frontiers | Research Topics

Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system

Edited by
Barbara M. P. Willekens, Beatrijs Wokke and Ilka Kleffner

Published in
Frontiers in Neurology
Frontiers in Immunology

ISSN 1664-8714
ISBN 978-2-8325-4212-5
DOI 10.3389/978-2-8325-4212-5

About Frontiers

Frontiers is more than just an open access publisher of scholarly articles: it is a pioneering approach to the world of academia, radically improving the way scholarly research is managed. The grand vision of Frontiers is a world where all people have an equal opportunity to seek, share and generate knowledge. Frontiers provides immediate and permanent online open access to all its publications, but this alone is not enough to realize our grand goals.

Frontiers journal series

The Frontiers journal series is a multi-tier and interdisciplinary set of open-access, online journals, promising a paradigm shift from the current review, selection and dissemination processes in academic publishing. All Frontiers journals are driven by researchers for researchers; therefore, they constitute a service to the scholarly community. At the same time, the *Frontiers journal series* operates on a revolutionary invention, the tiered publishing system, initially addressing specific communities of scholars, and gradually climbing up to broader public understanding, thus serving the interests of the lay society, too.

Dedication to quality

Each Frontiers article is a landmark of the highest quality, thanks to genuinely collaborative interactions between authors and review editors, who include some of the world's best academicians. Research must be certified by peers before entering a stream of knowledge that may eventually reach the public - and shape society; therefore, Frontiers only applies the most rigorous and unbiased reviews. Frontiers revolutionizes research publishing by freely delivering the most outstanding research, evaluated with no bias from both the academic and social point of view. By applying the most advanced information technologies, Frontiers is catapulting scholarly publishing into a new generation.

What are Frontiers Research Topics?

Frontiers Research Topics are very popular trademarks of the *Frontiers journals series*: they are collections of at least ten articles, all centered on a particular subject. With their unique mix of varied contributions from Original Research to Review Articles, Frontiers Research Topics unify the most influential researchers, the latest key findings and historical advances in a hot research area.

Find out more on how to host your own Frontiers Research Topic or contribute to one as an author by contacting the Frontiers editorial office: frontiersin.org/about/contact

Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system

Topic editors

Barbara M. P. Willekens — Antwerp University Hospital, Belgium

Beatrijs Wokke — Erasmus Medical Center, Netherlands

Ilka Kleffner — University Hospital Bochum GmbH, Germany

Citation

Willekens, B. M. P., Wokke, B., Kleffner, I., eds. (2024). *Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system*. Lausanne: Frontiers Media SA. doi: 10.3389/978-2-8325-4212-5

Table of contents

frontiers | Frontiers in Neurology

TYPE Editorial
PUBLISHED 13 December 2023
DOI 10.3389/fneur.2023.1342817

OPEN ACCESS

EDITED AND REVIEWED BY
Ronny Wickstrom,
Karolinska Institutet (KI), Sweden

*CORRESPONDENCE
Barbara M. P. Willekens
barbara.willekens@uza.be

RECEIVED 22 November 2023
ACCEPTED 28 November 2023
PUBLISHED 13 December 2023

CITATION
Willekens BMP, Kleffner I and Wokke B (2023) Editorial: Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system. *Front. Neurol.* 14:1342817. doi: 10.3389/fneur.2023.1342817

Editorial: Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system

Barbara M. P. Willekens[1,2,3]*, Ilka Kleffner[4] and Beatrijs Wokke[5]

[1]Department of Neurology, Antwerp University Hospital, Antwerp, Belgium, [2]Department of Translational Neurosciences, Faculty of Medicine and Health Sciences, University of Antwerp, Antwerp, Belgium, [3]Laboratory of Experimental Hematology, Faculty of Medicine and Health Sciences, Vaccine and Infectious Disease Institute, University of Antwerp, Antwerp, Belgium, [4]Department of Neurology, University Hospital Knappschaftskrankenhaus, Ruhr University Bochum, Bochum, Germany, [5]Department of Neurology, Erasmus Medical Center, Rotterdam, Netherlands

KEYWORDS

neuromyelitis optica spectrum disorder (NMOSD), MOGAD, rare disease, neurosarcoidosis, GFAP, systemic disorders, autoimmune encephalitis (AIE)

Editorial on the Research Topic
Epidemiology, diagnosis, prognosis and treatment of rare immune-mediated diseases of the central nervous system

Introduction

Rare immune-mediated disorders of the central nervous system (CNS) continue to pose challenges in diagnosis, prognosis and treatment, stressing the importance of sharing knowledge in the research community. In this Research Topic, we aimed to bring together clinical and case studies, epidemiological studies and reviews covering a variety of rare CNS immune-mediated disorders, including autoimmune encephalitis (AIE), neuromyelitis optica spectrum disorders (NMOSD), MOG antibody associated disease (MOGAD), Glial Fibrillary Astrocytic Protein (GFAP) autoimmune astrocytopathy and neurological involvement in systemic disorders such as lupus, rheumatoid arthritis, sarcoidosis and Sjögren's disease.

Autoimmune encephalitis: clinical findings and prognostication

In this Research Topic, several case reports (Ding C. et al., Khojah et al., Li et al.) address novel findings in autoimmune encephalitis (AIE), including a familiar case of LGI-1 AIE presented by Ding C. et al., suggesting a genetic background and advocating for Genome Wide Association Studies to discover the presence of risk alleles. Li et al. report a case of a patient with anti-GAD65 AIE following HPV vaccination, considering this temporal relationship as a trigger for development of AIE. Finally Khojah et al. have performed a systematic review, including a case vignette on mGluR-1 AIE, stressing the importance of the association of this antibody with cerebellar encephalitis and normal brain imaging in half of patients.

Two manuscripts concerned prognostication. Wu et al. analyzed four models to predict intensive care unit (ICU) admission of patients with AIE in a cohort of 234 patients of whom 40 were admitted to the ICU. The clinical assessment scale in autoimmune encephalitis (CASE) scale plus model, including prodromal symptoms, elevated fasting blood glucose and elevated cerebrospinal fluid (CSF) white blood cell (WBC) count, was selected as the best predictive model. The findings of this model should be externally validated. Ding J. et al. studied 34 patients with anti-gamma-aminobutyric-acid type B receptor (anti-$GABA_BR$) encephalitis and found that pulmonary infection and baseline mRS scores were independent risk factors for poor prognosis after a firstline immunotherapy. Finally, Bai et al. report the clinical spectrum, response to immunotherapy and outcomes of patients ($n = 55$) with GAD65 antibodies. The most frequent clinical syndromes were limbic encephalitis ($n = 34$, 61.82%), stiff-person syndrome (SPS; $n = 18$, 32.73%), cerebellar ataxia ($n = 11$, 20%) or overlap syndromes. Almost 60% of patients had other autoimmune conditions, including Hashimoto thyroiditis, type 1 diabetes mellitus and vitiligo. A minority ($n = 2$, 3.64%) of patients had underlying tumors, including thymoma and small cell lung carcinoma. Most patients had short-term favorable outcomes with Modified Ranking Scale ≤ 2 (87%). Longterm outcomes showed more variation and were dependent on the clinical phenotype.

Clinical presentation, prognostication and management of NMOSD, MOGAD and GFAP autoimmune astrocytopathy

GFAP antibodies were first described in 2016 (1) as a biomarker of relapsing meningoencephalomyelitis. Over the years, the clinical spectrum has extended. Zhu et al. report on 59 adults and children with GFAP antibodies in serum or CSF of whom 55 were positive only in the CSF. Interestingly, in almost a quarter of them multiple autoantibodies were detected, most frequently AQP4 antibodies. The most common phenotype in children was encephalomyelitis (9/18, 50%) and in adults encephalitis (15/41, 36.6%). More than 80% had a monophasic course over a median followup time of 9 months. Zhang, Xie et al. performed a similar retrospective analysis of 33 patients, with a slightly longer median followup time of 12 months, reporting relapses while steroids were tapered in four patients. Almost 80% had good outcomes in the short-term. A study by Sun et al. compared clinical and imaging features of GFAP and MOG antibody associated myelitis in 14 and 24 patients respectively, in order to differentiate these disorders. Higher protein CFS levels were found in GFAP vs. MOG antibody positive patients, which may help clinicians differentiate these diseases.

While many patients with NMOSD have a good response to rituximab, some may be none-responders. Zhang, Jiao et al. report a difficult to treat NMOSD case and present a successful treatment approach with ofatumumab and IVIg.

The intriguing observation and role of enlarged perivascular spaces in NMOSD is discussed by Yao et al. while a temporal association of NMOSD with SARS-CoV-2 vaccination is discussed in a systematic review by Harel et al.. The often difficult patient journey from diagnosis to chronic disease is well-described by Delgado-Garcia et al..

Neurological involvement in systemic disorders

Finally, some interesting cohorts are presented, discussing the clinical presentation, diagnostic approach and management of neurosarcoidosis (Sambon et al.), rheumatoid meningitis (Fan et al.), and Sjögren's syndrome (Hoshina et al.). A cohort of patients with MRI negative myelitis, show that this can be a presenting feature of lupus (Das et al.).

Concluding remarks

Overall, this Research Topic includes recent and emerging insights on clinical aspects of rare CNS immune-mediated disorders.

Author contributions

BMPW: Conceptualization, Writing—original draft, Writing—review & editing. IK: Writing—review & editing. BW: Writing—review & editing.

Funding

The author(s) declare that no financial support was received for the research, authorship, and/or publication of this article.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Fang B, McKeon A, Hinson SR, Kryzer TJ, Pittock SJ, Aksamit AJ, et al. Autoimmune glial fibrillary acidic protein astrocytopathy: a novel meningoencephalomyelitis. *JAMA Neurol.* (2016) 73:1297–307. doi: 10.1001/jamaneurol.2016.2549

frontiers | Frontiers in Neurology

CASE REPORT
published: 14 April 2022
doi: 10.3389/fneur.2022.855383

The First Case of Familiar Anti-leucine-rich Glioma-Inactivated1 Autoimmune Encephalitis: A Case Report and Literature Review

Chuhan Ding†, Qibing Sun†, Ran Li†, Hanli Li† and Yu Wang†*

Department of Neurology, The First Affiliated Hospital of Anhui Medical University, Hefei, China

Anti-leucine-rich glioma-inactivated1 (Anti-LGI1) autoimmune encephalitis is a rare autoimmune disease discovered in recent years. It is generally not defined as an inherited disease, though its etiology is still unclear. Herein, we report the first case of adult patients with familial anti-LGI1 encephalitis. Two biological siblings who worked in different regions were successively diagnosed with anti-LGI1 encephalitis in their middle age. The two patients had similar clinical manifestations including imaging results. Their clinical symptoms improved after immunotherapy and antiepileptic therapy. Given that some unique human leukocyte antigen (HLA) subtypes appear at a high frequency, multiple recent studies have revealed that anti-LGI1 encephalitis is associated with genetic susceptibility. One of the patients underwent HLA genotyping and whole-exome sequencing (WES), revealing the same HLA typing as in previous studies and two rare HLA variants. Therefore, further studies involving larger samples and more populations should be conducted to explore the possibility of other influencing factors such as environmental impacts.

Keywords: Anti-leucine-rich glioma-inactivated1 autoimmune encephalitis, autoimmune encephalitis, LGI1, HLA, case report

OPEN ACCESS

Edited by:
Barbara M. P. Willekens, Antwerp University Hospital, Belgium

Reviewed by:
Erdem Tüzün, Istanbul University, Turkey
Rinze Neuteboom, Erasmus Medical Center, Netherlands

***Correspondence:**
Yu Wang
wangyu18b@163.com

†These authors have contributed equally to this work and share first authorship

Specialty section:
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

Received: *15 January 2022*
Accepted: *14 March 2022*
Published: *14 April 2022*

Citation:
Ding C, Sun Q, Li R, Li H and Wang Y (2022) The First Case of Familiar Anti-leucine-rich Glioma-Inactivated1 Autoimmune Encephalitis: A Case Report and Literature Review. Front. Neurol. 13:855383. doi: 10.3389/fneur.2022.855383

INTRODUCTION

Anti-LGI1 encephalitis, a rare autoimmune encephalitis defined in recent years, is characterized by seizures, cognitive impairment, psychiatric disorders, and refractory hyponatremia. LGI1 is a type of neuron-secreted protein which dominantly expresses in the hippocampus and temporal cortex and transmits signals from the presynaptic to the posterior membrane (1, 2). Autoimmune encephalitis is a clinical syndrome of which the diagnosis is based on the detection of accurate antibodies, though it is not fully recognized. Currently, the first-line therapy for autoimmune encephalitis includes immunoglobulin, glucocorticoid, and plasma exchange, and the second-line therapy includes rituximab, cyclophosphamide, and mycophenolate mofetil (3). This report describes the first pair of siblings who were diagnosed with anti-LGI1 encephalitis after experiencing a convulsive seizure. According to previous gene association studies (3), we conducted genetic tests on the younger brother and found that his unique HLA haplotype was consistent with these studies, in addition to identifying two HLA variants. Therefore, more comprehensive genetic studies in a larger population are warranted.

PATIENT 1

A 39-year-old man was admitted to our department in July 2021 because of one-month history of short-term memory loss and a generalized tonic-clonic seizure (GTCS) attacking during sleep. The patient had no personal history of hypertension, diabetes, or other diseases and had no alcohol consumption but smoking for 10 years.

Neurological examination revealed normal except the spatial and temporal disorientation and memory impairment, especially the short-term memory impairment. The patient was able to recall three items immediately but, afterward, unable to recall any one. His Mini-mental State Examination (MMSE) was scored 19/30, and Montreal Cognitive Assessment (MoCA) was scored 12/30.

In detail, he had mild impairment in naming, severe destruction in visuospatial abilities, executive functioning, sentence repetition, abstract thinking, orientation, and delayed recall.

The brain magnetic resonance image (MRI) conducted on the admission day presented hyperintensities on T2 weighted image (T2WI) and fluid-attenuated inversion recovery (FLAIR) image but hypointensities on T1 weighted image (T1WI) in the bilateral temporal lobes and hippocampus with dominance in the left side (**Figure 1A**). Thoracic computed tomography (CT) revealed normal. The serum sodium concentration was 132.4 mmol/L which was lower than the normal (reference range: 137.0–147.0 mmol/L). The white blood cell count and C-reactive protein (CRP) were slightly higher than normal. The electroencephalogram (EEG) was detected with generalized slow wave (delta) activities. Cerebrospinal fluid (CSF) examination showed a slightly increased in the count of leukocytosis (7×10^6/L) but normal in the levels of chloride, glucose, and protein. Autoimmune encephalitis antibodies were detected with positive LGI1 antibody in CSF (1:100+) and serum (1:1000+). The clinical information of the patient is indicated in **Table 1**.

The patient was treated with intravenous immunoglobulin (IVIG) at a dose of 0.4 g/kg/day (25 g in total) for five days, followed by methylprednisolone pulse therapy (500 mg/day for five days, 250 mg/day for three days, 120 mg/day for three days). After treatment, his cognition improvement was remarkable from daily performance as he could realize he was a patient instead of mistaking himself as a caregiver for his wife, though no significant improvement in MMSE or MoCA scores after hospitalized treatment. He was discharged with oral prednisone and sustained-release sodium valproate tablets for seizure control. In a follow-up of three months after treatment, brain MRI imaging showed that the brain lesion improved (**Figure 1B**). The anti-LGI1 titer in serum decreased to 1:100+. MoCA score ameliorated to 15/30 for the noticeable improvement observed in visuospatial abilities, executive functioning, and retelling abilities. Nevertheless, severe impairment of orientation persisted.

PATIENT 2

In 2017, a 36-year-old woman, the biological sister of patient 1, was admitted to another hospital due to paroxysmal full-body numbness, "paroxysmal twitch" in upper limbs, blurred vision,

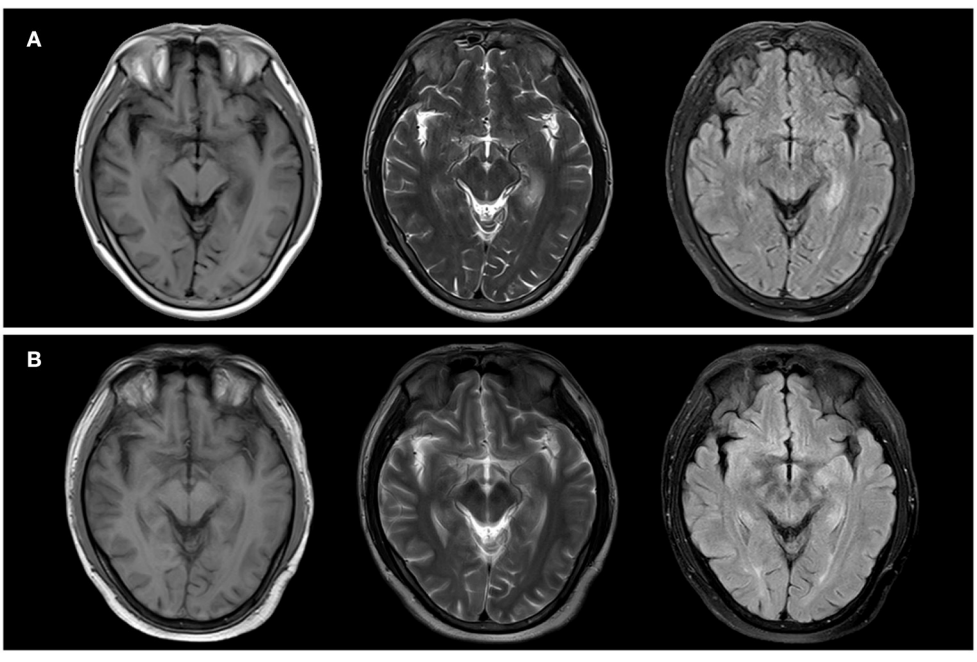


FIGURE 1 | Neuroradiologic MRI (1.5 T) of Patient 1. The bilateral temporal lobes and bilateral hippocampus showed hypointensity on T1WI and hyperintensity on T2WI and FLAIR. Notably, they were more pronounced in the left side **(A)**. After two months, the bilateral temporal lobes and bilateral hippocampus showed a slightly lower signal on T1WI and a slightly higher signal on T2 and FLAIR. Notably, they were more pronounced in the left side **(B)**.

TABLE 1 | Clinical profiles of the two patients.

	Test	Patient 1	Patient 2
Characteristics	Gender	male	female
	Age at onset	39	36
Clinical Symptom	Central nervous system	Seizure, memory decline	Seizure, memory decline, disorder of behavior, hallucinations, blurred vision
	Seizure types	Generalized tonic-clonic seizures	Simple partial seizures
	Peripheral nervous system	no	Neuropathic pain, muscle weakness and numbness
	Autonomic nervous system	tachycardia	Hyperhidrosis
Laboratory Studies	CBC	WBC 10.5×10^{9}/L, NUET 7.37×10^{9}/L	WBC 9.80×10^{9}/L
	Serum sodium	133.2 mmol/L	129 mmol/L
	Neoplasm	No	No effective basis for neoplasm
	Serological tumor markers	WNL	CA7-24: 8.72 U/mL
	Liver and kidney function tests	WNL	WNL
Brain Imaging	EEG	a delta (2-3c/s)activity	Extensive diffuse slow waves
	Initial MRI	Hyperintensities in bilateral parahippocampus	Hyperintensities in left temporal lobe
	Follow-up MRI	Slightly hiper signals on bilateral parahippocampus	None
LGI1-IgG	Serum (Cell-based assays, diluted 1:10)	1:1000+	1:100+
	CSF (Cell-based assays, without diluted)	1:100+	1:10+
Cerebrospinal Fluid Studies	Pressure	110 mm H_2O	82 mm H_2O
	Nucleated Cell Count	7×10^{6}/L	1×10^{6}/L
	Glucose	Normal	5.00 mmol/L
	Chloride	Normal	Normal
	Protein	Normal	Normal
	Microbiological and virological test	Normal	Normal
Other Auxiliary Examination	Chest CT	Normal	Normal
	Echocardiography	Sinus tachycardia	Normal
Treatment		Immunotherapy (IVIG and corticosteroids)	Immunotherapy (corticosteroids)
outcome		Returned to work	Returned to work

autonomic dysfunction, memory loss, confusion, and visual hallucinations. Notably, her clinical presentation first appeared two months before admission. However, after consulting in several hospitals, she was misdiagnosed with dysautonomia or peripheral neuropathy.

The medical record indicated she had depressed mood, poor orientation, and poor memory. The physical examination revealed mild weakness of extremities (muscle strength grade 5-/5), walking slowness, and unsteady gait.

Her blood routine test, blood biochemistry test, and anti-nuclear antibodies were all within physiological ranges. With regard to tumor markers, carbohydrate antigen 724 was mildly increased to 8.72 U/mL (reference range: 0–8.20 U/mL), but no definite tumor was detected. Brain MRI plain scan and enhancement revealed anomalous signals in the left temporal lobe and hippocampus (**Figure 2**). In addition, EEG showed generalized extensive diffuse slow waves.

CSF examination indicated WBC count and protein level were normal, chloride slightly increased and glucose slightly increased. Autoimmune encephalitis antibodies test ultimately indicated that anti-LGI1 was positive in CSF (1:10+) and serum (1:100+). She was forthwith treated with sodium valproate tablets and methylprednisolone pulse therapy for 20 days with dosage decreasing and discharged with oral prednisolone. She had difficulties in follow-up due to busy workloads after discharge. Long-term administration of prednisolone made her look swollen, thus she eventually stopped taking medication after three years' oral hormone. She is still plagued by memory

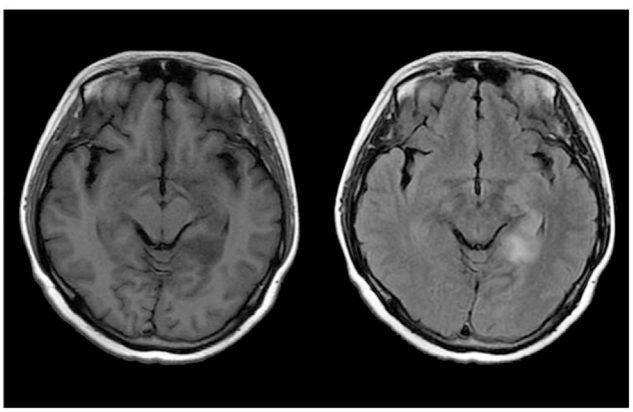


FIGURE 2 | Neuroradiologic MRI (3.0 T) of Patient 2. The left temporal lobe and hippocampus showed hypointensity on T1WI and hyperintensity on FLAIR.

TABLE 2 | Result of Patient 2's HLA genotyping.

Alleles/haplotypes	HLA-A	HLA-B	HLA-C	HLA-DRB1	HLA-DQB1	HLA-DQA1	HLA-DRB4	HLA-DPB1
	02:03,33:03	38:02,57:01*	06:02,07:02	07:01*,13:12	03:01,03:03*	02:01*,05:03	01:03*,/	05:01,13:01

*Special HLA genotypes that appeared in the literature studies on genetic susceptibility for anti-LGI1 encephalitis.

impairment while we followed her up, but she refused further treatment, examination, and further follow-up visits. This made it difficult to get her WES and other laboratory examination. She remained mild cognitive impairment with no disturbance of daily life as informed in a recent phone follow-up.

DISCUSSION

To date, almost all reported cases of autoimmune encephalitis were sporadic cases except for one familial autoimmune encephalitis reported in two pediatric brothers affected with voltage-gated potassium channel (VGKC) encephalitis (4).

Here, we report the first case of familiar LGI1 autoimmune encephalitis in adult patients. The clinical profiles are shown in **Table 1**. Specifically, two adult siblings at similar age were successively diagnosed with anti-LGI1 encephalitis within four years. Although both patients presented with subacute onset, and developed seizures and memory decline, some differences existed between the two siblings in brain images and EEG presentations as well as in clinical manifestations. The sister exhibited more severe and earlier autonomic dysfunction which had misled the physicians to diagnose it as peripheral neuropathy or autonomic dysfunction. Tests for autoimmune antibodies were undergone after her seizure attack, and this made the diagnosis of anti-LGI1 encephalitis established.

Recently, it was reported that adolescent siblings with acquired autoimmune syndrome after mercury exposure, and several autoantibodies including anti-LGI1 were detected (5). However, these two siblings we reported excluded toxicant exposure. As they had lived in distinct environments for more than 20 years before symptom onset, the living environment might not be a critically potential pathogenic factor.

Due to the consanguinity between the two patients, it should be taken into consideration that anti-LGI1 encephalitis is associated with genetic susceptibility. Multiple recent studies have indicated an association of anti-LGI1 encephalitis with HLA (6–11), but familial cases supporting this genetic association had never been reported. On the other hand, this definite and consistent genetic susceptibility has not been found in other types of autoimmune encephalitis. HLA genes are closely linked and obey Mendelian law of inheritance. Therefore, there is a 25% chance of two siblings being identical in the HLA genotype, a 50% chance of sharing the same HLA haplotype, and a 25% chance of not having the same HLA haplotype. It is well-known that HLA genes encode antigen-presenting proteins on the cell surface participating in the immune response directly. Studies have revealed genetic associations exist between HLA and various autoimmune diseases except for autoimmune encephalitis, among which HLA class II genes exert their effectiveness through autoantibody production. For instance, HLA haplotype DR3-DQ2 and DR4-DQ8 increase the risk of celiac disease (12), HLA-DR is associated with systemic lupus erythematosus and lupus nephritis (13), and HLA-DRB1*10:01-DQB1*05:01 is associated with IgLON5 encephalopathy (14). HLA class I genes are mostly expressed in diseases that do not produce antibodies, with the most famous example of the strong correlation between ankylosing spondylitis and HLA-B27 (15). Considering the fact that anti-LGI1 encephalitis is a disease caused by antibodies, it can be inferred that the disease is more closely associated with HLAII genes.

HLA class IIplays an important role in other humoral autoimmunity as well. Type 1 Diabetes (T1D) is characterized by destruction of islet β-cells. Insulin autoantibodies (IAA) as corresponding autoantibodies appear in children with DR4–DQ8 haplotype, which is located on HLA class II and can influence both etiology and pathogenesis of T1D (16). The HLA complex accounted for about 50% of genetic risk of T1D (17), and the risk of progression is conferred by specific HLA-DR/DQ alleles, while some haplotypes (i.e., DR2) could be protective factors (18). T lymphocyte differentiation, characterized by HLA, is identified as an independent pathway involved in systemic lupus erythematosus (SLE) susceptibility. Genes in the HLA region dominated associated genes in the T cell differentiation and antigen processing and presentation pathways, which was confirmed by gene-based association testing (19).

To date, HLA genotyping as well as genome-wide association study (GWAS) have been performed among patients with anti-LGI1 encephalitis across multiple populations, including Caucasian population (7–10), South Korean population (6), and southwestern Han Chinese population (11), demonstrating a significant association between unique HLA subtypes and anti-LGI1 encephalitis. In parallel, the frequencies of some definite sites located on major histocompatibility complex (MHC) class II were significantly higher in the anti-LGI1 encephalitis group than in the healthy control or epilepsy groups. Therefore, it is speculated that HLA isotypes might activate the immune response or work through initiating T–B cell interactions during disease onset (8). However, it should be noted that HLA subtypes in published studies are not consistent, possibly due to different ethnicities.

The first research on genetic susceptibility for anti-LGI1 encephalitis was conducted in the Netherlands (7). Interestingly, researchers explored the relationship between HLA and anti-LGI1 patients with or without tumors. They found a strong correlation of non-tumor anti-LGI1 encephalitis with HLA-DR7 and HLA-DRB4, as significant as the correlation between HLA-B27 and patients with ankylosing spondylitis. It suggested that the deficiency of HLA-DR7 or DRB4 appeared to boost

the prevalence of a tumor. Consequently, the researchers recommended an intensive tumor screen and long-term follow-up in anti-LGI1 encephalitis without HLA-DR7 or DRB4.

Conversely, a British research (8) revealed the uncorrelation between HLA and tumor in anti-LGI1 encephalitis patients. Hence, more studies in HLA and tumor in these patients are urgent to guide the clinical diagnosis and treatment. Moreover, this study found that the HLA class I and IIvariants (HLA-DRB1*07:01, HLA-DQA1*02:01, HLA-B*57:01) may increase the risk of adverse drug reactions (20).

The first study implementing genome-wide association (GWAS) analysis was conducted in Germany (9). The unprecedented discovery was that anti-LGI1 encephalitis was highly associated with 27 single-nucleotide polymorphisms (SNPs) located in the HLA class II region between HLA-DRB1 and HLA-DQA1 (leading SNP rs2858870) in the region of MHCIIgenes. It even found that DRB1*07:01 and DQA1*02:01 always appeared together in all participants. Considering that related alleles were associated with decreased total serum IgG levels, and LGI1 autoantibodies mainly belong to the IgG4 subclass (21), the result suggested that these haplotypes, in addition to improving peptide presentation of LGI1 peptides, may be associated with the disorder of the IgG4-LGI1, which is potentially pathogenic. In addition, a study in South Korea (6) discovered a higher frequency of B*44:03 and C*07:06 alleles of the HLA class I in anti-LGI1 encephalitis patients, illustrating that HLA class I is possible pathogenesis of anti-LGI1 encephalitis as well.

HLA genotyping was performed in anti-LGI1 encephalitis cohort in France recently (10) and found that 88% of patients carried DRB1*07:01. The study newly identified that non-carriers were younger, more frequently women, and presented less frequently with psychiatric and frontal symptoms, whereas non-carriers were not associated with poor outcomes. The HLA association in paraneoplastic or oncological patients has not been confirmed. The mechanisms of sex and age bias in HLA class II-associated diseases are unclear, while a study presumed that estrogens may change HLA expression (22).

The only study in the Chinese Han population was conducted in China (11), however, the study showed no evidence that the DRB1*07:01 ∼ DQB1*02:02 haplotype was associated with this disease. Researchers attributed this inconsistency to ethnic differences. All of the results of these researches mentioned above possessed homogeneity and heterogeneity, seen in **Table S1**.

WES is a promising tool in genetic testing methods, which offers the possibility of identifying rare or novel alleles responsible for the disease. In 2014, a child with cerebral lupus was identified a homozygous mutation in the Three Prime Repair Exonuclease 1 (TREX1) by this method (23). Some studies found possible mechanisms as $TREX1^{R97H}$ mutant protein had a severe reduction in exonuclease activity that leads to defects in clearance of nucleic acids, and triggers signaling pathways that promote secretion of type I IFNs and inflammation.

Herein, we performed HLA typing and WES on Patient 1. His HLA typing was DRB1*07:01, DQA1*02:01, and DQB1*03:03 (**Table 2**), which perfectly matched the H3 haplotype reported by the previous study from Germany. HLA-B*57:01 was also detected, which has been shown to perhaps induce adverse drug reactions (20). It should be noted that these alleles were recurrent in studies based on multiple populations, which indicated a highly possible association with the clinical onset of anti-LGI1 encephalitis. Given that this disease mainly occurs in middle-aged and elderly individuals, the pathogenesis may also be associated with environmental effects. According to the result of WES, we found one homozygous HLA-DRB1 variant (NM_002124:exon2:c.101-1G>A) and one heterozygous HLA-DPA1 variant (NM_001242524:exon5:c.746G>A:p.R249H), and details are given in **Table S2**. These variants were absent or rare in the general Chinese population. Regrettably, these mutations have not been verified in his sister and pedigree, and could not be clarified the exact role in the disease process. Apart from HLA variants, investigation on WES did not reveal any disease-causing variants associated with anti-neuronal autoimmune encephalitis. This unremarkable result was possibly associated with undetected problems in gene expression or epigenetics. In addition, tumor screening of two patients was performed by serum tumor markers and chest CT, not in the whole body, which was a limitation. Hence, there is an urgent need to verify these speculations by expanding the samples and performing further genome-wide association analysis.

In summary, the latest studies above have confirmed that anti-LGI1 encephalitis is genetically susceptible, highly associated with specific alleles located on HLA class II. However, more researches with large samples and more races are necessary to verify it. In addition, the frequencies of these HLA variants were much higher than the prevalence of anti-LGI1 encephalitis, suggesting that people with unique alleles may not develop the disease. Therefore, further studies should focus on the possibility of other influencing factors of anti-LGI1 encephalitis, for instance, additional haplotypes, environmental impacts, and other random effects.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**, further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the First Affiliated Hospital of Anhui Medical University. The patients/participants provided their written informed consent to participate in this study. Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

CD explained the data and wrote the manuscript. QS, RL, and HL acquired and analyzed data. CD and YW revised the manuscript. All authors approved the final manuscript.

FUNDING

This study was financially supported by the National Natural Science Foundation of China (YW, Grant No. 82071460).

ACKNOWLEDGMENTS

We would like to thank the patients for agreeing and providing their case histories. We acknowledge Dr. Xingui Chen (Department of Neurology) for diagnosing and taking care of the patient, Dr. Tianjuan Wang, MD (Department of Obstetrics and Gynecology) and Dr. Liqiong Cai, MD (Basic Medical Sciences) for helpful advice in genetics.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.855383/full#supplementary-material

REFERENCES

1. Irani SR, Alexander S, Waters P, Kleopa KA, Pettingill P, Zuliani L, et al. Antibodies to Kv1 potassium channel-complex proteins leucine-rich, glioma inactivated 1 protein and contactin-associated protein-2 in limbic encephalitis, Morvan's syndrome and acquired neuromyotonia. *Brain.* (2010) 133:2734–48. doi: 10.1093/brain/awq213
2. Ohkawa T, Fukata Y, Yamasaki M MT, Yokoi N, Takashima H, Watanabe M, et al. Autoantibodies to epilepsy-related LGI1 in limbic encephalitis neutralize LGI1-ADAM22 interaction and reduce synaptic AMPA receptors. *J Neurosci.* (2013) 33:18161–74. doi: 10.1523/JNEUROSCI.3506-13.2013
3. Zuliani L, Nosadini M, Gastaldi M, Spatola M, Iorio R, Zoccarato M, et al. Management of antibody-mediated autoimmune encephalitis in adults and children: literature review and consensus-based practical recommendations. *Neurol Sci.* (2019) 40:2017–30. doi: 10.1007/s10072-019-03930-3
4. Gillespie LE, Dave A, Goldstein A, A. Tale of two brothers: familial voltage-gated potassium channel autoimmune encephalitis. *Cureus.* (2020) 12:e8723. doi: 10.7759/cureus.8723
5. Pérez CA, Shah EG, Butler IJ. Mercury-induced autoimmunity: report of two adolescent siblings with Morvan syndrome "plus" and review of the literature. *J Neuroimmunol.* (2020) 342:577197. doi: 10.1016/j.jneuroim.2020.577197
6. Kim TJ, Lee ST, Moon J, Sunwoo JS, Byun JI, Lim JA, et al. Anti-LGI1 encephalitis is associated with unique HLA subtypes. *Ann Neurol.* (2017) 81:183–92. doi: 10.1002/ana.24860
7. van Sonderen A, Roelen DL, Stoop JA, Verdijk RM, Haasnoot GW, Thijs RD, et al. Anti-LGI1 encephalitis is strongly associated with HLA-DR7 and HLA-DRB4. *Ann Neurol.* (2017) 81:193–8. doi: 10.1002/ana.24858
8. Binks S, Varley J, Lee W, Makuch M, Elliott K, Gelfand JM, et al. Distinct HLA associations of LGI1 and CASPR2-antibody diseases. *Brain.* (2018) 141:2263–71. doi: 10.1093/brain/awy109
9. Mueller SH, Färber A, Prüss H, Melzer N, Golombeck KS, Kümpfel T, et al. Genetic predisposition in anti-LGI1 and anti-NMDA receptor encephalitis. *Ann Neurol.* (2018) 83:863–9. doi: 10.1002/ana.25216
10. Muñiz-Castrillo S, Haesebaert J, Thomas L, Vogrig A, Pinto AL, Picard G, et al. Clinical and prognostic value of immunogenetic characteristics in anti-LGI1 encephalitis. *Neurol Neuroimmunol Neuroinflamm.* (2021) 8:e974. doi: 10.1212/NXI.0000000000000974
11. Hu F, Liu X, Zhang L, Chen C, Gong X, Lin J, et al. Novel findings of HLA association with anti-LGI1 encephalitis: HLA-DRB1*03:01 and HLA-DQB1*02:01. *J Neuroimmunology.* (2020) 344:577243. doi: 10.1016/j.jneuroim.2020.577243
12. Liu E, Lee HS, Aronsson CA, Hagopian WA, Koletzko S, Rewers MJ, et al. Risk of pediatric celiac disease according to HLA haplotype and country. *N Engl J Med.* (2014) 371:42–9. doi: 10.1056/NEJMoa1313977
13. Niu Z, Zhang P, Tong Y. Value of HLA-DR genotype in systemic lupus erythematosus and lupus nephritis: a meta-analysis. *Int J Rheum Dis.* (2015) 18:17–28. doi: 10.1111/1756-185X.12528
14. Gaig C, Ercilla G, Daura X, Ezquerra M, Fernández-Santiago R, Palou E, et al. HLA and microtubule-associated protein tau H1 haplotype associations in anti-IgLON5 disease. *Neurol Neuroimmunol Neuroinflamm.* (2019) 6:e605. doi: 10.1212/NXI.0000000000000605
15. Sollid LM, Pos W, Wucherpfennig KW. Molecular mechanisms for contribution of MHC molecules to autoimmune diseases. *Curr Opin Immunol.* (2014) 31:24–30. doi: 10.1016/j.coi.2014.08.005
16. Krischer JP, Lynch KF, Lernmark K, Hagopian WA, Rewers MJ, She JX, et al. Genetic and environmental interactions modify the risk of diabetes-related autoimmunity by 6 years of age: the TEDDY study. *Diabetes Care.* (2017) 40:1194–202. doi: 10.2337/dc17-0238
17. Cerolsaletti K, Hao W, Greenbaum CJ. Genetics coming of age in type 1 diabetes. *Diabetes Care.* (2019) 42:189–91. doi: 10.2337/dci18-0039
18. Noble JA, Valdes AM, Cook M, Klitz W, Erlich HA. The role of HLA class II genes in insulin-dependent diabetes mellitus: molecular analysis of 180 Caucasian, multiplex families. *Am J Hum Genet.* (1996) 59:1134–48.
19. Sandling JK, Pucholt P, Rosenberg LH, Farias F, Rnnblom L. Molecular pathways in patients with systemic lupus erythematosus revealed by gene-centred DNA sequencing. *Ann Rheum Dis.* (2021) 80:109–17. doi: 10.1136/annrheumdis-2020-218636
20. Yip VLM, Alfirevic A, Pirmohamed M. Genetics of immune-mediated adverse drug reactions: a comprehensive and clinical review. *Clin Rev Allergy Immunol.* (2015) 48:165–75. doi: 10.1007/s12016-014-8418-y
21. Jonsson S, Sveinbjornsson G, de Lapuente Portilla AL, Swaminathan B, Plomp R, Dekkers G, et al. Identification of sequence variants influencing immunoglobulin levels. *Nat Genet.* (2017) 49:1182–91. doi: 10.1038/ng.3897
22. Taneja V. Sex hormones determine immune response. *Front Immunol.* (2018) 9:1931. doi: 10.3389/fimmu.2018.01931
23. Ellyard JI, Jerjen R, Martin JL, Lee A, Vinuesa CG. Whole exome sequencing in early-onset cerebral SLE identifies a pathogenic variant in TREX1. *Arthritis Rheumatol.* (2014) 66:3382–6. doi: 10.1002/art.38824

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Immunology

ORIGINAL RESEARCH
published: 10 June 2022
doi: 10.3389/fimmu.2022.916111

Risk Prediction Models for Early ICU Admission in Patients With Autoimmune Encephalitis: Integrating Scale-Based Assessments of the Disease Severity

Chunmei Wu, Yongkang Fang, Yingying Zhou, Huiting Wu, Shanshan Huang and Suiqiang Zhu**

Department of Neurology, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, China

OPEN ACCESS

Edited by:
Barbara M. P. Willekens, Antwerp University Hospital, Belgium

Reviewed by:
Jeroen Kerstens, Antwerp University Hospital, Belgium
Justin R. Abbatemarco, Cleveland Clinic, United States

***Correspondence:**
Shanshan Huang
shanahuang3@gmail.com
Suiqiang Zhu
zhusuiqiang@163.com

Specialty section:
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Immunology

Received: *08 April 2022*
Accepted: *16 May 2022*
Published: *10 June 2022*

Citation:
Wu C, Fang Y, Zhou Y, Wu H, Huang S and Zhu S (2022) Risk Prediction Models for Early ICU Admission in Patients With Autoimmune Encephalitis: Integrating Scale-Based Assessments of the Disease Severity. Front. Immunol. 13:916111. doi: 10.3389/fimmu.2022.916111

Background: In patients with autoimmune encephalitis (AE), the prediction of progression to a critically ill status is challenging but essential. However, there is currently no standard prediction model that comprehensively integrates the disease severity and other clinical features. The clinical assessment scale in autoimmune encephalitis (CASE) and the modified Rankin Scale (mRS) have both been applied for evaluating the severity of AE. Here, by combining the two scales and other clinical characteristics, we aimed to investigate risk factors and construct prediction models for early critical care needs of AE patients.

Methods: Definite and probable AE patients who were admitted to the neurology department of Tongji Hospital between 2013 and 2021 were consecutively enrolled. The CASE and mRS scores were used to evaluate the overall symptom severity at the time of hospital admission. Using logistic regression analysis, we analyzed the association between the total scores of the two scales and critical illness individually and then we evaluated this association in combination with other clinical features to predict early intensive care unit (ICU) admission. Finally, we constructed four prediction models and compared their performances.

Results: Of 234 patients enrolled, forty developed critical illness and were early admitted to the ICU (within 14 days of hospitalization). Four prediction models were generated; the models were named CASE, CASE-plus (CASE + prodromal symptoms + elevated fasting blood glucose + elevated cerebrospinal fluid (CSF) white blood cell (WBC) count), mRS and mRS-plus (mRS + prodromal symptoms + abnormal EEG results + elevated fasting blood glucose + elevated CSF WBC count) and had areas under the ROC curve of 0.850, 0.897, 0.695 and 0.833, respectively. All four models had good calibrations. In general, the models containing "CASE" performed better than those including "mRS", and the CASE-plus model demonstrated the best performance.

Conclusion: Overall, the symptom severity at hospital admission, as defined by CASE or mRS, could predict early ICU admission, especially when assessed by CASE. Adding other clinical findings, such as prodromal symptoms, an increased fasting blood glucose level and an increased CSF WBC count, could improve the predictive efficacy.

Keywords: autoimmune encephalitis, intensive care unit, risk factor, prediction, model

INTRODUCTION

The severity of autoimmune encephalitis (AE) is highly heterogeneous because it can range from mild impairments in working memory to the most severe, persistent disorders of consciousness that would require lasting care or could even cause death (1–3). During the acute stage of the disease, the rapid progression of an immune inflammatory response may cause severe neurological deficits, status epilepticus, coma, and respiratory failure (4). Moreover, with a high risk of suffering from multiple concurrent complications, such as lung infections and sepsis, the reported mortality of AE was as high as 40% in some studies (1, 4). Therefore, some patients require admission to intensive care units (ICUs) for the maximum standard of care. It is still unknown why some patients with AE survive the acute phase of the disease, while others are overwhelmed by the life-threatening acute phase. Previously, several variables, such as anemia, a definite diagnosis of AE (5), cerebrospinal fluid (CSF) WBC >20 cells/mm3 (6), failure of first-line immunotherapy (7) and a high CSF IL-17A concentration (8), were found to be associated with critical illness and subsequent ICU admission. However, to date, there is no standard prediction model that comprehensively includes both the clinical symptom severity and laboratory tests. For the subset of the AE patients who are critically ill, delayed admission to the ICU may be an independent risk factor for poor outcome (9). Therefore, identifying the risk factors for the deterioration to critical illness is crucial for early administration to the intensive care and for timely therapeutic implementation in order to improve prognosis.

Scales are ubiquitously used to assess the severity of symptoms in neurological diseases (10–12). Due to the lack of customized scales, the modified Rankin Scale (mRS) is usually applied for evaluating the neurological severity and outcomes in AE patients (2, 13, 14). However, mRS was originally designed to measure disability after stroke, and it was weighed toward motor deficits and functional independence and apparently with shortage in measuring the non-motor symptoms that frequently occur in AE (15). The clinical assessment scale in autoimmune encephalitis (CASE) is a novel tool that was developed in 2019 to specifically evaluate the clinical severity of a series of syndromes, including definite AE, definite autoimmune limbic encephalitis (ALE), autoantibody negative but probable AE, definite acute disseminated encephalomyelitis (ADEM), and definite and probable brainstem encephalitis (16). The CASE is composed of nine major clinical features of AE, with a total score ranging from 0 to 27 (16), and this makes it a fine quantification tool that has great potential in the assessment of AE. Two studies that evaluated Chinese patients with antibody-positive AE confirmed the accuracy of the clinical evaluation of CASE (17, 18). However, CASE has not been popularized in clinical practice, nor is there a study comparing the performance of the mRS and CASE in predicting ICU admission independently or in combination with other clinical factors.

Prolonged hospital length of stay may increase the risk of hospital-acquired infection (19), leading to an increased likelihood of ICU admission for reasons not directly related to AE. Therefore, to be representative of ICU admission for AE-related reasons, this study aimed to investigate the association between symptom severity at hospital admission, as assessed by the CASE or mRS score, and early deterioration, requiring ICU care, in patients diagnosed with definite and probable AE, and to construct scale-based risk prediction models and compare the performances of these models. Finally, we explored whether the addition of other clinical factors to the models that evaluated symptom severity could improve their predictive efficacy.

MATERIALS AND METHODS

Patients

We retrospectively extracted data from the medical records of consecutive patients diagnosed with encephalitis who were treated from January 2013 to October 2021 in the Department of Neurology, Tongji Hospital, and we screened those patients who met the clinical diagnostic criteria for definite and probable AE proposed by Mittal and Graus et al. in 2016 (3). Specifically, the patients with definite AE, definite ALE and autoantibody-negative but probable AE were included in this study. The detailed diagnostic criteria for each AE are described in the **Supplementary Table 1**. Patients were excluded when 1) they had infectious encephalitis with laboratory evidence, including tuberculosis or bacterial, fungal, viral (IgM), or parasitic

Abbreviations: ADEM, acute disseminated encephalomyelitis; AE, autoimmune encephalitis; AIC, Akaike's information criterion; ALE, autoimmune limbic encephalitis; AMPA, α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; ASMs, anti-seizure medications; AUC, area under the receiver operating characteristic (ROC) curve; CASE, the clinical assessment scale in autoimmune encephalitis; CASPR, contactin associated protein; CSF, cerebrospinal fluid; D_2R, Dopamine 2 receptor; DPPX, dipeptidyl-peptidase-like protein; EEG, electroencephalography; GABA, gamma-aminobutyric acid; GAD, glutamic acid decarboxylase; GBS, Guillain–Barre syndrome; ICU, intensive care unit; LGI1, leucine-rich glioma inactivated 1; MG, myasthenia gravis; mGluR, metabotropic glutamate receptor; MOG, myelin oligodendrocyte glycoprotein; MRI, magnetic resonance imaging; mRS, the modified Rankin Scale; NMDA, N-methyl-D-aspartate; WBC, white blood cell.

infections; format correction: 2) they did not fulfill the probable AE criteria (e.g, AE mimics such as Creutzfeldt–Jakob disease, metabolic encephalopathy, neoplastic disorders and cerebrovascular disease (Identification of these disorders was based on history, physical examination, laboratory tests, and auxiliary tests; auxiliary tests used are listed in **Supplementary Table 2**); other diseases screened from the electronic database such as meningitis); 3) they had received immunotherapy before hospital admission or this admission was not their index AE admission; 4) they were under the age of 18; 5) they were admitted to the ICU immediately at hospitalization; or 6) they had incomplete medical records. Early ICU admission was defined as admission to the ICU at any time point within two weeks of hospitalization and patients admitted to the ICU beyond 14 days of hospitalization were classified into "non-early ICU admission" group. The protocol was approved by the institutional ethics board of Tongji Hospital, Tongji Medical College of Huazhong University of Science and Technology (ID: TJ-IRB20211221).

Data Collection

The following details about the acute phase of the disease were obtained by 3 neurologists (W-CM, W-HT and Z-YY): (1) demographic information (sex, age); (2) clinical features: comorbidities including hypertension, diabetes and autoimmune diseases; prodromal symptoms such as fever, headache, nonspecific respiratory or gastrointestinal symptoms and other nonspecific viral-like symptoms; the symptoms at onset and all of the symptoms that were present from the onset to hospital admission; the date of onset, the date of hospital admission and discharge; (3) laboratory results: the results of blood tests within 24 hours and the first laboratory CSF sample analysis after admission. For antibody detection, blood and CSF samples were sent to the same laboratory for detection of antibody types and titers using cell-based assay (CBA) in an indirect immunofluorescence (IIF) test and immunospot assay. Antibody titer was defined as low (+, 1:10 in blood or 1:1 in CSF), moderate (++, ≤1:100 in blood or ≤1:10 in CSF), or high (+++, ≥1:320 in blood or ≥1:32 in CSF), with initial dilution titers of CSF and serum of 1:1 vs. 1:10. Six basic types of antibodies were detected for every patient: anti-N-methyl-D-aspartate receptor (NMDAR) antibody, anti-α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid 1 (AMPA1) receptor antibody, and anti-AMPA2 receptor antibody, anti-leucine-rich glioma-inactivated 1 (LGI1) antibody, anti-gamma-aminobutyric acid-B receptor ($GABA_BR$) antibody and anti-contactin-associated protein-like 2 (CASPR2) antibody. Other optional antibody types including anti-$GABA_AR$ antibody, anti-dipeptidyl-peptidase-like protein-6 (DPPX) antibody, anti-mGluR5 antibody, anti-glutamic acid decarboxylase 65 (GAD65) antibody, anti-myelin oligodendrocyte glycoprotein (MOG) antibody, anti-Ma2 antibody, anti-Dopamine 2 receptor (D_2R) antibody, anti-Hu antibody and so on. (4) imaging and electroencephalography (EEG) data: the first results of magnetic resonance imaging (MRI) and EEG; (5) therapeutic data, including first-line immunotherapy (corticosteroids, intravenous immunoglobulin, plasma exchange) and second-line immunotherapy (cyclophosphamide, mycophenolate mofetil, and rituximab) (3, 20, 21); and (6) the scale data: the CASE and mRS scores at the time of hospital admission.

Scale Assessment

The CASE and mRS scores were assessed simultaneously upon hospital admission. The CASE contains nine items: seizures (current status), memory dysfunction, psychiatric symptoms (delusion, hallucination, disinhibition, aggression), consciousness, language problems, dyskinesia/dystonia, gait instability and ataxia, brainstem dysfunction, and weakness. Each item was based on a 3-point grading system, except for the item ''brainstem dysfunction", which is rated by the number of symptoms (gaze paresis, tube feeding, and ventilator care due to hypoventilation), with one point given for each symptom and a maximum of three points (16). The mRS has six grades (0-5) as follows: 0 = no symptoms, 1 = no significant disability: able to carry out all usual activities despite the presence of symptoms, 2 = Slight disability: unable to perform all usual activities but able to look after their own affairs without assistance, 3 = Moderate disability: requiring some help but able to walk without assistance, 4 = Moderately severe disability: unable to walk or attend bodily needs without assistance, 5 = Severe disability: bedridden, incontinent and requiring constant nursing care and attention (15). Two neurologists (W-HT and Z-YY), who were blinded to the research purpose, evaluated the scales independently by reviewing the detailed medical records, and consensus was achieved after discussion in any discrepant cases. If no consensus was reached, a third senior neurologist (H-SS) made the final decision.

Statistical Analysis

Categorical variables are shown as counts and percentages and were analyzed by Pearson's chi-squared test or Fisher's exact test. Continuous variables are expressed as the mean ± SD or medians (quartiles), and Student's t test or the Mann–Whitney U test was used to compare the group data accordingly. EEG and MRI data were missing in 75 (32.1%) and 8 (3.4%) patients because these examinations were either not completed or the results were not recorded in the medical records, and these patients were classified into the "unknown" group. Variables with P values < 0.05 in univariate analyses were subjected to multivariate logistic regression analysis with a stepwise elimination procedure to obtain an optimized model in terms of a minimal Akaike's information criterion (AIC) value. The AE-related management decision based on the judgment of the attending clinicians was a reflection of the disease characteristics, and not all treatments were performed prior to ICU admission. Therefore, the treatment data were not included as risk factors in the multivariate logistic regression analysis. Variables with P values < 0.05 in the optimized multivariate regression model were used to build the final prediction model. Each model was calibrated by a calibration curve, which is actually a visualization of the Hosmer–Lemeshow test. The discriminatory ability of the models was assessed by calculating the area under the receiver operating characteristic (ROC) curves (AUC). ROC analysis was also used to calculate the optimal cutoff values, and these were determined by maximizing the Youden index. The accuracy of the optimal cutoff value was assessed by the sensitivity and

specificity. Internal validation of the models was performed using bootstraps with 1000 replicates (22). To determine if any of the candidate models outperformed the others, we used the DeLong test (23) to explore each of the model pairs for a difference in the AUC values. Two-sided values of $P<0.05$ were considered statistically significant. All analyses were performed with IBM SPSS software, version 24 (SPSS Inc., Chicago, IL, USA), R software version 3.6.1 (The R Foundation for Statistical Computing, Vienna, Austria; http://www.r-project.org) and GraphPad Prism 8. We prepared this article using STROBE, which is the guideline for observational study reports.

RESULTS

Clinical Characteristics of AE

From January 2013 to October 2021, a total of 769 patients with potential encephalitis were screened from the electronic database, of which 344 met the diagnosis criteria of probable or definite AE, and 234 patients were included in the final analysis after excluding the following patients: 43 patients received immunotherapy before hospitalization/non-index AE admission, 27 patients aged under 18 years old, and 40 patients admitted to the ICU immediately upon hospitalization. The flow chart of the study is shown in **Figure 1**. We did not formally calculate the sample size because the current number of patients was determined by the availability of existing data from the introduction of autoimmune encephalitis antibody testing. Of the enrolled patients, 54.7% (128/234) tested positive for neuronal antibodies, which included anti-NMDAR (n=69), LGI1 (n=14), CASPR2 (n=9), $GABA_BR$ (n =14), AMPA (n=3), DPPX (n=7), GAD65 (n=1), mGluR 5 (n=1), MOG (n=3), NMDAR/AMPA (n=1), LGI1/$GABA_BR$ (n=2), LGI1/CASPR2 (n=2), LGI1/AMPA (n=1) and $GABA_BR$/MOG (n=1). Details of antibodies are shown in **Supplementary Table 3**. Five patients were autoantibody negative but were clinically diagnosed with definite autoimmune limbic encephalitis, and the remaining 101 (43.2%) patients fulfilled the criteria for autoantibody-negative but probable AE. The characteristics of the patients are summarized in **Table 1**. The median age of the 234 patients (56.0% males) was 39.0 (IQR 26.0-54.3) years. All patients were in the acute phase of the index admission, and 73.5% (172/234) had an interval of less than 1 month from symptom onset to hospital admission. The timeline of patients from hospital admission to discharge is shown in **Supplementary Figure 1**. Epilepsy was the most common initial symptom (42.3%) and was the most common symptom from onset to hospital admission (52.6%), followed by psychiatric symptoms (50.4%). Half of the patients (52.6%) had prodromal symptoms. Two hundred and one (85.9%) patients received immunotherapy, while 14.1% of

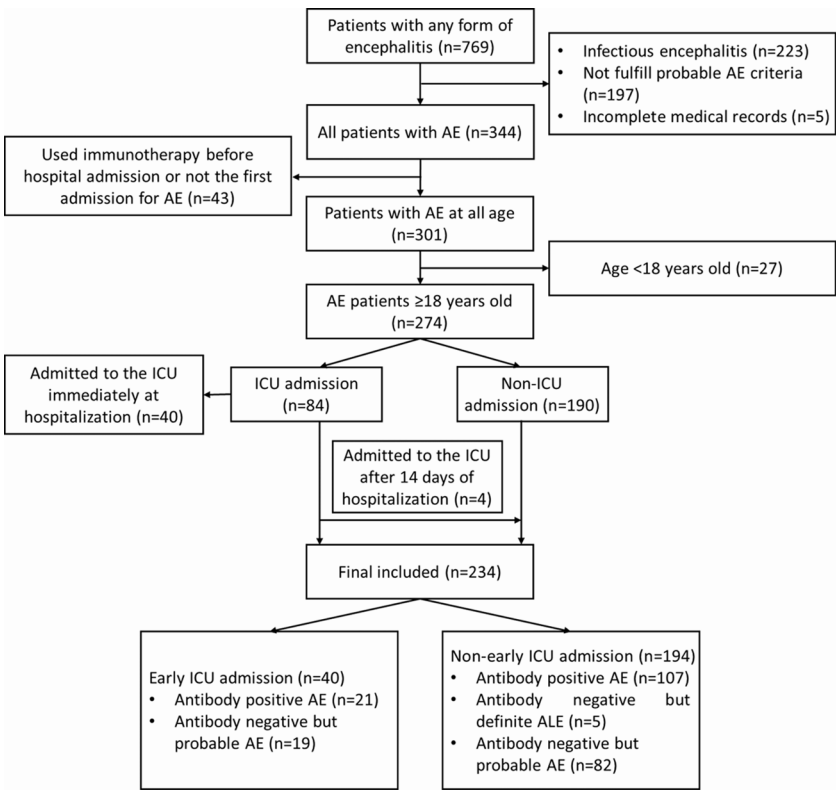


FIGURE 1 | Flow chart of study patients.

TABLE 1 | Characteristics of AE patients with and without need for ICU care.

Characteristics	Total (n = 234)	Non-early ICU admission (n = 194)	Early ICU admission (n = 40)	*P*
Age	39.0 (26.0-54.3)	39.5 (25.8-56.0)	35.0 (27.0-48.0)	0.181
Gender (male)	131 (56.0%)	107 (55.2%)	24 (60.0%)	0.604
Positive antibody[a]	128 (54.7%)	107 (55.2%)	21 (52.5%)	0.862
Definite AE	133 (56.8%)	112 (57.7%)	21 (52.5%)	0.600
Comorbidities				
Tumor	17 (7.3%)	14 (7.2%)	3 (7.5%)	1.000
Hypertension	35 (15.0%)	32 (16.5%)	3 (7.5%)	0.222
Diabetes mellitus	18 (7.7%)	16 (8.2%)	2 (5.0%)	0.707
Autoimmune disease	12 (5.1%)	9 (4.6%)	3 (7.5%)	0.724
Interval from symptoms onset to hospital admission (months)				0.024
>3	12 (5.1%)	12 (6.2%)	0 (0.0%)	
1-3	50 (21.4%)	46 (23.7%)	4 (10.0%)	
≤1	172 (73.5%)	136 (70.1%)	36 (90.0%)	
Prodromal symptoms	123 (52.6%)	93 (47.9%)	30 (75.0%)	0.003
Onset symptoms				
Epilepsy	99 (42.3%)	79 (40.7%)	20 (50.0%)	0.296
Psychiatric/cognition disturbances	102 (43.6%)	85 (43.8%)	17 (42.5%)	1.000
Consciousness disorders	14 (6.0%)	11 (5.7%)	3 (7.5%)	0.938
Symptoms from onset to hospital admission				
Epilepsy	123 (52.6%)	98 (50.5%)	25 (62.5%)	0.223
Short-term memory dysfunction	65 (27.8%)	58 (29.9%)	7 (17.5%)	0.124
Psychiatric symptoms	118 (50.4%)	90 (46.4%)	28 (70.0%)	0.009
Consciousness disorders	52 (22.2%)	33 (17.0%)	19 (47.5%)	< 0.001
Language dysfunction	38 (16.2%)	30 (15.5%)	8 (20.0%)	0.483
Extrapyramidal symptoms	13 (5.6%)	10 (5.2%)	3 (7.5%)	0.833
Autonomic dysfunction	12 (5.1%)	9 (4.6%)	3 (7.5%)	0.724
Sleep disorders	26 (11.1%)	23 (11.9%)	3 (7.5%)	0.602
CSF test				
Elevated CSF pressure[b]	59 (25.2%)	40 (20.6%)	19 (47.5%)	0.001
Elevated CSF WBC count[b]	116 (49.6%)	85 (43.8%)	31 (77.5%)	<0.001
Elevated CSF total protein[b]	81 (34.6%)	66 (34.0%)	15 (37.5%)	0.716
Blood test				
Elevated WBC count[b]	61 (26.1%)	46 (23.7%)	15 (37.5%)	0.078
Anemia[b]	77 (32.9%)	65 (33.5%)	12 (30.0%)	0.716
Platelet (×10^9/L)	226.0 (188.0-272.0)	224.0 (188.8-272.0)	235.5 (185.5-277.8)	0.801
Elevated fasting blood glucose[b]	49 (20.9%)	31 (16.0%)	18 (45.0%)	< 0.001
Impaired hepatic function	42 (17.9%)	33 (17.0%)	9 (22.5%)	0.497
Hypokalemia[b]	26 (11.1%)	21 (10.8%)	5 (12.5%)	0.783
Na^+				0.006
Normal [b]	195 (83.3%)	168 (86.6%)	27 (67.5%)	
Decreased	33 (14.1%)	23 (11.9%)	10 (25.0%)	
Increased	6 (2.6%)	3 (1.5%)	3 (7.5%)	
Cl^-				0.060
Normal [b]	175 (74.8%)	150 (77.3%)	25 (62.5%)	
Decreased	55 (23.5%)	42 (21.6%)	13 (32.5%)	
Increased	4 (1.7%)	2 (1.0%)	2 (5.0%)	
Hypocalcemia[b]	36 (15.4%)	26 (13.4%)	10 (25.0%)	0.089
Creatinine (umol/L)	65.5 (55.0-76.0)	66.5 (55.0-76.0)	64.0 (53.3-77.0)	0.655
Uric acid (umol/L)	258.7 (191.5-321.5)	264.0 (202.5-324.3)	231.7 (150.8-304.6)	0.064
EEG				0.031
Normal	68 (29.1%)	63 (32.5%)	5 (12.5%)	
Abnormal[c]	91 (38.9%)	70 (36.1%)	21 (52.5%)	
Unknown	75 (32.1%)	61 (31.4%)	14 (35.0%)	
MRI				0.052
Normal	90 (38.5%)	76 (39.2%)	14 (35.0%)	
Abnormal[c]	136 (58.1%)	114 (58.8%)	22 (55.0%)	
Unknown	8 (3.4%)	4 (2.1%)	4 (10.0%)	
Treatment				
Immunotherapy	201 (85.9%)	163 (84.0%)	38 (95.0%)	0.082
First-line	200 (85.5%)	162 (83.5%)	38 (95.0%)	0.082
Second-line	7 (3.0%)	4 (2.1%)	3 (7.5%)	0.184
ASMs	124 (53.0%)	95 (49.0%)	29 (72.5%)	0.009
Scale				
mRS	2 (1-3)	2 (1-3)	3 (2-4)	< 0.001
CASE	4 (2-5)	3 (2-5)	7 (5-11)	< 0.001

[a]*Antibodies against cell-surface, synaptic, or onconeural protein.*

[b]*Normal values: CSF pressure (80-180 mmH2O), CSF WBC count (≤5/mm3), CSF protein (150-450 mg/L), blood WBC ((4-10) ×10^9/L), Na+ (135-145 mmol/L) and Cl- (98-110 mmol/L). anemia was defined as < 120 g/L in females and children and < 135 g/L in males; elevated fasting blood glucose was defined as >6.1mmol/L; hypokalemia was defined as < 3.5 mmol/L; hypocalcemia was defined as < 2.15 mmol/L.*

[c]*Abnormal EEG results: epileptic discharge, delta brush, or slow wave. Abnormal brain MRI results: brain MRI hyperintense signal on T2-weighted fluid-attenuated inversion recovery sequences highly restricted to one or both medial temporal lobes or in multifocal areas involving gray matter, white matter, or both compatible with demyelination or inflammation.*

patients rejected any immunotherapies due to a mild disease severity, poor economic conditions, or an intolerance of side effects. Forty patients (17.1%) deteriorated for the early of their hospitalization and were admitted to the ICU. The common direct reasons for early ICU admission were status epilepticus (32.5%, 13/40), unstable vital sign (respiratory failure or blood pressure drop) (17.5%, 7/40), severe psychiatric symptoms (15.0%, 6/40) and decreased level of consciousness (7.5%, 3/40), details are shown in **Supplementary Table 4**.

Factors Associated With Early ICU Admission Among AE Patients

As shown in **Table 1**, the total CASE and mRS scores were both significantly associated with ICU admission (P<0.001). We found that the CASE and mRS scores were statistically correlated (r=0.642, P<0.001) (**Figure 2**), which was consistent with previous studies (7, 17, 18). Other variables that were significant associated with admission to the ICU included time from symptom onset to hospital admission of less than 1 month (P=0.024), prodromal symptoms (P=0.003); symptoms from onset to hospital admission: psychiatric symptoms (P=0.009), consciousness disorders (P<0.001); laboratory tests: elevated CSF pressure (P=0.001), elevated CSF WBC count (P<0.001), elevated fasting blood glucose (P<0.001), Na^+ (P=0.006); abnormal or unknown EEG results (P=0.031), and anti-seizure medications (ASMs) therapy (P=0.009).

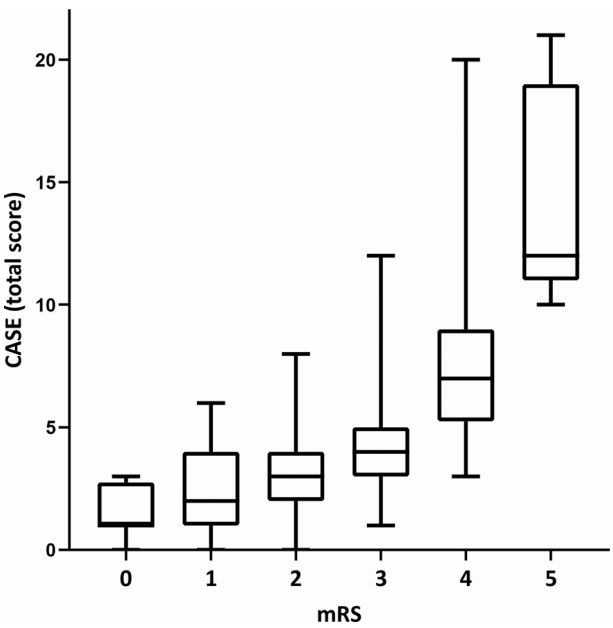


FIGURE 2 | The total CASE score according to the mRS at the time of hospital admission. CASE, the Clinical Assessment Scale for Autoimmune Encephalitis. mRS, the modified Rankin scale. The CASE and mRS scores were statistically correlated (r = 0.642, P < 0.001).

Risk Models for Prediction Early ICU Admission

Model Construction

Next, we conducted two multivariate logistic regression analyses. One included the CASE score and the variables with P<0.05 in the univariate analysis, and the other included the mRS score and variables with P<0.05 in the univariate analysis. The results of the optimized multivariate regression model after the variable selection are shown as forest plots (**Figure 3**). As seen from the forest plots, there were four significant independent predictors (CASE, prodromal symptoms, elevated fasting blood glucose and elevated CSF WBC count) in the CASE model and five (mRS, prodromal symptoms, abnormal EEG results, elevated fasting blood glucose and elevated CSF WBC count) in the mRS model.

Based on the results of the multivariate analysis, we developed the following four candidate predictive models:

Model 1: CASE: CASE alone;

Model 2: CASE-plus: CASE + prodromal symptoms + elevated fasting blood glucose + elevated CSF WBC count;

Model 3: mRS: mRS alone;

Model 4: mRS-plus: mRS + prodromal symptoms + abnormal EEG results + elevated fasting blood glucose + elevated CSF WBC count

Model Evaluation

Discrimination

The ROC curves for each model are presented in **Figure 4A1**. **Figure 4A2** shows the uncorrected AUC values and the bootstrapped optimism corrected AUC values for each model. The bootstrap-adjusted AUC values were similar to the uncorrected AUC values. The optimal cutoff scores, which were derived from the ROC analysis, of Model 1 and Model 3 were 4.5 and 2.5, respectively. We also summarized the sensitivity and specificity in estimating the risk of ICU admission using 4.5 and 2.5 as the cutoff values (**Figure 4A2**). To determine if any of the four candidate models outperformed the others, we used DeLong's test (23) to test each of the four correlated possible model pairs for a difference in predicting the uncorrected AUC scores. We found a significant difference in the discriminant ability of each pair (P<0.05), except for Model 1 and Model 4 (P=0.671), and the P values are listed in **Figure 4A3**. By combining **Figure 4A**, we can conclude that Model 2 performed best, followed by Model 1, Model 4 and Model 3, with AUCs of 0.897 (95% CI 0.842-0.953, P<0.001), 0.850 (95% CI 0.773-0.927, P<0.001), 0.833 (95% CI 0.760-0.906, P<0.001) and 0.695 (95% CI 0.599-0.792, P<0.001), respectively (**Figure 4A2**).

Calibration

The Hosmer–Lemeshow test of these four models showed (with P values of 0.14, 0.44, 0.35 and 0.40 for Models 1, 2, 3 and 4, respectively) that the calibrations of the four models were adequate and that the models were correctly specified. The apparent and bootstrapped calibration curves for each model (**Figure 4B**) showed the excellent agreement between the observed outcomes and the predictions, with predicted probabilities positioned on or around a 45° line of the plot.

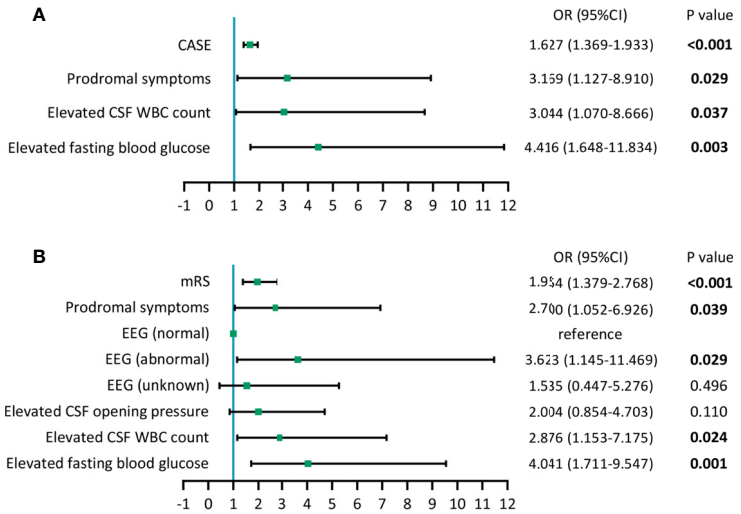
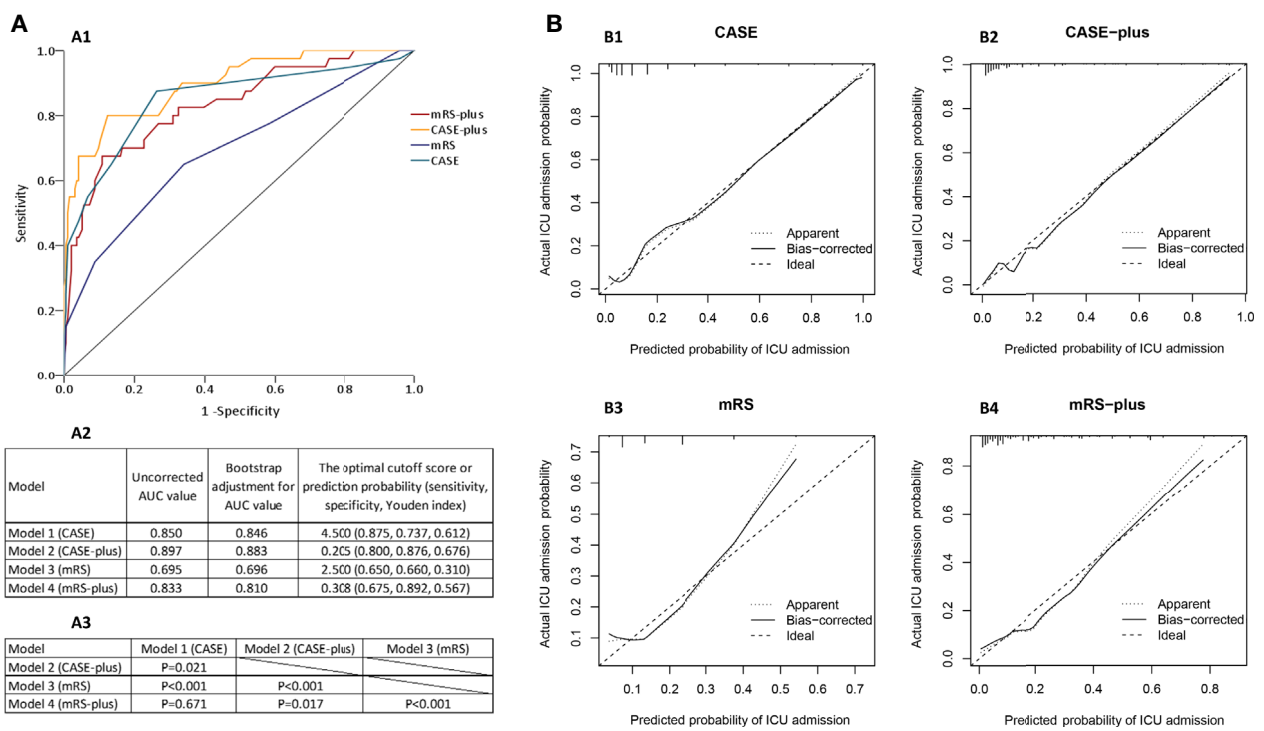


FIGURE 3 | Forest plots of models of multivariate logistic regression analysis. **(A)** Results of multivariate logistic regression analysis containing CASE. **(B)** Results of multivariate logistic regression analysis containing mRS.



Model	Uncorrected AUC value	Bootstrap adjustment for AUC value	The optimal cutoff score or prediction probability (sensitivity, specificity, Youden index)
Model 1 (CASE)	0.850	0.846	4.500 (0.875, 0.737, 0.612)
Model 2 (CASE-plus)	0.897	0.883	0.265 (0.800, 0.876, 0.676)
Model 3 (mRS)	0.695	0.696	2.500 (0.650, 0.660, 0.310)
Model 4 (mRS-plus)	0.833	0.810	0.308 (0.675, 0.892, 0.567)

Model	Model 1 (CASE)	Model 2 (CASE-plus)	Model 3 (mRS)
Model 2 (CASE-plus)	P=0.021		
Model 3 (mRS)	P<0.001	P<0.001	
Model 4 (mRS-plus)	P=0.671	P=0.017	P<0.001

FIGURE 4 | Risk prediction models for early ICU admission in patients with AE. **(A1)** Receiver operating characteristic (ROC) curves of the four models for predicting early ICU admission. **(A2)** Uncorrected and the bootstrap-adjusted AUC values and optimal cutoff values of the four models. The AUC values of CASE, CASE-plus, mRS, mRS-plus were 0.850 (95% CI 0.773-0.927, P<0.001), 0.897 (95% CI 0.842-0.953, P<0.001), 0.695 (95% CI 0.599-0.792, P<0.001), and 0.833 (95% CI 0.760-0.906, P<0.001), respectively. **(A3)** P values for the pairwise comparison of original AUC values of the four models. **(B)** The calibration curves of the four models in predicting ICU admission. The y-axis represents the actual probability of ICU admission, and the x-axis represents the predicted probability of ICU admission. A perfect model would fully match the 45° ideal line.

DISCUSSION

In this retrospective study of critically ill and noncritically ill patients with AE who were treated in a tertiary hospital, we investigated the association between the patient characteristics and early ICU admission. In the univariate analysis, both the CASE and mRS scores were significantly associated with ICU admission. Next, we developed four risk prediction models: CASE, CASE-plus (CASE plus = CASE + prodromal symptoms + elevated fasting blood glucose + elevated CSF WBC count), mRS, and mRS-plus (mRS plus = mRS + prodromal symptoms + abnormal EEG results + elevated fasting blood glucose + elevated CSF WBC count). Among the four models, the CASE-plus model demonstrated the best performance.

CASE is the first clinical severity scale that was specifically designed for the various syndromes of AE. Although lacking large-scale validation, Cai and Zhang et al. proved that CASE performed well and had a significant positive correlation with mRS in two groups of Chinese patients with antibody-positive AE (17, 18). As a comprehensive scale covering multiple domains of AE, CASE has inherent advantages when compared to mRS. First, the detailed assessment of various specific clinical manifestations allows CASE to represent the overall severity of the disease, especially in patients with nonmotor symptoms and who develop common intensive care signs, such as status epilepticus, coma and mechanical ventilation due to central hypoventilation, and these variables are not included in mRS. Second, the total score of CASE ranges from 0 to 27, and mRS is a 6-point scale (15, 16). This discrepancy makes CASE more precise and sensitive in differentiating the severity of disease within the same range of measurements that are also defined by mRS (16). In our study, total CASE scores of 4.5/27 and total mRS scores of 2.5/6 at the time of hospitalization were the optimal cutoff values in predicting ICU admission; to some extent, these values reflect the early predictive value and sensitivity of the CASE score compared with the mRS score. The cutoff value of the total CASE score also implied that patients with multiple moderate to severe symptoms at the time of admission were more likely to progress to critical conditions. In such cases, quantified symptoms can serve as an alert, which allows patients to receive advanced treatment in a timely manner. Several limitations of CASE may also exist. First, CASE is more complicated and time-consuming than mRS, and it is difficult to use CASE to evaluate some symptoms in specific situations. For example, in sedated patients, the assessment of symptoms such as language and memory can be challenging. Second, the score of each item is unweighted, and CASE is a three-point scale, which may be unfair in assessing some fatal symptoms, such as central hypoventilation. Overall, the total CASE score might be a better optimal predictor of early ICU admission than the mRS score because of its more comprehensive characteristics.

CASE and mRS reflect the pro tempore status of the patient; however, other symptoms can develop in the early course of AE. Also, laboratory and imaging abnormalities may contribute to disease deterioration. We then included more variables associated with AE to screen out other potential risk factors for ICU admission. We found that prodromal symptoms, abnormal EEG results, elevated fasting blood glucose and elevated CSF WBC count were independent predictors in the multivariable analysis. In our study, 52.6% of patients had prodromal symptoms, which is consistent with previous studies (34%-62.8%) (2, 24, 25). Prodromal symptoms are nonspecific, vary in presentation and, more importantly, indicate infection (26). In fact, infections have long been suspected to play a role in triggering or enhancing the autoimmune process (26, 27). Accumulating evidence suggests that viral infections may be associated with the development of AE (28, 29). In other autoimmune diseases, such as myasthenia gravis (MG) (30) and Guillain–Barre syndrome (GBS) (31, 32), patients who triggered by infection often have a higher ICU admission rate and a more unfavorable prognosis. Elevated CSF WBC count, an indicator of inflammation within the central nervous system (33), was also found to be associated with ICU admission in another study (6). Previously elevated fasting blood glucose might have been ignored as a variable in AE. However, in the setting of acute inflammation, stress hyperglycemia is often observed, which is common in critically ill patients and appears to be a marker of disease severity (34). We found that patients with elevated fasting glucose levels were four times more likely to be admitted to the ICU than patients without elevated fasting glucose levels. However, only 11 of the 49 patients with abnormal fasting glucose levels were diagnosed with diabetes or with an impaired fasting glucose tolerance, indicating that the majority of these patients had acute glucose instability. Therefore, it is feasible for an abnormal fasting blood glucose level at admission to serve as an indicator for the early identification of critical illness in AE cases. Thus, we postulate that infection-triggered and multisystem-involved patients with AE may suffer a more severe disease index.

To generate prediction models with a higher sensitivity, we integrated the above risk factors into CASE and mRS. We found that the predictability of each ICU admission model (except for mRS), as measured by the area under the ROC curve, was more than 0.80. Models containing the CASE score performed better than those containing the mRS score. The CASE-plus and mRS-plus model performed better than the CASE or mRS models, respectively. The best-performing model was the CASE-plus model. This result confirmed that considering both clinical phenotypes and biological disturbances would precisely predict disease progression.

Several limitations should be noted in our study. First, the retrospective nature of the design makes it difficult to control for confounding factors and may lead to possible information bias. AE is a disease that has gradually received attention with the development of antibody detection technology in recent years. Many of the large studies on AE are also retrospective (35, 36), and the results are repeatable. The assessment of CASE was performed retrospectively, and there would inevitably be a small number of patients with incomplete documentation of some items, such as the grading of dyskinesia/dystonia and memory dysfunction. For these patients, we carefully reviewed the medical records, and if no relevant symptoms were recorded throughout the course of the disease, it was considered not present. Nonetheless, CASE has been used in retrospective studies (17, 18, 37) and it is feasible to consider the results of our study are reliable. Second, this

is a single-center study, and selection bias may exist. For example, the ICU admission rate was lower than that in previous studies in Western countries (13, 38). As a national tertiary hospital, Tongji Hospital receives a wide coverage and a great number of patients, which makes the patients representative of the general population. In fact, the overall severity and severity distribution of the patients in our study are comparable to those from several domestic studies (17, 18, 39, 40). Third, although the internal validation in our study showed good efficacy, this study was not externally validated. Generalizing the conclusions of this study requires validation in further external datasets, and a prospective, multicenter study with a larger sample size will be necessary in the future.

To the best of our knowledge, this is the first study to integrate the scale-based disease severities of patients with AE into predictive models for ICU admission. Both the CASE and mRS models could accurately predict the risk of ICU admission in AE patients, but the CASE model performed better. Patients with CASE scores ≥5 were more likely to be admitted to the ICU. Adding prodromal symptoms, elevated fasting blood glucose and CSF WBC count to the CASE model could improve the predictive ability of the existing grading scale.

DATA AVAILABILITY STATEMENT

The data supporting the findings of this study are available from the corresponding authors, upon reasonable request.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Institutional Ethics Board of Tongji Hospital Tongji Medical College of Huazhong University of Science and Technology (ID: TJ-IRB20211221). Written informed consent from the participants' legal guardian/next of kin was not required to participate in this study in accordance with the national legislation and the institutional requirements.

AUTHOR CONTRIBUTIONS

CW designed the study, acquired data, performed the analysis and drafted the work; YF analyzed and interpreted of the data; YZ and HW acquired data; SH designed the study. SH and SZ led the study, critically reviewed and revised the manuscript. All authors contributed to the article and approved the submitted version.

FUNDING

This study was supported by the Hubei Technological Innovation Special Fund (CN) [Grant NO.2019ACA132] and Hubei Natural Science Foundation Grant NO. 2020CFB805.

ACKNOWLEDGMENTS

The authors would like to thank all the participants for their valuable data, cooperation, and participation.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fimmu.2022.916111/full#supplementary-material

REFERENCES

1. Diaz-Arias LA, Pardo CA, Probasco JC. Autoimmune Encephalitis in the Intensive Care Unit. *Neurointensive Care Unit* (2020) 249–63. doi: 10.1007/978-3-030-36548-6_17
2. Schubert J, Brämer D, Huttner HB, Gerner ST, Fuhrer H, Melzer N, et al. Management and Prognostic Markers in Patients With Autoimmune Encephalitis Requiring ICU Treatment. *Neurol Neuroimmunol Neuroinflamm* (2019) 6(1):e514. doi: 10.1212/nxi.0000000000000514
3. Graus F, Titulaer MJ, Balu R, Benseler S, Bien CG, Cellucci T, et al. A Clinical Approach to Diagnosis of Autoimmune Encephalitis. *Lancet Neurol* (2016) 15 (4):391–404. doi: 10.1016/s1474-4422(15)00401-9
4. Mittal MK, Rabinstein AA, Hocker SE, Pittock SJ, Wijdicks EF, McKeon A. Autoimmune Encephalitis in the ICU: Analysis of Phenotypes, Serologic Findings, and Outcomes. *Neurocrit Care* (2016) 24(2):240–50. doi: 10.1007/s12028-015-0196-8
5. Harutyunyan G, Hauer L, Dünser MW, Moser T, Pikija S, Leitinger M, et al. Risk Factors for Intensive Care Unit Admission in Patients With Autoimmune Encephalitis. *Front Immunol* (2017) 8:835. doi: 10.3389/fimmu.2017.00835
6. Broadley J, Wesselingh R, Seneviratne U, Kyndt C, Beech P, Buzzard K, et al. Prognostic Value of Acute Cerebrospinal Fluid Abnormalities in Antibody-Positive Autoimmune Encephalitis. *J Neuroimmunol* (2021) 353:577508. doi: 10.1016/j.jneuroim.2021.577508
7. Broadley J, Wesselingh R, Seneviratne U, Kyndt C, Beech P, Buzzard K, et al. Peripheral Immune Cell Ratios and Clinical Outcomes in Seropositive Autoimmune Encephalitis: A Study by the Australian Autoimmune Encephalitis Consortium. *Front Immunol* (2020) 11:597858. doi: 10.3389/fimmu.2020.597858
8. Levraut M, Bourg V, Capet N, Delourme A, Honnorat J, Thomas P, et al. Cerebrospinal Fluid IL-17a Could Predict Acute Disease Severity in Non-NMDA-Receptor Autoimmune Encephalitis. *Front Immunol* (2021) 12:673021. doi: 10.3389/fimmu.2021.673021
9. Sonneville R, Gault N, de Montmollin E, Klein IF, Mariotte E, Chemam S, et al. Clinical Spectrum and Outcomes of Patients With Encephalitis Requiring Intensive Care. *Eur J Neurol* (2015) 22(1):6–16.e1. doi: 10.1111/ene.12541
10. Krishnan S, Sarma G, Sarma S, Kishore A. Do Nonmotor Symptoms in Parkinson's Disease Differ From Normal Aging? *Mov Disord* (2011) 26 (11):2110–3. doi: 10.1002/mds.23826
11. Lee SA, Choi EJ, Jeon JY, Han SH, Kim HW, Lee GH, et al. Insomnia Moderates the Association Between Recurrent Seizures and Emotional Instability in Persons With Epilepsy. *Epilepsy Behav* (2021) 125:108414. doi: 10.1016/j.yebeh.2021.108414
12. Ryu WS, Hong KS, Jeong SW, Park JE, Kim BJ, Kim JT, et al. Association of Ischemic Stroke Onset Time With Presenting Severity, Acute Progression, and Long-Term Outcome: A Cohort Study. *PLoS Med* (2022) 19(2):e1003910. doi: 10.1371/journal.pmed.1003910

13. Titulaer MJ, McCracken L, Gabilondo I, Armangué T, Glaser C, Iizuka T, et al. Treatment and Prognostic Factors for Long-Term Outcome in Patients With Anti-NMDA Receptor Encephalitis: An Observational Cohort Study. *Lancet Neurol* (2013) 12(2):157–65. doi: 10.1016/s1474-4422(12)70310-1
14. Pruetarat N, Netbaramee W, Pattharathitikul S, Veeravigrom M. Clinical Manifestations, Treatment Outcomes, and Prognostic Factors of Pediatric Anti-NMDAR Encephalitis in Tertiary Care Hospitals: A Multicenter Retrospective/Prospective Cohort Study. *Brain Dev* (2019) 41(5):436–42. doi: 10.1016/j.braindev.2018.12.009
15. van Swieten JC, Koudstaal PJ, Visser MC, Schouten HJ, van Gijn J. Interobserver Agreement for the Assessment of Handicap in Stroke Patients. *Stroke* (1988) 19(5):604–7. doi: 10.1161/01.str.19.5.604
16. Lim JA, Lee ST, Moon J, Jun JS, Kim TJ, Shin YW, et al. Development of the Clinical Assessment Scale in Autoimmune Encephalitis. *Ann Neurol* (2019) 85 (3):352–8. doi: 10.1002/ana.25421
17. Cai MT, Lai QL, Zheng Y, Fang GL, Qiao S, Shen CH, et al. Validation of the Clinical Assessment Scale for Autoimmune Encephalitis: A Multicenter Study. *Neurol Ther* (2021) 10(2):985–1000. doi: 10.1007/s40120-021-00278-9
18. Zhang Y, Tu E, Yao C, Liu J, Lei Q, Lu W. Validation of the Clinical Assessment Scale in Autoimmune Encephalitis in Chinese Patients. *Front Immunol* (2021) 12:796965. doi: 10.3389/fimmu.2021.796965
19. Jeon CY, Neidell M, Jia H, Sinisi M, Larson E. On the Role of Length of Stay in Healthcare-Associated Bloodstream Infection. *Infect Control Hosp Epidemiol* (2012) 33(12):1213–8. doi: 10.1086/668422
20. Abboud H, Probasco J, Irani SR, Ances B, Benavides DR, Bradshaw M, et al. Autoimmune Encephalitis: Proposed Recommendations for Symptomatic and Long-Term Management. *J Neurol Neurosurg Psychiatry* (2021) 92(8):897–907. doi: 10.1136/jnnp-2020-325302
21. Tan C, Jiang Y, Zhong M, Hu Y, Hong S, Li X, et al. Clinical Features and Outcomes in Pediatric Autoimmune Encephalitis Associated With CASPR2 Antibody. *Front Pediatr* (2021) 9:736035. doi: 10.3389/fped.2021.736035
22. Choi H, Detyniecki K, Bazil C, Thornton S, Crosta P, Tolba H, et al. Development and Validation of a Predictive Model of Drug-Resistant Genetic Generalized Epilepsy. *Neurology* (2020) 95(15):e2150–e60. doi: 10.1212/wnl.0000000000010597
23. DeLong ER, DeLong DM, Clarke-Pearson DL. Comparing the Areas Under Two or More Correlated Receiver Operating Characteristic Curves: A Nonparametric Approach. *Biometrics* (1988) 44(3):837–45. doi: 10.2307/2531595
24. Yu Y, Wu Y, Cao X, Li J, Liao X, Wei J, et al. The Clinical Features and Prognosis of Anti-NMDAR Encephalitis Depends on Blood Brain Barrier Integrity. *Mult Scler Relat Disord* (2021) 47:102604. doi: 10.1016/j.msard.2020.102604
25. Yang RN, Ge FF, Jiang JW, Wang Y, Zhang W. [Disease Characteristics, Treatment, and Prognosis of Chinese Patients With Autoimmune Encephalitis: A Retrospective Study]. *Sichuan Da Xue Xue Bao Yi Xue Ban* (2022) 53(1):142–8. doi: 10.12182/20220160206
26. Joubert B, Dalmau J. The Role of Infections in Autoimmune Encephalitides. *Rev Neurol (Paris)* (2019) 175(7-8):420–6. doi: 10.1016/j.neurol.2019.07.004
27. Venkatesan A, Benavides DR. Autoimmune Encephalitis and its Relation to Infection. *Curr Neurol Neurosci Rep* (2015) 15(3):3. doi: 10.1007/s11910-015-0529-1
28. Armangue T, Spatola M, Vlagea A, Mattozzi S, Cárceles-Cordon M, Martinez-Heras E, et al. Frequency, Symptoms, Risk Factors, and Outcomes of Autoimmune Encephalitis After Herpes Simplex Encephalitis: A Prospective Observational Study and Retrospective Analysis. *Lancet Neurol* (2018) 17 (9):760–72. doi: 10.1016/s1474-4422(18)30244-8
29. Linnoila J, Pulli B, Armangué T, Planagumà J, Narsimhan R, Schob S, et al. Mouse Model of Anti-NMDA Receptor Post-Herpes Simplex Encephalitis. *Neurol Neuroimmunol Neuroinflamm* (2019) 6(2):e529. doi: 10.1212/nxi.0000000000000529
30. Sakaguchi H, Yamashita S, Hirano T, Nakajima M, Kimura E, Maeda Y, et al. Myasthenic Crisis Patients Who Require Intensive Care Unit Management. *Muscle Nerve* (2012) 46(3):440–2. doi: 10.1002/mus.23445
31. Nithyashree N, Dhanaraj M, Kumar S, Saraswathi MB. Factors Predicting Poor Outcome in Patients With Fulminant Guillaine-Barré Syndrome. *Ann Indian Acad Neurol* (2014) 17(4):463–5. doi: 10.4103/0972-2327.144040
32. Palace JA, Hughes RA. Guillain-Barré Syndrome With Severe Persistent Disability: Relationship to Hyperacute Guillain-Barré Syndrome. *Eur J Neurol* (1994) 1(1):21–7. doi: 10.1111/j.1468-1331.1994.tb00046.x
33. Baunbæk Egelund G, Ertner G, Langholz Kristensen K, Vestergaard Jensen A, Benfield TL, Brandt CT. Cerebrospinal Fluid Pleocytosis in Infectious and Noninfectious Central Nervous System Disease: A Retrospective Cohort Study. *Medicine* (2017) 96(18):e6686. doi: 10.3389/fimmu.2021.774664
34. Marik PE, Bellomo R. Stress Hyperglycemia: An Essential Survival Response! *Crit Care* (2013) 17(2):305. doi: 10.1186/cc12514
35. Shan W, Yang H, Wang Q. Neuronal Surface Antibody-Medicated Autoimmune Encephalitis (Limbic Encephalitis) in China: A Multiple-Center, Retrospective Study. *Front Immunol* (2021) 12:621599. doi: 10.3389/fimmu.2021.621599
36. Balu R, McCracken L, Lancaster E, Graus F, Dalmau J, Titulaer MJ. A Score That Predicts 1-Year Functional Status in Patients With Anti-NMDA Receptor Encephalitis. *Neurology* (2019) 92(3):e244–e52. doi: 10.1212/wnl.0000000000006783
37. Shim Y, Kim SY, Kim H, Hwang H, Chae JH, Choi J, et al. Clinical Outcomes of Pediatric Anti-NMDA Receptor Encephalitis. *Eur J Paediatr Neurol* (2020) 29:87–91. doi: 10.1016/j.ejpn.2020.10.001
38. Cohen J, Sotoca J, Gandhi S, Yeshokumar AK, Gordon-Lipkin E, Geocadin RG, et al. Autoimmune Encephalitis: A Costly Condition. *Neurology* (2019) 92 (9):e964–e72. doi: 10.1212/wnl.0000000000006990
39. Li A, Gong X, Guo K, Lin J, Zhou D, Hong Z. Direct Economic Burden of Patients With Autoimmune Encephalitis in Western China. *Neurol Neuroimmunol Neuroinflamm* (2020) 7(6):e891. doi: 10.1212/nxi.0000000000000891
40. Gu Y, Zhong M, He L, Li W, Huang Y, Liu J, et al. Epidemiology of Antibody-Positive Autoimmune Encephalitis in Southwest China: A Multicenter Study. *Front Immunol* (2019) 10:2611. doi: 10.3389/fimmu.2019.02611

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 25 August 2022
DOI 10.3389/fimmu.2022.918064

OPEN ACCESS

EDITED BY
Barbara M. P. Willekens,
Antwerp University Hospital, Belgium

REVIEWED BY
Ernestina Santos,
University Hospital Center of Porto, Portugal
Tatjana Reynders,
University of Antwerp, Belgium

*CORRESPONDENCE
Yajun Lian
lianyajun369@sina.com

†These authors contributed equally

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Immunology

RECEIVED 12 April 2022
ACCEPTED 01 August 2022
PUBLISHED 25 August 2022

CITATION
Ding J, Xu D, Lv J, Wu T, Li J, Tian M and Lian Y (2022) Pulmonary infection and baseline mRS scores predict poor prognosis in anti-$GABA_BR$ encephalitis. *Front. Immunol.* 13:918064. doi: 10.3389/fimmu.2022.918064

Pulmonary infection and baseline mRS scores predict poor prognosis in anti-$GABA_BR$ encephalitis

Junqing Ding[1†], Dingkang Xu[2,3†], Jie Lv[4†], Tianwen Wu[1], Jinghong Li[1], Mi Tian[1] and Yajun Lian[1*]

[1]Department of Neurology, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, China, [2]Department of Neurosurgery, Beijing Hospital, National Center of Gerontology, Institute of Geriatric Medicine, Chinese Academy of Medical Sciences, Beijing, China, [3]Graduate School of Peking Union Medical College, Beijing, China, [4]Department of Nephrology and Rheumatology, Dongzhimen Hospital, The First Affiliated Hospital of Beijing University of Chinese Medicine, Beijing, China

Purpose: Anti-gamma-aminobutyric-acid type B receptor (anti-$GABA_BR$) encephalitis is a rare autoimmune condition caused by the presence of $GABA_BR$ antibodies in the limbic system. However, its clinical features and prognostic factors are poorly understood. In this study, we aimed to explore factors that affect the response to first-line treatment in patients with anti-$GABA_BR$ encephalitis.

Methods: Thirty-four patients with an initial diagnosis of anti-$GABA_BR$ encephalitis were retrospectively enrolled from December 2015 to June 2021. Clinical features and experimental data recorded within 24 h of admission were extracted from the patients' medical records. The modified Rankin Scale (mRS) was utilized to assess disease severity at admission and functional recovery after immunotherapy. Independent prognostic factors were determined by ordinal logistic regression analysis.

Results: Of the 34 anti-$GABA_BR$ encephalitis patients, 12 (35%) presented with cancer; all of these patients had lung cancer. According to multivariate regression analysis, the cancer group exhibited a decrease in the peripheral blood absolute lymphocyte count (ALC) (odds ratio [OR]: 0.063, 95% confidence interval [CI]: 0.006-0.639, P=0.019) and hyponatremia (OR: 9.268, 95% CI: 1.054-81.502, 0.045). In addition, the neutrophil/lymphocyte ratio (NLR), monocyte/lymphocyte ratio (MLR) and platelet/lymphocyte ratio (PLR) did not significantly differ according to mRS scores in patients receiving first-line treatment. No patients with mild or moderate mRS scores (0-2) at admission developed symptoms after treatment; in contrast, only 11 patients with a severe mRS scores (≥3, 11/18) experienced symptom alleviation. Ordinal regression analysis indicated that worse prognosis was associated with pulmonary infection (OR=9.885, 95% CI: 1.106-88.323, P=0.040) and baseline mRS scores (OR= 24.047, 95% CI: 3.294-175.739, P=0.002) in the adjusted model.

Conclusion: Our findings demonstrate that pulmonary infection and baseline mRS scores are independent risk factors for poor prognosis in patients with anti-$GABA_BR$ encephalitis after first-line treatment. ALC and hyponatremia are potential biomarkers for anti-$GABA_BR$ encephalitis cases accompanied by lung cancer.

KEYWORDS

anti-gamma-aminobutyric-acid B receptor (anti-GABABR) encephalitis, Baseline mRS score, pulmonary infection, prognosis, absolute lymphocyte count (ALC), Hyponatremia

Introduction

Autoimmune encephalitis (AE) is an inflammation of the central nervous system (CNS) triggered by immune system attack of the CNS and the production of aberrant pathogenic autoantibodies (1). AE can be divided into various types according to the production of autoantibodies against neuronal cell surface or synaptic proteins. Anti-$GABA_BR$ encephalitis is the third most frequent AE after anti-N-methyl-D-aspartate receptor (anti-NMDAR) encephalitis and anti-leucine-rich, glioma-inactivated 1 receptor (anti-LGI1) encephalitis. However, anti-$GABA_BR$ encephalitis is relatively rare, accounting for approximately 5% of AE cases (2). Anti-$GABA_BR$ encephalitis, first reported by Lancaster et al. in 2010 (3), is characterized by the presence of limbic encephalitis, including the acute or subacute onset of prominent seizures, cognitive dysfunction, and psychiatric behavior (4). Approximately 50% of these patients harbor an underlying cancer, particularly small-cell lung cancer (SCLC) or a pulmonary neuroendocrine tumor (5–7); therefore, anti-$GABA_BR$ encephalitis is also known as paraneoplastic limbic encephalitis (PLE).

As anti-$GABA_BR$ encephalitis is chiefly mediated by humoral immunity, management of this condition focuses on immunotherapy and the detection and removal of tumors (8). First-line treatments include steroids, intravenous immunoglobulin (IVIG), and plasma exchange (PLEX), either alone or in combination; rituximab, cyclophosphamide, and bortezomib comprise second-line immunotherapies (9). Patients usually respond well to immunotherapy, which alleviates 70%–83.3% of neurological symptoms (10), and treatment of the associated cancer (11).

In general, the interaction between peripheral immune cell ratios and clinical outcomes in AE patients has attracted significant attention. Recent studies of AE have found that a high NLR significantly correlates with long-term functional disability, as measured by the mRS scores, and a reduced response to first-line immunotherapy (12, 13). James Broadley et al. (14) showed that a high NLR was associated with failure of first-line treatment but that a high MLR was not associated with AE patient prognosis. The PLR has recently been associated with prognosis in various diseases, such as lung cancer, affective disorders and diabetic kidney disease (15–17). However, no studies have examined PLR as a prognostic biomarker in AE.

Previous studies of anti-$GABA_BR$ encephalitis have mostly been descriptive, utilizing individual cases or small samples and evaluating clinical symptoms and long-term prognosis. No study has focused on predictive factors for evaluating the use of immunotherapy as first-line treatment. In this study, data from 34 patients admitted to our hospital with an initial diagnosis of anti-$GABA_BR$ encephalitis were analyzed to explore the clinical characteristics of anti-$GABA_BR$ encephalitis and to identify factors that predicted poor prognosis after first-line treatment, allowing combined first-line immunotherapy and second-line immunotherapy to be administered in a timely manner.

Methods

Participants

This retrospective study was approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University in accordance with Helsinki declaration. The patients/proxy provided written informed consent prior to participation in this study. Thirty-four patients who were admitted to the First Affiliated Hospital of Zhengzhou University from December 2015 to June 2021 with an initial diagnosis of anti-$GABA_BR$ encephalitis were selected for inclusion. The diagnosis was based on the consensus for diagnosis and treatment of AE proposed by Chinese experts in 2017. All included patients met the following diagnostic criteria for anti-$GABA_BR$ encephalitis: (1) clinical manifestations of limbic encephalitis, such as the acute or subacute onset of prominent seizures, cognitive dysfunction, and psychiatric behavior; (2) positive results on tests for anti-$GABA_BR$ antibodies in cerebrospinal fluid (CSF) and/or serum; and (3)

received first-line treatment. The exclusion criteria were as follows: (1) anti-$GABA_BR$ encephalitis was confirmed and treated before admission; (2) diagnosis of infectious, toxic, or metabolic encephalopathy and/or another nervous system disease prior to the onset of anti-$GABA_BR$ encephalitis; (3) incomplete clinical data; or (4) loss at follow-up. For each patient, follow-up evaluations were conducted by telephone or outpatient interviews for at least 6 months.

Data collection

The following basic clinical data were collected: demographic characteristics (age and sex), interval from onset to admission, clinical manifestations (prodrome, initial symptoms, and primary clinical manifestations), immunotherapy latency, treatment methods, admission to the ICU, and complications (pulmonary infection, central hypoventilation, hypoproteinemia, and hyponatremia). We defined immunotherapy latency as the interval from onset to the initiation of immunotherapy. Pulmonary infection was diagnosed by respiratory physicians according to relevant criteria.

The results of laboratory tests and imaging examinations were also extracted from medical records and electronic databases for review. Abnormal cranial magnetic resonance imaging (MRI) results were confirmed as consistent with neuroinflammation (18), including T2-weighted fluid-attenuated inversion recovery (FLAIR) hyperintensities on one or both sides of the mesial temporal lobes (hippocampus and amygdala). We determined the CSF pressure, white blood cell (WBC) count, lymphocyte ratios, total protein, and autoantibody results from serum and CSF samples based on the first lumbar puncture after admission. Immunoglobulin anti-$GABA_BR$ antibodies in the CSF were detected by cell-based assays (CBAs) in all patients. To prevent potential impacts on peripheral immune cell counts, we excluded patients with systemic infections or who underwent immunotherapy. In addition, we obtained the total WBC count, platelet count (PLT), absolute neutrophil count (ANC), absolute lymphocyte count (ALC) and absolute monocyte count (AMC) from the patient's first full blood analysis within 24 h of admission. The NLR was calculated as the ratio of ANC to ALC; the MLR and PLR were calculated in a similar manner. In this study, all patients received examinations that screened for tumors, including computed tomography (CT) scans of the thorax, and ultrasounds of the abdomen, pelvic area and reproductive regions during hospitalization.

Disease prognosis evaluation

The mRS was used to evaluate the neurological function of the patient at the time of admission, in the first 4 weeks of treatment (19), and during the follow-up period. The mRS scores include 6 categories (20). Patients were divided into the mild or moderate group (0-2) and severe group (3-6) according to their mRS scores at admission.

Statistical analysis

Missing data were imputed using multiple imputation methods. The Shapiro–Wilk test was applied to assess the distribution of data. Continuous variables with a normal distribution are presented as the mean ± standard deviation. For data with a skewed distribution, the median (1^{st} quartile, 3^{rd} quartile) was utilized to describe their features, and Kruskal–Wallis tests were employed for comparisons. Categorical variables are presented as frequencies (proportions), and Fisher's exact tests were applied for comparisons. Parameters with $P< 0.05$ in the univariate analysis were included in the ordinal logistic regression analysis to estimate the effect of treatment on the full range of the mRS scores. Tolerance and the variance inflation factor (VIF) were used to examine multicollinearity. ALC, CSF WBC count, and the PLR were included in binary logistic regression analysis with the outcome of tumor presentation; the presence of psychiatric behavior was ignored due to its extreme effect. Ordinal logistic regression was performed to investigate risk factors. Model 1 included mRS score at admission, hospital stay, psychiatric behavior, tumor presentation, central hypoventilation, pneumonia, hypoproteinemia and mRS score after immunotherapy. To further test the stability of the model, age and sex were included as covariates in Model 2, while hospital stay was not adjusted for. A P value < 0.05 was considered statistically significant. Descriptive analysis of the baseline and univariate analyses was performed using IBM SPSS version 25.0 for Windows.

Results

Clinical characteristics

In total, 42 potential patients were screened; of these, 34 met the inclusion criteria. The baseline clinical features of the study population are shown in Table 1. The median age of the patients was 62.5 (15-82) years old, and the sample included 26 (76.50%) men and 8 (23.50%) women. All patients had an acute or subacute onset, and the median time from onset to admission was 10 (1-180) days. 13(38.2%) exhibited prodromal symptoms, with 6 having a fever and 5 having headaches. Other prodromal symptoms included dizziness, fatigue, vomiting, diarrhea, and sore throat. The most common initial symptom was seizure (26/34, 76.5%). 3 (8.8%) initially experienced behavioral changes, and 3 (8.8%) patients presented with memory deficits as the

10.3389/fimmu.2022.918064

TABLE 1 Patient characteristics in the cancer and noncancer groups.

	Total	Cancer	Noncancer	P value	OR (95% CI), P
Variable	34	12	22	–	
Males, n (%)	26	10	16	0.548	
Age, median (IQR), years	62.5 (54.75-65.25)	64.5 (59.25-66.5)	59 (48-64.5)	0.784	
mRS score at admission, mild or moderate, n (%)	17	4	13	0.102	
Symptoms					
Psychiatric behavior, n (%)	23	12	11	0.003*	
Seizure, n (%)	30	11	19	1.000	
Consciousness declination, n (%)	12	7	5	0.062	
Cognitive dysfunction, n (%)	23	8	15	1.000	
Movement disorder, n (%)	5	2	3	1.000	
Speech dysfunction, n (%)	4	0	4	0.273	
Sleep disorder, n (%)	10	5	5	0.271	
Autonomic dysfunction, n (%)	4	2	2	0.602	
Prodromal symptoms, n (%)	11	6	5	0.138	
ICU admission, n (%)	12	5	7	0.711	
Central hypoventilation, n (%)	5	3	2	0.319	
Pulmonary infection, n (%)	17	7	10	0.721	
Hypoproteinemia, n (%)	10	5	5	0.271	
Hyponatremia, n (%)	8	6	2	0.013*	9.268 (1.054-81.502), 0.045
Abnormal brain MRI, n (%)	19	7	12	1.000	
CSF tests					
CSF pressure, median (IQR)	165.00 (129.52-192.50)	155 (140.00-187.50)	170.00 (126.07-202.50)	0.709	
WBC count (n×10^6), median (IQR)	8.00 (2.00-26.50)	22.0 (8.5-37.00)	3 (2-13.5)	0.004*	
CSF protein, n × g/L	376.70 (267.58-550.58)	445.55 (367.7-620.625)	343.85 (234.75-466.75)	0.102	
Blood tests					
WBC count, median (IQR), n × 10^9/L	8.57 (6.80-10.27)	7.90 (5.55-10.83)	8.67 (7.08-10.22)	0.466	
Platelets, median (IQR)	216.00 (178.25-261.50)	206.00 (154.25-279.50)	216 (183.5-260.25)	0.817	
Neutrophils, median (IQR)	5.65 (4.56-7.62)	5.65 (3.50-8.95)	5.64 (4.78-7.14)	0.986	
Lymphocytes, median (IQR)	1.46 (0.85-1.88)	0.83 (0.60-1.48)	1.68 (1.30-2.29)	0.001*	0.063 (0.006-0.639), 0.019
Monocytes, median (IQR)	0.58 (0.46-0.75)	0.49 (0.40-0.99)	0.64 (0.48-0.75)	0.345	
NLR	3.50 (2.45-8.90)	6.67 (2.95-11.76)	3.17 (2.28-5.81)	0.080	
MLR	0.47 (0.28-0.62)	0.60 (0.36-0.75)	0.37 (0.26-0.59)	0.110	
PLR	127.60 (88.55-214.68)	217.94 (124.01-312.78)	120.86 (82.07-156.54)	0.018*	
mRS score after immunotherapy, median (IQR)	1 (1-2)	2 (1.25-2.00)	1 (0.75-2.00)	0.033*	
Hospital stay, median (IQR), days	62.5 (54.75-65.25)	26.5 (17.25-32.25)	22 (13-32)	0.736	

* indicates P<0.05, OR, odds ratio; CI, confidence interval.

initial symptom. The primary clinical manifestations included seizure (n = 30, 88.2%), psychiatric behavior (n = 23, 67.6%), cognitive dysfunction (n = 23, 67.6%), Consciousness declination (n = 12, 35.3%), sleep disorders (n=10, 29.4%), movement disorders (n = 5, 14.7%), speech dysfunction (n = 4, 11.8%) and autonomic dysfunction (n = 4, 11.8%). Among these patients, 12 (35.3%) were admitted to the ICU for supportive treatment. Regarding complications, half of the patients in this cohort (n = 17, 50%) had pulmonary infections, followed by those with hypoproteinemia (n = 10, 29.4%), hyponatremia (n = 8, 23.5%) and central hypoventilation (n = 5, 14.7%).

Laboratory and imaging findings

The initial CSF, brain MRI and laboratory findings are presented in Table 1. Lumbar puncture was performed in 33 patients. The CSF intracranial pressure was higher than 180 mmH_2O in 10 (30.3%) patients, and the CSF WBC count was increased (> 5 × 10^6/L) in 18 (54.5%) patients. The CSF lymphocyte ratios and total protein were elevated in 27 (81.8%) and 9 (27.3%) patients, respectively. AE-related antibodies, including anti-NMDAR, $GABA_BR$, LGI1, α-amino-3-hydroxyl-5-methyl-4-isooxazolpropionic acid receptor (AMPAR1, AMPAR2), and contact protein-associated protein-

2 (CASPR2) antibodies, were detected in 18 serum samples and 33 CSF samples. A total of 33 patients were positive for anti-$GABA_BR$ antibodies in CSF and 17 patients were positive for anti-$GABA_BR$ antibodies in serum. 33 patients underwent a brain MRI. Of these, 18 (54.5%) exhibited increased signals on T2-weighted or FLAIR images, of which 14 (41.2%) were distributed in the limbic system: 7 patients had bilateral lesions, 6 patients had left-sided lesions, and 2 patients had right-sided lesions (1 patient showed lesions of the right medial temporal lobe and bilateral hippocampus).

Treatment and follow-up

All patients received first-line treatment, and the median time from onset of the disease to the initiation of immunotherapy was 12.5 (4-186) days. No patients received second-line treatment. In our center, selection of immunotherapy was based on consensus principles. In mild cases, a single first-line immunotherapy was the primary choice. For patients without contraindications, steroids were preferred; otherwise, IVIG was preferred. In patients positive for serum antibodies, PLEX was preferred. For patients with a poor response to monotherapy or severe cases, combined first-line immunotherapy was considered, such as steroids combined with IVIG and/or plasma exchange. First-line immunotherapy could be repeated according to the specific patient status. If patients did not respond well to first-line immunotherapy, second-line immunotherapy was initiated as soon as possible. 12 received steroids (1 g/d for 5 days) alone, 3 received IVIG (0.4 g/kg/d for 5 days) alone, and 1 received PLEX alone. In addition, 18 patients were administered combined first-line immunotherapy: 14 were administered steroids combined with IVIG, 2 were administered steroids combined with PLEX, and 2 were administered steroids combined with IVIG and PLEX. At follow-up, neurological function, relapse, presence of tumors, and mortality were evaluated. The median follow-up time was 22.5 months (0.1-63 months). 7 experienced relapse, with a median time from discharge to relapse of 187 (81-772) days. Additionally, 12 (35.3%) patients had lung cancer: 7 cases were diagnosed at admission, and 6 presented during follow-up. Among these patients with cancer, 6 were confirmed to have SCLC *via* pathological biopsy. All of the patients presented with neurologic symptoms that preceded the diagnosis of cancer. 14 died, with 7 deaths due to lung cancer.

Predictive factors for poor prognosis of patients with anti-$GABA_BR$ encephalitis

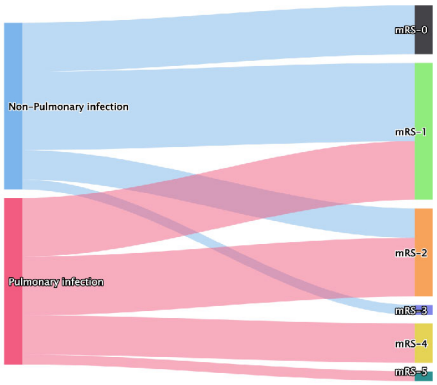
To explore factors related to prognosis of patients with anti-$GABA_BR$ encephalitis, we conducted ordinal regression analysis according to mRS scores (Table 2). Univariate analysis indicated baseline mRS scores (P=0.002), psychiatric behavior (P=0.009), hypoproteinemia (P=0.05), pulmonary infection (P=0.036), central hypoventilation (P=0.021), and accompanying tumors (P=0.033) to be associated with significant differences in mRS scores after first-line treatment. All of the above factors were included in the ordinal logistic regression model, and the results showed that pulmonary infection [odds ratio (OR)=17.444, 95% confidence interval (CI): 1.713-177.683, P=0.016] and baseline mRS scores (OR= 17.392, 95% CI: 2.237-135.098, P=0.006) were independent risk factors for failure of first-line treatments in patients with anti-$GABA_BR$ encephalitis (Figures 1, 2). Moreover, the adjusted ORs of pulmonary infection (OR=9.885, 95% CI: 1.106-88.323, P=0.040) and baseline mRS score (OR= 24.047, 95% CI: 3.294-175.739, P=0.002) were still significant when age and sex were included as covariates in the multiple regression model, further demonstrating the robust predictive value of pulmonary infection and baseline mRS score in anti-$GABA_BR$ encephalitis therapy.

Comparisons between the cancer and noncancer groups

To explore whether anti-$GABA_BR$ encephalitis interacts with cancer, we performed logistic analysis between the cancer and noncancer groups. Univariate analysis indicated significant differences between the group with cancer and the group without cancer with regard to psychiatric behavior (P=0.003), CSF WBC count (P=0.004), ALC (P=0.001), the PLR (P=0.018), and mRS scores (P=0.033) after first-line treatment. All factors with a P value < 0.05 were included in the multivariate logistic regression model. Due to the extreme distribution of psychiatric behavior (all patients in the cancer group had a psychiatric behavior), we performed multivariate logistic regression analysis excluding this variable; we found that ALC (OR: 0.063, 95% CI: 0.006-0.639, P=0.019) and hyponatremia (OR: 9.268, 95% CI: 1.054-81.502, p=0.045) were independent risk factors for anti-$GABA_BR$ encephalitis accompanied by lung cancer.

Discussion

In this study, we retrospectively analyzed the clinical features and risk factors for poor prognosis of patients with anti-$GABA_BR$ encephalitis who received first-line treatment. Moreover, we identified factors related to cases of anti-$GABA_BR$ encephalitis accompanied by cancer. We found that pulmonary infection and baseline mRS score may be crucial predictors of a poor prognosis in patients with anti-$GABA_BR$ encephalitis and that low ALC and hyponatremia at the time of admission may predict an underlying risk of developing cancer. However, the NLR, MLR and PLR had no predictive value in terms of the success of first-line treatment.

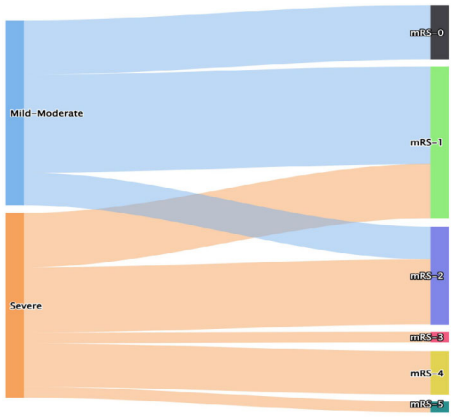


	mRS-0	mRS-1	mRS-2	mRS-3	mRS-4	mRS-5	Univariate P value	Multivariate P value
Non-Pulmonary infection	5	8	3	1	0	0	0.036*	0.040*
Pulmonary infection	6	6	0	0	4	1		

FIGURE 1
Univariate and multivariate analyses of pulmonary infection presentation and mRS score after immunotherapy. * indicates P<0.05.

Of the 34 patients, 26 were male (76.5%), and 8 were female. This result suggests that anti-$GABA_BR$ encephalitis is more common in males, which is consistent with previous research (6, 21). The median time from onset to admission was 10 days, which is shorter than the 4-week (2–104-week) duration described by Hoftberger B (6). Viral infection is a principal cause of AE (22). However, in our study, only 13 patients (38.2%) exhibited prodromal symptoms of infection, such as fever and headache, indicating that infection was not a trigger for onset in most of our patients.

The $GABA_BR$ is a G-protein-coupled receptor that belongs to the family of inhibitory synaptic proteins; this family plays an important role in neurotransmitter transmission and synaptic plasticity (23). $GABA_BRs$ reduce neuronal activity by inhibiting



	mRS-0	mRS-1	mRS-2	mRS-3	mRS-4	mRS-5	Univariate P value	Multivariate P value
Mild-Moderate	5	9	3	0	0	0	0.026*	0.002*
Severe	0	5	6	1	4	1		

FIGURE 2
Univariate and ordinal analyses of mRS scores at admission and after immunotherapy. * indicates P<0.05.

10.3389/fimmu.2022.918064

TABLE 2 Univariate and ordinal regression analysis of predictors for outcomes of anti-$GABA_BR$ encephalitis immunotherapy.

	Univariate analysis	Ordinal logistics regression			
	P value	Model 1** OR (95% CI)	P value	Model 2# OR (95% CI)	P value
Sex	0.975				
Age	0.548				
mRS score at admission	0.026*	17.392 (2.237-135.098)	0.006*	24.047 (3.294-175.739)	0.002*
Symptoms					
Psychiatric behavior	0.009*	1.388 (0.120-16.071)	0.793		
Seizures	0.398				
Consciousness declination	0.144				
Cognitive dysfunction	0.561				
Movement disorder	0.111				
Speech dysfunction	0.895				
Sleep disorder	0.152				
Autonomic dysfunction	0.082				
Prodromal symptoms	0.382				
Tumor presentation	0.033*	2.737 (0.424-17.655)	0.290		
ICU admission	0.144				
Central hypoventilation	0.021*	3.216 (0.179-57.858)	0.428		
Pulmonary infection	0.036*	17.444 (1.713-177.683)	0.016*	9.885 (1.106-88.323)	0.040*
Hypoproteinemia	0.050*	2.889 (0.271-19.317)	0.447		
Hyponatremia	0.177				
Abnormal MRI	0.512				
CSF tests					
CSF pressure	0.366				
WBC count	0.133				
CSF protein	0.327				
Blood tests					
WBC count	0.602				
Platelets	0.188				
Neutrophils	0.512				
Monocytes	0.152				
NLR	0.777				
MLR	0.487				
PLR	0.404				
Hospital stay	0.034*	1.068 (0.999-1.142)	0.055		

* indicates P<0.05, OR, odds ratio; CI, confidence interval.
**Model 1 included mRS score at admission, hospital stay, Psychiatric behavior, tumor presentation, central hypoventilation, pulmonary infection, and hypoproteinemia.
#Model 2 included all factors from Model 1 plus age and sex.

presynaptic calcium channels and thereby reducing calcium influx. $GABA_BRs$ are widely distributed in the CNS and highly localized in the cerebral cortex, hippocampus, cerebellum and thalamus (24). In our study, seizure was the initial symptom of AE in 26 patients (76.5%). In the whole course of the disease, seizure occurred in 30 patients, psychiatric behavior occurred in 23 patients, and cognitive dysfunction occurred in 23 patients, further confirming the above point. Previous studies (25, 26) have verified that anti-$GABA_BR$ encephalitis should be considered when patients are admitted to the hospital with characteristic manifestations of new-onset seizure or status epilepticus. Seizures may be the major or only clinical symptom of anti-$GABA_BR$ encephalitis, and approximately 3/4 of patients develop refractory epilepsy (27). In this study, 18 (54.5%) showed abnormal inflammation on the T2-weighted FLAIR, which is essentially consistent with the results of Dalmau J (28). Previous studies have found inflammatory changes when analyzing the CSF (29). Although an abnormal MRI is important for diagnosing anti-$GABA_BR$ encephalitis, lack of MRI abnormalities cannot rule out this disease. Our study further supports this view.

In this study, the baseline mRS score was a crucial predictor for response to first-line treatment. The mRS score was originally developed and validated to assess a patient's neurological outcome

after stroke (30). Later, researchers applied it to assess the severity and prognosis of AE, and, in most studies, patients with AE are divided into groups with a cutoff value of 2. Based on previous research, we assessed the mRS scores of patients with anti-$GABA_BR$ encephalitis at admission and after first-line treatment. We found that the higher the mRS score at admission was, the higher the mRS score after first-line treatment; that is, the more serious the condition was, the less effective the therapy. The severity of anti-$GABA_BR$ encephalitis fundamentally reflected the disease-induced inflammation, and the efficacy of treatment largely depended on disease severity, which is consistent with clinical practice. The findings further indicate that patients with a high baseline mRS score should be given more aggressive treatment (combined first-line immunotherapy). Additionally, these results suggest that doctors should give close attention to patients with a high baseline mRS score and communicate with relatives in advance about the possibility of a poor prognosis.

The results of the ordinal analysis showed pulmonary infection is an independent risk factor for failure to response to first-line treatment in patients with anti-$GABA_BR$ encephalitis. The incidence of pulmonary infection is high in these patients. According to a study by Jingfang Lin, more than two-thirds of anti-$GABA_BR$ encephalitis patients (18/28, 64.3%) have pneumonia, which is the major cause of short-term mortality (31). In our study, 50% of patients (17/34) developed a pulmonary infection during immunotherapy, and all of them had a worse response to first-line treatments. Additionally, pulmonary infection may be a crucial risk factor for poor prognosis in anti-NMDAR encephalitis (13, 32). The possible reasons are as follows. First, immune dysfunction results in low antibacterial activity of alveolar macrophages. Second, the administration of corticosteroids and immunosuppressants further reduce patient immune function. Third, central hypoventilation might aggravate the infection. Fourth, long-term bedridden status and intubation may increase the risk of pneumonia. In addition, some studies have found that the risk of pulmonary infection is related to the dose of corticosteroids and immunosuppressants: the higher the dose is, the higher the risk of infection (33, 34). All patients in our study received first-line treatment: 12 received steroids, 14 received steroids combined with IVIG, 2 received steroids combined with PLEX, and 2 received steroids combined with IVIG and PLEX. To treat this condition, patients are administered high doses of corticosteroids for long durations. Moreover, pulmonary infection in patients with immune dysfunction differs from that in patients with normal immune function because of the increased risks of opportunistic infections and severe bacterial infections. Therefore, close attention should be devoted to the occurrence of pulmonary infections in patients with anti-$GABA_BR$ encephalitis. In the present study, all patients were assessed for the risk of pneumonia before immunotherapy and regularly over the course of immunotherapy. In addition to a CT scan of the thorax, we also recommend examination of pathogens. If pneumonia developed, we immediately initiated anti-infective therapy. Mild pneumonia had little influence on immunotherapy; however, in cases of definite severe infection, IVIG was given priority, and the use of steroids was discontinued until the infection was controlled. Overall, appropriate prophylactic measures and aggressive therapy for pulmonary infection might help to improve patient prognosis.

Previous studies (28) have reported that approximately 50% of patients with anti-$GABA_BR$ encephalitis harbor an underlying cancer, particularly SCLC. The pathogenesis of cancer is related to abnormalities in the immune system. In this study, 12 (35.3%) were complicated with lung cancer, with 6 confirmed to have SCLC. The lower incidence of cancer in this study may be related to the short follow-up time. Once patients are diagnosed with anti-$GABA_BR$ encephalitis, cancer screening (especially for lung cancer) should be initiated as soon as possible. If the first cancer screening is negative, regular follow-up screening should be implemented. Additionally, screening is recommended at 3–6 months after discharge and then once a year for at least 4 years (35). In our univariate analysis, lung cancer was indicated to result in significant differences in mRS scores after first-line treatment (P=0.033). However, in the ordinal logistic regression model, the influence of lung cancer was not significant. In this study, our purpose was to find out the potential factors that affect the response to first-line treatment rather than survival in patients with anti-$GABA_BR$ encephalitis. Therefore, it is worthwhile to explore if the presence of lung cancer affecting the survival in a larger cohorts. This result might have been due to the small sample size of our study, which is a limitation. In the future, larger study cohorts are needed to confirm this hypothesis. Moreover, we found that a lower ALC might be a predictor of anti-$GABA_BR$ encephalitis accompanied by lung cancer. Normally, lymphocyte subpopulations maintain a dynamic balance to ensure stable immune function. The immune system, especially the strength of cellular immune function, is an important intrinsic protective factor against cancer occurrence. In recent years, many important studies have shown that the strength of the immune system is strongly related to the aggressiveness and prognosis of cancer. ALC represents the strength of the immune system and is an independent factor that influences cancer prognosis. In general, lymphocytes inhibit the proliferation of malignant cells in the body (36). In this study, patients with anti-$GABA_BR$ encephalitis and reduced ALC had a higher incidence of lung cancer, similar to the findings of a previous study. As the present study was retrospective in nature, lymphocyte subsets were not evaluated, and the specific mechanism underlying this relationship needs to be clarified.

Recent studies have found that the NLR, MLR and PLR, which are new biomarkers of inflammation (15), can stably reflect the body's inflammatory state and correlate with classic inflammatory mediators [such as levels of C-reactive protein (CRP), interleukin-6 (IL-6) and tumor necrosis factor alpha

(TNFa)]. The NLR serves as a biomarker of systemic inflammation in systemic lupus erythematosus (37), ulcerative colitis (38), and rheumatoid arthritis (39). Furthermore, some studies have suggested that the NLR is related to the severity, treatment and prognosis of CNS autoimmune diseases, such as multiple sclerosis (40) and AE (12, 13). In this study, we found no correlations of the NLR, MLR, or PLR with mRS scores after first-line treatment for anti-$GABA_BR$ encephalitis. James Broadley et al. (14) showed that a high NLR is associated with first-line treatment failure but that a high MLR was not associated with AE prognosis, consistent with our previous research on the MLR. We also utilized the PLR for the first time in the present study but found that it did not affect prognosis. Differences in the effects of the NLR on prognosis may be due to differences among study cohorts. Anti-$GABA_BR$ encephalitis is a type of AE mediated by neuronal cell surface antibodies, which are currently believed to be largely moderated by humoral immunity, but the exact pathological mechanisms of immune proliferation and transmission remain unclear (28). The NLR, MLR and PLR may be more closely related to encephalitis mediated by intracellular antibodies rather than neuronal cell surface antibodies; the former are considered to be cellular immune responses mediated mainly by T cells and pathology is characterized by a large number of infiltrating macrophages and microglia (14). The relationships between peripheral inflammatory indicators and AE prognosis require further multicenter studies with larger sample sizes.

In summary, our study had several limitations. First, this study had a retrospective design. Second, although we applied strict inclusion criteria, the sample size at our single center was still relatively small due to the low incidence of anti-$GABA_BR$ encephalitis in the general population. In the future, multicenter prospective studies are needed to confirm our results.

Conclusions

To date, studies have yet to identify the exact clinical characteristics that predict poor prognosis of patients with anti-$GABA_BR$ encephalitis. This study demonstrates that pulmonary infection and baseline mRS scores were independent risk factors for a poor prognosis of patients with anti-$GABA_BR$ encephalitis after first-line treatment. Moreover, ALC and hyponatremia might be potential biomarkers in the clinical evaluation of patients with anti-$GABA_BR$ encephalitis accompanied by lung cancer.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

Ethics statement

The experiments involving human participants were reviewed and approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University. The patients/participants provided written informed consent prior to participation in this study.

Author contributions

JD and YL designed the research. DX, JLv performed the research and data analysis. MT and JHL collected the data. JD wrote the paper; and JD, DX, JLv, TW and JHL critically revised the paper. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

References

1. Dalmau JE, Lancaster E, Martinez-Hernandez E, Rosenfeld MR, Balice-Gordon R. Clinical experience and laboratory investigations in patients with anti-NMDAR encephalitis. *Lancet Neurol* (2011) 10(1):63–74. doi: 10.1016/S1474-4422(10)70253-

2. Lancaster E, Martinez-Hernandez E, Dalmau J. Encephalitis and antibodies to synaptic and neuronal cell surface proteins. *Neurology* (2011) 77:179–89. doi: 10.1212/WNL.0b013e318224afde

3. Lancaster E, Lai M, Peng X, Hughes E, Constantinescu R, Raizer J, et al. Antibodies to the GABA(B) receptor in limbic encephalitis with seizures: Case series and characterisation of the antigen. *Lancet Neurol* (2010) 9:67–76. doi: 10.1016/S1474-4422(09)70324-2

4. Graus F, Titulaer MJ, Balu R, Benseler S, Bien CG, Cellucci T, et al. A clinical approach to diagnosis of autoimmune encephalitis. *Lancet Neurol* (2016) 15:391–404. doi: 10.1016/S1474-4422(15)00401-9

5. Dalmau J, Rosenfeld MR. Autoimmune encephalitis update. *Neuro Oncol* (2014) 16:771–8. doi: 10.1093/neuonc/nou030

6. Hoftberger R, Titulaer MJ, Sabater L, Dome B, Rózsás A, Hegedus B, et al. Encephalitis and GABAB receptor antibodies: Novel findings in a new case series of 20. *Neurology* (2013) 81:1500–6. doi: 10.1212/WNL.0b013e3182a9585f

7. Leypoldt F, Armangue T, Dalmau J. Autoimmune encephalopathies. *Ann N Y Acad Sci* (2015) 1338:94–114. doi: 10.1111/nyas.12553

8. Shin YW, Lee ST, Park KI, Jung KH, Jung KY, Lee SK, et al. Treatment strategies for autoimmune encephalitis. *Ther Adv Neurol Disorder* (2017) 11:1756285617722347. doi: 10.1177/1756285617722347

9. Titulaer MJ, McCracken L, Gabilondo I, Armangué T, Glaser C, Iizuka T, et al. Treatment and prognostic factors for long-term outcome in patients with anti-NMDA receptor encephalitis: An observational cohort study. *Lancet Neurol* (2013) 12:157–65. doi: 10.1016/S1474-4422(12)70310-1

10. Wen X, Wang B, Wang C, Han C, Guo S. A retrospective study of patients with GABABR encephalitis: Therapy, disease activity and prognostic factors. *Neuropsychiatric Disease and Treatment* (2021) 17:99–110. doi: 10.2147/NDT.S289942

11. Jeffery OJ, Lennon VA, Pittock SJ, Gregory JK, Britton JW, McKeon A. GABAB receptor autoantibody frequency in service serologic evaluation. *Neurology* (2013) 81:882–7. doi: 10.1212/WNL.0b013e3182a35271

12. Qiu X, Zhang H, Li D, Wang J, Jiang Z, Zhou Y, et al. Analysis of clinical characteristics and poor prognostic predictors in patients with an initial diagnosis of autoimmune encephalitis. *Front Immunol* (2019) 10:1286. doi: 10.3389/fimmu.2019.01286

13. Zhang X, Wang C, Zhu W, Wang B, Liang H, Guo S. Factors affecting the response to first-line treatments in patients with anti-N-Methyl-DAspartate receptor encephalitis. *J Clin Neurol* (2019) 15(3):369–75. doi: 10.3988/jcn.2019.15.3.369

14. Broadley J, Wesselingh. R, Seneviratne U, Kyndt C, Beech P, Buzzard K, et al. Peripheral immune cell ratios and clinical outcomes in seropositive autoimmune encephalitis: A study by the Australian autoimmune encephalitis consortium. *Front Immunol* (2021) 11:597858. doi: 10.3389/fimmu.2020.597858

15. Ding N, Pang Z, Shen H, Ni Y, Du J, Liu Q. The prognostic value of PLR in lung cancer, a meta-analysis based on results from a Large consecutive cohort. *Sci Rep* (2016) 6:34823. doi: 10.1038/srep34823

16. Wei Y, Feng J, Ma J, Chen D, Chen J.. Neutrophi/lymphocyte,platelet/lymphocyte and monocyte/lymphocyte ratios in patients with affective disorders. *J Affect Disord* (2022) 309:221–8. doi: 10.1016/j.jad.2022.04.092

17. Chen X, Wang Q, Li C. A retrospective analysis of hematologic parameters in patients with early diabetic kidney disease. *Clin Appl THROMB-HEM* (2022) 28:10760296221083681. doi: 10.1177/10760296221083681

18. Heine J, Pruss H, Bartsch T, Ploner CJ, Paul F, Finke C. Imaging of autoimmune encephalitis–relevance for clinical practice and hippocampal function. *Neuroscience* (2015) 309:68–83. doi: 10.1016/j.neuroscience.2015.05.037

19. Balu R, McCracken L, Lancaster E, Graus F, Dalmau J, Titulaer MJ. A score that predicts 1-year functional status in patients with anti-NMDA receptor encephalitis. *Neurology* (2019) 92(3):e244–e52. doi: 10.1212/WNL.0000000000006783

20. Lim JA, Lee ST, Jung KH, Kim S, Shin JW, Moon J, et al. Anti-nmethyl-d-aspartate receptor encephalitis in Korea: clinical features, treatment, and outcome. *J Clin Neurol* (2014) 10:157–61. doi: 10.3988/jcn.2014.10.2.157

21. Maureille A, Fenouil T, Joubert B, Picard G, Rogemond V, Pinto AL, et al. Isolated seizures are a common early feature of paraneoplastic anti-GABA(B) receptor encephalitis. *J Neurol* (2019) 266(1):195–206. doi: 10.1007/s00415-018-9132-0

22. Zhao MM. The relationship between anti-N-methyl-Daspartate receptor encephalitis and viral encephalitis. *Foreign Med Sci (Section of Pediatrics)* (2016) 43 (6):453–6. doi: 10.1111/jog.14984

23. Collingridge GL, Isaac JT, Wang YT. Receptor tracking and synaptic plasticity. *Nat Rev Neurosci* (2004) 5:952–62. doi: 10.1038/nrn1556

24. Benarroch EE. GABA(B) receptors structure, functions, and clinical implications. *Neurology* (2012) 78(8):578–84. doi: 10.1212/WNL.0b013e318247cd03

25. Mckay JH, Dimberg EL, Lopez CA. A systematic review of gammaaminobutyric acid receptor type b autoimmunity. *Neurol Neurochir Pol* (2019) 53(1):1–7. doi: 10.5603/PJNNS.a2018.0005

26. Guan HZ, Ren HT, Yang XZ, Lu Q, Peng B, Zhu YC, et al. Limbic encephalitis associated with anti-γ-aminobutyric acid b receptor antibodies: A case series from China. *Chin Med J (Engl)* (2015) 128(22):3023–8. doi: 10.4103/0366-6999.168989

27. Huang Q, Ma M, Wei X, Liao Y, Qi H, Wu Y, et al. Characteristics of seizure and antiepileptic drug utilization in outpatients with autoimmune encephalitis. *Front Neurol* (2019) 9:1136. doi: 10.3389/fneur.2018.01136

28. Dalmau J, Graus F. Antibody-mediated encephalitis. *N Engl J Med* (2018) 378(9):840–51. doi: 10.1056/NEJMra1708712

29. Ehling P, Melzer N, Budde T, Meuth SG. CD8 + T cell-medited neuronal dysfunction and degeneration in limbic encephalitist. *Front Neurol* (2015) 6:163. doi: 10.3389/fneur.2015.00163

30. van Swieten JC, Koudstaal PJ, Visser MC, Schouten HJ, van Gijn J. Interobserver agreement for the assessment of handicap in stroke patients. *Stroke* (1988) 19(5):604–7. doi: 10.1161/01.STR.19.5.604

31. Lin. J, Li C, Li A, Liu X, Wang R, Chen C, et al. Encephalitis with antibodies against the GABAB receptor: High mortality and risk factors. *Front Neurol* (2019) 10:1030. doi: 10.3389/fneur.2019.01030

32. Chi X, Wang W. Risk factors for mortality in patients with anti-NMDA receptor encephalitis *Acta Neurologica Scandinavica* (2017) 136(4):298–304. doi: 10.1111/ane.12723

33. Braga BP, Prieto – Gonzalez S, Hernández-Rodríguez J.. Pneumocyst is jirovecii pneumonia prophylaxis in immunocompromised patients with systemic autoimmune diseases. *Medicina Clinica* (2019) 152(12):502–7. doi: 10.1016/j.medcli.2019.01.010

34. Long W, Cai F, Wang X, Zheng N, Wu R. High risk of activation of latent tuberculosis infection in rheumatic disease patients. *Infect Dis (Lond)* (2020) 52 (2):80–6. doi: 10.1080/23744235.2019.1682187

35. Lin J, Li C, Li A, et al. Encephalitis with antibodies against the GABA(B) receptor: high mortality and risk factors. *Front Neurol* (2019) 10:1030. doi: 10.3389/fneur.2019.01030

36. Junttila MR, De Sauvage FJ. Influence of tumour micro-environment heterogeneity on therapeutic response. *Nature* (2013) 501(7467):346–54. doi: 10.1038/nature12626

37. Qin B, Ma N, Tang Q, Wei T, Yang M, Fu H, et al. Neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) were useful markers in assessment of inflammatory response and disease activity in SLE patients. *Mod Rheumatol* (2016) 26(3):372–6. doi: 10.3109/14397595.2015.1091136

38. Celikbilek M, Dogan S, Ozbakir O, Zararsiz G, Kucuk H, Gursoy S, et al. Neutrophil-lymphocyte ratio as a predictor of disease severity in ulcerative colitis. *J Clin Lab Anal* (2013) 27(1):72–6. doi: 10.1002/jcla.21564

39. Mercan R, Bitik B, Tufan A, Bozbulut UB, Atas N, Ozturk MA, et al. The association between Neutrophil/Lymphocyte ratio and disease activity in rheumatoid arthritis and ankylosing spondylitis. *J Clin Lab Anal* (2016) 30 (5):597–601. doi: 10.1002/jcla.21908

40. Hemond CC, Glanz BI, Bakshi R, Chitnis T, Healy BC. The neutrophil-tolymphocyte and monocyte-to-lymphocyte ratios are independently associated with neurological disability and brain atrophy in multiple sclerosis. *BMC Neurol* (2019) 19(1):23. doi: 10.1186/s12883-019-1245-2

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 06 September 2022
DOI 10.3389/fneur.2022.966428

OPEN ACCESS

EDITED BY
Barbara M. P. Willekens,
Antwerp University Hospital, Belgium

REVIEWED BY
Catherine Vignal Clermont,
Fondation Ophtalmologique Adolphe de Rothschild, France
Jinming Han,
Capital Medical University, China

*CORRESPONDENCE
Michael Levy
mlevy11@mgh.harvard.edu

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

RECEIVED 10 June 2022
ACCEPTED 02 August 2022
PUBLISHED 06 September 2022

CITATION
Delgado-Garcia G, Lapidus S, Talero R and Levy M (2022) The patient journey with NMOSD: From initial diagnosis to chronic condition. *Front. Neurol.* 13:966428. doi: 10.3389/fneur.2022.966428

The patient journey with NMOSD: From initial diagnosis to chronic condition

Guillermo Delgado-Garcia[1,2], Sheryl Lapidus[3], Rosa Talero[4] and Michael Levy[5]*

[1]Department of Clinical Neurosciences, Cumming School of Medicine, University of Calgary, Calgary, AB, Canada, [2]Centro de Investigacion y Desarrollo en Ciencias de la Salud (CIDICS), Universidad Autonoma de Nuevo Leon, Monterrey, Mexico, [3]Patient Advocacy, Horizon Therapeutics, Deerfield, IL, United States, [4]Patient at Neuromyelitis Optica Clinic and Research Laboratory, Massachusetts General Hospital, Boston, MA, United States, [5]Neuromyelitis Optica Clinic and Research Laboratory, Department of Neurology, Massachusetts General Hospital/Harvard Medical School, Boston, MA, United States

Objective: To better understand the patient experience with neuromyelitis optica spectrum disorder (NMOSD) through the course of the illness.

Background: NMOSD is a rare autoimmune disorder that causes recurrent inflammatory attacks of the optic nerve, spinal cord, and brain. Knowledge and awareness of NMOSD in the general medical community are often limited, resulting in potential delays in diagnosis and treatment.

Design/methods: We developed a comprehensive 101-question survey to understand the patient's perspective on their journey from initial presentation to present condition. The survey covered basic demographics, symptoms, medical tests used to reach a diagnosis, and the patient's psychosocial responses to their diagnosis. The survey included questions to determine internal consistency in responses. We shared the survey with members of the Neuromyelitis Optica (NMO) Clinic Facebook group and received responses from 151 patients. All data collected were self-reported and presented as summary statistics.

Results: The majority of survey responses were from patients who were female (83%) and White (76%), Asian (7%), or African American (7%). Initial symptoms of disease included fatigue, pain, stiffness/spasticity, bladder and bowel dysfunction, cognitive/emotional symptoms, and visual disturbances. Initial reactions to NMOSD diagnosis were frequently fear, anxiety, and/or depression. Mean (SD) time to diagnosis was 2.2 (3.2) years. First contact with a medical professional was felt to be not helpful or somewhat helpful for many patients (71%), in part due to uncertain diagnosis and/or treatment. However, once referred to specialists (primarily neurologists), the majority of patients (87%) reported finding a professional who could help. Tests leading to diagnosis included magnetic resonance imaging, lumbar puncture, and blood tests for autoantibodies including aquaporin-4 (AQP4) and myelin oligodendrocyte glycoprotein (MOG). While approximately 30% of patients still felt challenged for a variety of reasons, most patients reported that having a diagnosis and being under the care of a specialist contributed to a comprehensive plan with hope for their future.

Conclusions: The NMOSD patient journey frequently begins with anxiety, fear, and frustration. Finding the right specialist and identifying appropriate screening tests can lead to earlier diagnosis and progression toward better patient outcomes.

KEYWORDS

NMOSD, neuromyelitis optica spectrum disorder, patient journey, diagnosis, patient experience, patient perspectives

Introduction

Neuromyelitis optica spectrum disorder (NMOSD) is a rare and severe autoimmune disease characterized by inflammation of the optic nerve and spinal cord (1–3). This chronic and potentially debilitating condition is typically marked by multiple relapses that can result in progressive neurologic disabilities, blindness, and even death (1, 3–5). NMOSD has prevalence ranging from 0.5 to 10 per 100,000 in most populations, with considerable global and regional variation (5–8). African Americans are overrepresented in the US patient population (9). A recent survey of patients with NMOSD in North America reported a population who was White (53%), African American (24%), Hispanic (12%), and Asian (9%) (6).

NMOSD was initially considered to be a clinical subtype of multiple sclerosis (MS) as both disorders present with similar symptoms including optic neuritis, myelitis, and demyelination (1, 9–11). NMOSD generally manifests as a series of discrete attacks (1, 9). Relapses occur in 80%-90% of patients, frequently within 1 to 3 years after the initial episode (1, 9). Recovery after an attack often is partial, and the level of disability increases with each relapse, leading to impaired mobility or blindness (1, 9).

Initial symptoms of NMOSD include mild to severe paralysis and ocular pain with loss of vision (1, 9). Other symptoms include intractable hiccups, nausea and vomiting, hearing loss, cranial nerve dysfunctions, sleep abnormalities, narcolepsy, bladder and bowel dysfunction, and acute respiratory failure (1, 4, 12, 13). NMOSD and MS are difficult to distinguish in the early course of disease. The identification of autoantibodies to aquaporin-4 (AQP4-IgG) as highly specific markers of NMOSD has facilitated differential diagnosis (10). Approximately 80% of patients with NMOSD express detectable levels of AQP4-IgG; however, antibody titers by themselves do not seem to be predictive of disease course or outcome (14–16).

The diagnostic odyssey for a patient with NMOSD can be complicated because there is significant variability in clinical presentation and disease course over time (17). NMOSD is frequently misdiagnosed, especially in patients with clinical signs who are seronegative for established biomarkers such as AQP4-IgG and myelin oligodendrocyte glycoprotein autoantibodies (MOG-IgG) (3, 10, 17). Primary care providers and emergency departments, who are often the first points of health care contact, generally have limited or no experience diagnosing and/or treating patients with NMOSD (17–19).

To better understand the challenges and experiences of patients with NMOSD, we explored how patients navigate the early stages of their disease using a survey. The aims of this survey were to identify what patients perceive to be their challenges to diagnosis and treatment and to help health care providers better understand this journey from the patients' point of view.

Methods

We worked with rareLife Solutions, Inc. to develop a detailed survey to explore the patient's perspective on their initial diagnostic journey from early symptoms to diagnosis and treatment of NMOSD. The survey was shared with members of the Neuroimmunology Clinic (formerly NMO Clinic, Boston, MA, USA) private Facebook group. A pilot survey was administered to a group of 23 volunteers who self-identified as patients. Responses were assessed for completeness, consistency with known baseline values, and demographics for the NMOSD population. The responses obtained from the pilot survey were used to develop a final survey, which was made available in an online format to the full group of patients in the Neuroimmunology Clinic private Facebook group. Survey questions focused on patient population (baseline demographics), signs and symptoms of patients' first clinical events, their initial experiences with the health care system, the diagnostic process, and treatment options. We also focused on the psychological reactions that patients with NMOSD experienced as they were diagnosed with this rare disease. Questions were primarily multiple choice with additional opportunities for patient narratives through inclusion of 6 free-form questions. Survey responses were fielded through SurveyMonkey in a de-identified case report form, and results were collected in September 2020. All data collected were self-reported by the respondents, and the survey could only be completed one time. To participate in the survey, respondents had to agree and grant permission *via* an active response for their

data to be used in an aggregated and anonymized manner. Data were anonymized in accordance with General Data Protection Regulation and presented as summary statistics. When narrative responses were reported, any details that could be used to identify respondents were removed.

Results

Respondents were required to agree to the following statement before they could proceed with the survey: "Please be aware that we will be gathering and processing your responses in total and that while no individual information will be shared with anyone, your responses will be combined and analyzed with all other respondents. Most importantly, your responses will be held in strict confidence. If you are comfortable with that, please continue with the survey, by clicking the button below."

Patient responses obtained during the pilot survey indicated that patients understood the questions and were actively engaged with the project, as demonstrated by the following: (1) a large percentage of patients answered most, if not all, the questions; (2) patient responses were complete and consistent with known facts about NMOSD; and (3) answers were internally consistent with information provided in response to other related questions in the survey.

Of the 160 volunteers who participated in the final survey, 151 identified themselves as patients, and 9 were advocates and caregivers. Only data from self-identified patients are reported in this article. These data were presented in part as a poster for the 2021 annual meeting of the American Academy of Neurology (20).

Patient demographics and baseline physical condition

Respondents to this survey were predominantly female (83%), White (76%), and from the United States (71%) (Table 1), which is representative of the group in general. More than half had completed college or advanced degrees. Median age was 48 (<10 to >70) years (Figure 1) and mean age at disease onset was 40.3 years. Time from diagnosis to this survey was within 4 years for 66/123 (54%) respondents; an additional 40/123 (33%) were diagnosed between 5 and 9 years before this survey, and 15/123 (12%) were diagnosed between 10 and 19 years before this survey. Fifty-two patients reported problems with mobility (requiring a cane, walker, or wheelchair, or being homebound).

TABLE 1 Patient demographics and current level of mobility.

Characteristic	Responses, no. (%)
Age, median (range), y	48 (<10 to >70)
Sex, $n = 151$	
Female	126 (83%)
Male	18 (12%)
Other/NA	7 (5%)
Race, $n = 151$	
White	115 (76%)
Asian	11 (7%)
African American	10 (7%)
Native American	3 (2%)
Hawaiian/Pacific Islander	2 (1%)
Other/PNtS	10 (7%)
Ethnicity, $n = 146$	
Not Hispanic/Latino	125 (86%)
Hispanic/Latino	12 (8%)
PNtS or NA	9 (6%)
Level of education, $n = 151$	
Advanced degree	37 (24%)
Completed college	50 (33%)
Some college	34 (22%)
Completed high school	22 (15%)
Some high school	4 (3%)
PNtS	4 (3%)
Country/region of residence, $n = 147$	
USA	104 (71%)
Australia	11 (7%)
Canada	9 (6%)
EU	8 (5%)
UK	6 (4%)
Asia	6 (4%)
Other	3 (3%)
Level of mobility at time of survey, $n = 126$	
None	74 (59%)
Need a cane to get around	24 (19%)
Need a walker	11 (9%)
Need a wheelchair	12 (9%)
Confined to home	5 (4%)

NA, no answer; PNtS, prefer not to say.

Characteristics of first NMOSD attack

In all, 73% (110/151) of patients described their first attack as serious or worse, with 5% (7/151) reporting it as life-threatening (Figure 2). Eightythree percent (125/151) of respondents experienced pain, 81% (123/151) experienced fatigue, and 63% (95/151) experienced stiffness or spasticity (Figure 3A). Of the patients who reported an impact on their vision, 94% (88/94) experienced visual disturbances, 39% (37/95) experienced double vision, 71% (67/94) experienced loss of peripheral vision, and 61% (58/95) experienced loss of central vision (Figure 3B). Patients also reported other physical

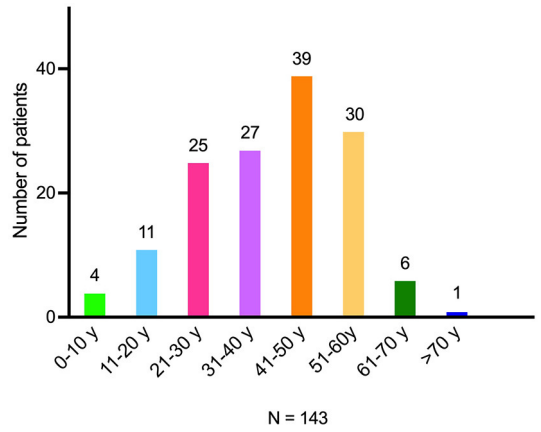


FIGURE 1
Age at disease onset for NMOSD. NMOSD indicates neuromyelitis optica spectrum disorder.

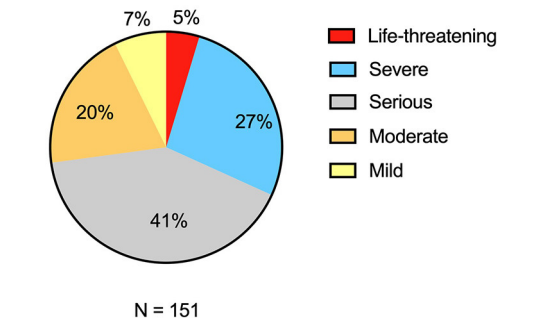


FIGURE 2
Severity of the first attack of NMOSD. Patients reported severity of first events to range from mild to life-threatening. NMOSD indicates neuromyelitis optica spectrum disorder.

symptoms including bladder problems 47% (71/151), bowel problems 39% (58/150), and sexual dysfunction 36% (54/148) (Figure 3C). Additionally, cognitive and emotional symptoms were reported by 59% (89/150) of patients and included brain fog, mood swings, and anxiety (Figure 3D).

This survey contained questions that afforded patients the opportunity to write narrative comments about various aspects of their diagnostic journey. Initial attacks of NMOSD were often described as painful and frightening (Supplementary Table 1). One patient described their initial experience as follows: *"Two weeks of severe cold that developed into flu symptoms with headache, weakness, and body aches. I was placed on an antibiotic. The headache worsened and I developed blurred vision and loss of vision in one eye. My antibiotic was changed. Two days later, I developed severe abdominal pain. While in the ER, the weakness progressed to paralysis from the chest down."*

First experience with health care system

Patients often described their the initial contact with the health care system using terms such as "scared," frustrated," and "bewildered" (Table 2). It was noted that 107 of 151 (71%) patients responded that their first contact with a medical professional was "not helpful" or only "somewhat helpful" in guiding them toward their next steps. Fewer than 10% of patients described their initial contact with a medical provider as "hopeful." Only 16 of 144 (11%) were diagnosed as having preliminary NMOSD. Initial treatments were prescribed for ~75% of patients and included prednisone/methylprednisolone, gabapentin, baclofen, azathioprine, or rituximab. Other initial treatments offered included antibiotics, pain medications, exercise, and a referral to a psychiatrist. Almost all (148/151) patients provided brief narrative acounts of their initial experiences, coping strategies, and emotional responses to the sudden challenges of their attack (Supplementary Table 2). One patient described their experience as follows: *"Initially, I felt scared and bewildered. No one understood what was going on. There was nothing to help me see better to start school, no treatment suggested to correct my vision[,] and no reason why it was happening. They were just unanswered questions. When the doctors couldn't figure out what was wrong and was happening, they accused me of faking and suggested a psychiatrist to my parents."*

Path toward a diagnosis and treatment

Time from the first onset of symptoms to a diagnosis of NMOSD ranged from 1 month (20%) to more than 10 years (9%) (Table 3). The mean (SD) time to diagnosis was 2.2 (3.2) years and the median time was 7 months. Many patients subsequently proceeded to seek additional help, and care often transitioned from a general practitioner to a specialist, who was a neurologist for 98% of patients. Over half of patients reported feeling relieved after meeting their NMOSD specialist. Approximately half of patients had to go to a major academic medical center to see their specialist. Travel and time away from home were frequently required for patients to see their specialist, but travel was rarely international. Clinical and laboratory tests used to confirm NMOSD included physical examination, blood tests, magnetic resonance imaging, and lumbar puncture (Table 4). After the first series of tests, 99 of 151 (66%) of patients had to undergo further extensive tests which often included additional imaging and radiology. Seventy-six (69%) of the 110 patients who reported being tested for AQP4-IgG; had a positive response, and 18 (32%) of the 56 patients who reported being tested for MOG-IgG had antibodies. Approximately two-thirds of patients reported that they were provided with the appropriate information to help them understand their diagnosis of NMOSD. Patients reported

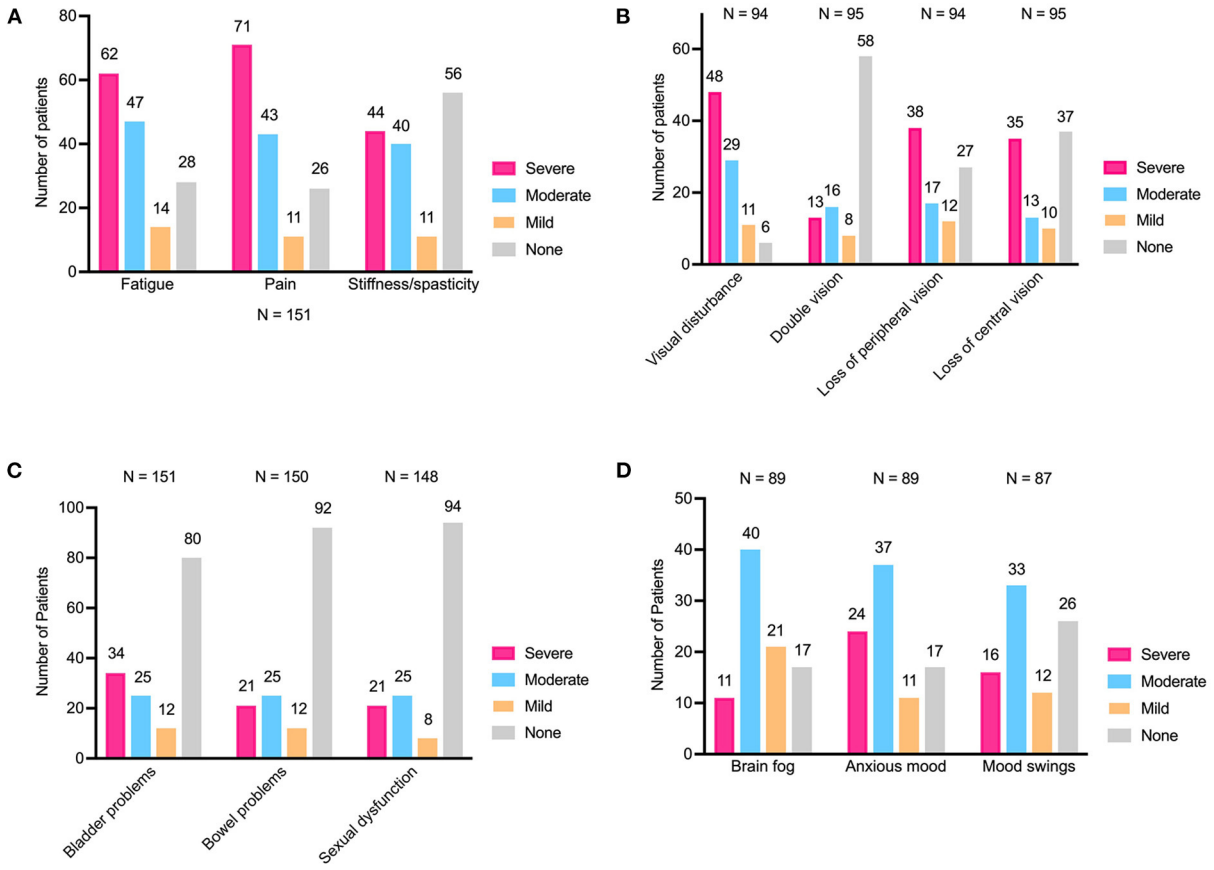


FIGURE 3
Signs and symptoms encountered during an initial attack of NMOSD. **(A)** Fatigue, pain, and stiffness/spasticity. **(B)** Visual disturbances, double vision, loss of peripheral vision, and loss of central vision. **(C)** Bladder and bowel problems, sexual dysfunction. **(D)** Brain fog, anxious mood, and mood swings. NMOSD indicates neuromyelitis optica spectrum disorder.

that they were taking a mean of 1.8 medications for NMOSD, and almost two-thirds of them were taking rituximab (Figure 4). Approximately half of the respondents had received at least one plasmapheresis treatment.

After meeting with an NMOSD specialist, 132 of 151 (87%) patients reported that they felt they had access to a professional who could guide them with treatment decisions (Table 5). In all, 106 of 150 (71%) respondents stated that they understood and could take advantage of their best treatment options, and 105 of 150 (70%) had a comprehensive care and recovery plan in place. After receiving their diagnosis and beginning to work with an NMOSD specialist, the majority of patients reported feeling relieved; however, others felt unhappy or lost. Upon diagnosis, patients had to confront their new reality of having NMOSD (Supplementary Table 3). *"It was hard being diagnosed. I was a month and a half away from getting married. I had always been healthy up until I wasn't. I had no real medical history. I was so scared of what the future would hold. Would I be blind? Would I be in a wheelchair? Would I be able to have children? Would I be dead in 5 years?"*

After a period of mourning their old lives and accepting the permanent losses, patients frequently began adjusting to a "new normal." When asked whether patients felt confident that they can now "live your best life," the responses were more positive than negative, although many patients still struggle with a life of limitations (Supplementary Table 4). *"I'm adjusting to my new normal. But I feel like every time something new goes numb, or something doesn't feel right, I have to wonder if it's an [NMOSD] attack. So, dealing with the unknown is a fear I live with every day."*

Discussion

Our survey provides information that describes the symptoms of the initial attack of NMOSD and patients' reactions

10.3389/fneur.2022.966428

TABLE 2 Patient first interaction with a health care provider.

Question	Responses, no. (%)
What type of health care provider did you first visit? $n = 144$	
ER doctor	49 (34%)
Primary care doctor	49 (34%)
Neurologist	26 (18%)
Ophthalmologist	13 (9%)
Other	7 (5%)
What was the first contact with a medical care provider like? How did you feel during, then after the appointment (check all that apply)?[a] $n = 150$	
Scared	86 (57%)
Frustrated	60 (40%)
Bewildered	56 (37%)
It will go away	40 (27%)
Alone	36 (24%)
Annoyed	30 (20%)
Impatient	20 (13%)
Relieved	15 (10%)
Hopeful	14 (9%)
Grateful	4 (3%)
Was there an initial diagnosis? $n = 151$	
Yes	81 (54%)
No	70 (46%)
What did they attribute your signs and symptoms to (check all that apply)?[a] $n = 144$	
Preliminary MS	50 (35%)
Stress	28 (19%)
Nonspecific neurologic issue	25 (17%)
Anxiety	20 (14%)
Autoimmune issue	20 (14%)
Preliminary NMOSD	16 (11%)
Other	35 (24%)
Was an initial treatment suggested? $n = 149$	
Yes	113 (76%)
No	36 (24%)
Was the first point of contact with a medical provider helpful in guiding you to what to do next? $n = 151$	
Very helpful	23 (15%)
Yes	21 (14%)
Somewhat helpful	41 (27%)
No	66 (44%)

[a]Because patients can select more than one option, the total percentage may exceed 100%. ER, emergency room; MS, multiple sclerosis; NMOSD, neuromyelitis optica spectrum disorder.

TABLE 3 Patient transition from a general practioner to a specialist.

Question	Responses, no. (%)
Time from symptom onset to NMOSD diagnosis, $n = 150$	
1 month	30 (20%)
2 months	10 (6%)
3 months	11 (7%)
4 months	13 (9%)
5 months	6 (4%)
6-11 months	13 (9%)
1 year	11 (7%)
2–5 years	31 (21%)
6–10 years	12 (8%)
>10 years	13 (9%)
What type of specialist did you see (check all that apply)?[a] $n = 136$	
Neurologist	133 (98%)
Immunologist	11 (8%)
Psychiatrist	6 (4%)
Other	5 (4%)
To see this specialist, did you have to go to a major academic medical center? $n = 134$	
Yes	76 (57%)
No	58 (43%)
Did this require significant travel and time away from home? $n = 76$	
Yes	44 (58%)
No	32 (42%)
Was the travel international? $n = 75$	
Yes	4 (5%)
No	71 (95%)
Did it present any language barriers? $n = 4$	
Yes	2 (50%)
No	2 (50%)

[a]Because patients can select more than one option, the total percentage may exceed 100%. NMOSD, neuromyelitis optica spectrum disorder.

to this experience while navigating the health care system to the point where a correct diagnosis was obtained. This survey is the first, to the best of our knowledge, that focuses on the patient's initial NMOSD attack and provides a substantial opportunity for patients to provide narrative responses regarding their feelings and reactions to their experience. Our patient population had essentially the same characteristics as those in other surveys of patients with NMOSD, suggesting that they are representative of the NMOSD populations who participate in surveys (17, 21–24). Unlike in previous surveys that used standardized assessment instruments, we intentionally designed ours to allow patients to express their feelings in a free form. Despite the subjective nature of our survey, our results were very similar to those from surveys that utilized standardized tools with the additional important benefit that we were able to obtain very personal insights into

10.3389/fneur.2022.966428

TABLE 4 Medical procedures/tests informing the diagnosis of NMOSD.

Question	Responses, no. (%)
What initial medical testing did you receive as part of your first visit (check all that apply)?[a] $n = 128$	
Blood tests	114 (89%)
MRI	112 (88%)
Physical exam	97 (76%)
Spinal tap	88 (69%)
X-rays	37 (29%)
Other	3 (2%)
Did you then undergo more extensive and invasive medical tests after the first series? $n = 151$	
Yes	99 (66%)
No	52 (34%)
If yes, what more extensive and invasive tests were performed (check all that apply)?[a] $n = 99$	
MRI	91 (92%)
Spinal tap	61 (62%)
Other imaging	45 (45%)
Radiology	34 (34%)
Other	9 (9%)
Did you undergo more extensive blood tests, including detailed screens for a range of autoantibodies? $n = 151$	
Yes	126 (83%)
No	12 (8%)
Not sure	13 (9%)
Which autoantibodies were you positive for (check all that apply)?[a] $n = 122$	
AQP-4	76 (62%)
MOG	18 (15%)
Not sure	24 (20%)
None	11 (9%)
Other	3 (2%)
As the patient, were you provided with the appropriate information to better understand your diagnosis of NMOSD? $n = 149$	
Yes	92 (62%)
No	57 (38%)
Once you received a diagnosis of NMOSD, did you wonder about how your disease would progress? $n = 151$	
Yes	144 (95%)
No	7 (5%)
What questions did you have (check all that apply)?[a] $n = 151$	
What will my future look like?	135 (89%)
Will I get better?	118 (78%)
Will I get back to feeling normal?	120 (79%)
If not, what will be my new normal be like?	110 (73%)

[a]Because patients can select more than one option, the total percentage may exceed 100%. AQP-4, aquaporin-4; MOG, myelin oligodendrocyte glycoprotein; MRI, magnetic resonance imaging; NMOSD, neuromyelitis optica spectrum disorder.

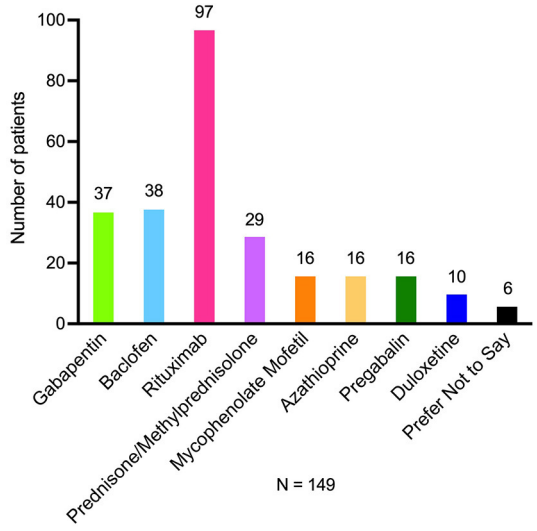


FIGURE 4
Current medications being used for treatment of NMOSD symptoms. One hundred forty-nine patients reported taking 1.8 medications each (mean) for NMOSD symptoms. NMOSD indicates neuromyelitis optica spectrum disorder.

patients' feelings and psychological state as they navigated their path through diagnosis and treatment (17, 21–24). In future surveys, it would be of interest to go even deeper into patient experiences to explore issues such as how regional differences affect their journey and how NMOSD has affected their ability to work and interact in society.

We believe that the NMO Clinic private Facebook community was highly motivated to share their journeys, as indicated by the number of patients completing the long and detailed survey. A large percentage of patients provided thoughtful narrative answers where appropriate. We believe adding questions that could elicit narrative responses enabled the patients to delve more deeply into questions about their quality of life and emotional experiences. For example, 148 out of 151 patients (98%) responded to questions about their coping strategies and emotional reactions to their diagnostic experience.

Patients' descriptions of their first attack of NMOSD and their contact with medical professionals clearly demonstrate how distressing the process can be. Patients describe fear, frustration, and disappointment. Patients describe how they were often confronted with sudden, distressing, and painful attacks of NMOSD with relatively little support or understanding from the medical community, especially emergency departments, primary care physicians, and neurologists, due to lack of knowledge of NMOSD (19). Increased understanding of NMOSD by physicians can help preserve vision and avoid permanent disability as well as help patients transition more efficiently to the right specialists (19). Finding the right specialist and identifying appropriate

TABLE 5 Identification of treatment options after a definitive diagnosis.

Question	Responses, no. (%)
How did you feel after meeting your NMOSD specialist? $n = 111$	
Relieved	86 (77%)
Unhappy and lost	16 (14%)
Other	9 (8%)
Do you feel like a comprehensive care and recovery plan is in place? $n = 150$	
Yes	105 (70%)
No	45 (30%)
Based on the details of my specific situation, do I feel that I understand and can take advantage of my best options? $n = 150$	
Yes	106 (71%)
No	14 (9%)
Not sure	30 (20%)
Do you feel like you know, and have access to, the professional who will guide/help you in making these decisions? $n = 151$	
Yes	132 (87%)
No	19 (13%)
If not, why do you feel that you do not know and/or have access to this professional? [Free-form answer] $n = 17$	NMOSD specialist is too far away No expert doctor Months to get an appointment Diagnostic issues Public health care limitations

NMOSD, neuromyelitis optica spectrum disorder.

screening tests can lead to an earlier correct diagnosis and faster progress toward better treatment and outcomes (10, 19).

Understanding the patient journey can yield important insights that could have a beneficial impact on patient care. This Facebook group and other social media networks like PatientsLikeMe provide access to many patients who have NMOSD and should be utilized to expand awareness to a broader patient and physician population (23). Patient responses to this survey provided detailed insights into the challenges that they encountered as they tried to find the best path forward in their new life. Utilizing patient narratives in publications can help clinicians empathize with the experiences that are often so frightening and disturbing to their patients (25–27). We believe that adding narrative questions within this survey may have allowed respondents to more freely express their feelings, helped them believe that they were being heard, and helped them to be more engaged in this survey.

The goal of this survey was to gather information on current patient experiences to help improve the patient journey in the future. Based on the responses of several patients, it appears that more education for the medical community could help raise awareness of NMOSD and could help physicians correctly diagnose the disease as early as possible. Many patients spend a long time with a misdiagnosis, which not only aggravates their medical condition but also subjects them to great emotional and financial hardship. An early and correct diagnosis with immediate treatment would be of great value in controlling the damage caused by NMOSD.

Limitations

As this survey was designed to elicit self-reported responses, individual experiences can be very subjective and less likely to provide quantitative data about specifics of NMOSD. Respondents may have very different perceptions of what "mild" or "serious" means with respect to disease or symptom severity. Moreover, they were often asked subjective questions about their feelings and perceptions. There are also challenges validating a patient's identity and diagnosis through a social media platform. There was no restriction on members of the group sharing the survey link externally, and no validation process was used to confirm that the respondents were in fact patients with NMOSD.

Data collected in this survey came primarily from patients in the United States (71%). Results cannot necessarily be generalized and may differ between regions and health care systems. A potential limitation of this study is that respondents were those who volunteered to complete this online survey. Therefore, individuals without access to the internet or who were unable to see or have the strength to participate were unlikely to complete the survey unless they had a friend or family member complete it with them. Although we queried the status of each respondent (patient, caregiver, or advocate), we did not expressly ask whether respondents were being aided by another person. No person was purposely excluded from the survey, and we did not specifically ask whether respondents were capable of completing the survey unaided.

Conclusions

Patients with NMOSD face a diagnostic journey that frequently begins with fear, confusion, and frustration. Initial contact with the medical community in the form of emergency departments or primary care physicians can often lead to misdiagnosis due to lack of knowledge about this rare disease. The survey indicates that when given the opportunity, patients are willing to share their experiences in their own words. As patients connect with specialists who provide the correct

diagnosis of NMOSD and a treatment plan is developed, patients frequently experience hope for an improved "new normal."

Data availability statement

The raw data supporting the conclusions of this article will be made available on reasonable request.

Ethics statement

Ethical review and approval was not required for the study on human participants in accordance with the local legislation and institutional requirements. The patients/participants provided their written informed consent to participate in this study.

Author contributions

GD-G and ML were involved in the development of the survey. All authors were involved in the interpretation of the results.

Funding

Medical writing services were supported by Horizon Therapeutics.

Acknowledgments

The authors thank the participants of this survey for giving their time and volunteering their information. The authors would also like to thank rareLife solutions for developing the survey under their guidance and analyzing the results and Eliza A. Donovan, PharmD candidate, University of Southern California, for collating the responses and assisting with the analysis. Medical writing support was provided by Michele Kinrade, Ph.D., of rareLife solutions, Westport, CT, USA, and supported by Horizon Therapeutics.

Conflict of interest

GD-G has received research support from the Consejo Nacional de Ciencia y Tecnologia, Mexico. ML received consulting fees from Alexion (now Alexion, AstraZeneca Rare Disease), Genentech/Roche/Chugai, Sanofi, UCB Pharmaceuticals, and Viela Bio (since acquired by Horizon Therapeutics). SL is an employee of Horizon Therapeutics and holds stock in the company. RT is a patient participating in a clinical trial of ravulizumab sponsored by Alexion, AstraZeneca Rare Disease.

The authors declare that this study received funding from Horizon Therapeutics for the preparation and submission of the manuscript. The funder was involved in the review of this article and the decision to submit it for publication.

Publisher's note

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.966428/full#supplementary-material

References

1. Wingerchuk DM, Lennon VA, Lucchinetti CF, Pittock SJ, Weinshenker BG. The spectrum of neuromyelitis optica. *Lancet Neurol.* (2007) 6:805–15. doi: 10.1016/S1474-4422(07)70216-8

2. Wingerchuk DM, Fujihara K, Palace J, Berthele A, Levy M, Kim HJ, et al. Long-term safety and efficacy of eculizumab in aquaporin-4 IgG-positive NMOSD. *Ann Neurol.* (2021) 89:1088–98. doi: 10.1002/ana.26049

3. Wingerchuk DM, Banwell B, Bennett JL, Cabre P, Carroll W, Chitnis T, et al. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. *Neurology.* (2015) 85:177–89. doi: 10.1212/WNL.0000000000001729

4. Borisow N, Mori M, Kuwabara S, Scheel M, Paul F. Diagnosis and treatment of NMO spectrum disorder and MOG-encephalomyelitis. *Front Neurol.* (2018) 9:888. doi: 10.3389/fneur.2018.00888

5. Tenembaum S, Yeh EA, Guthy-Jackson Foundation International Clinical Consortium. Pediatric NMOSD: a review and position statement on approach to work-up and diagnosis. *Front Pediatr.* (2020) 8:339. doi: 10.3389/fped.2020.00339

6. Cook LJ, Rose JW, Alvey JS, Jolley AM, Kuhn R, Marron B, et al. Collaborative international research in clinical and longitudinal experience study in NMOSD. *Neurol Neuroimmunol Neuroinflamm.* (2019) 6:e583. doi: 10.1212/NXI.0000000000000583

7. Pandit L, Asgari N, Apiwattanakul M, Palace J, Paul F, Leite MI, et al. Demographic and clinical features of neuromyelitis optica: a review. *Mult Scler.* (2015) 21:845–53. doi: 10.1177/1352458515572406

8. Bukhari W, Khalilidehkordi E, Mason DF, Barnett MH, Taylor BV, Fabis-Pedrini M, et al. NMOSD and MS prevalence in the indigenous populations of Australia and New Zealand. *J Neurol.* (2022) 269:836–45. doi: 10.1007/s00415-021-10665-9

9. Oh J, Levy M. Neuromyelitis optica: an antibody-mediated disorder of the central nervous system. *Neurol Res Int.* (2012) 2012:460825. doi: 10.1155/2012/460825

10. Jarius S, Wildemann B. Aquaporin-4 antibodies (NMO-IgG) as a serological marker of neuromyelitis optica: a critical review of the literature. *Brain Pathol.* (2013) 23:661–83. doi: 10.1111/bpa.12084

11. Dobson R, Giovannoni G. Multiple sclerosis—a review. *Eur J Neurol.* (2019) 26:27–40. doi: 10.1111/ene.13819

12. Misu T, Fujihara K, Nakashima I, Sato S, Itoyama Y. Intractable hiccup and nausea with periaqueductal lesions in neuromyelitis optica. *Neurology.* (2005) 65:1479–82. doi: 10.1212/01.wnl.0000183151.19351.82

13. Rosenthal JF, Hoffman BM, Tyor WR. CNS inflammatory demyelinating disorders: MS, NMOSD and MOG antibody associated disease. *J Investig Med.* (2020) 68:321–30. doi: 10.1136/jim-2019-001126

14. Tradtrantip L, Yeaman MR, Verkman AS. Cytoprotective IgG antibodies in sera from a subset of patients with AQP4-IgG seropositive neuromyelitis optica spectrum disorder. *Sci Rep.* (2021) 11:21962. doi: 10.1038/s41598-021-01294-3

15. Kessler RA, Mealy MA, Jimenez-Arango JA, Quan C, Paul F, Lopez R, et al. Anti-aquaporin-4 titer is not predictive of disease course in neuromyelitis optica spectrum disorder: a multicenter cohort study. *Mult Scler Relat Disord.* (2017) 17:198–201. doi: 10.1016/j.msard.2017.08.005

16. Dauby S, Dive D, Lutteri L, Andris C, Hansen I, Maquet P, et al. Comparative Study of AQP4-NMOSD, MOGAD and Seronegative NMOSD: a single-center belgian cohort. *Acta Neurol Belg.* (2021) doi: 10.1007/s13760-021-01712-3

17. Beekman J, Keisler A, Pedraza O, Haramura M, Gianella-Borradori A, Katz E, et al. Neuromyelitis optica spectrum disorder: patient experience and quality of life. *Neurol Neuroimmunol Neuroinflamm.* (2019) 6:e580. doi: 10.1212/NXI.0000000000000580

18. Starfield B, Shi L, Macinko J. Contribution of primary care to health systems and health. *Milbank Q.* (2005) 83:457–502. doi: 10.1111/j.1468-0009.2005.00409.x

19. Thomas K, Ocran C, Monterastelli A, Sadun AA, Cockerham KP. Bridging the gap between ophthalmology and emergency medicine in community-based emergency departments (EDs): a neuro-ophthalmology guide for ED practitioners. *Clin Pract.* (2021) 11:919–32. doi: 10.3390/clinpract11040106

20. Delgado-Garcia G, Lapidus S, Stefani-Hunyady D, Levy M. "Patient attitudes toward NMOSD diagnosis and treatment: final survey results (4233)," in *Poster session presented at: 73rd American Academy of Neurology Annual Meeting* (2021).

21. Ayzenberg I, Richter D, Henke E, Asseyer S, Paul F, Trebst C, et al. Pain, depression, and quality of life in neuromyelitis optica spectrum disorder: a cross-sectional study of 166 AQP4 antibody-seropositive patients. *Neurol Neuroimmunol Neuroinflamm.* (2021) 8(3). doi: 10.1212/NXI.0000000000000985

22. Mealy MA, Boscoe A, Caro J, Levy M. Assessment of patients with neuromyelitis optica spectrum disorder using the EQ-5D. *Int J MS Care.* (2019) 21:129–34. doi: 10.7224/1537-2073.2017-076

23. Eaneff S, Wang V, Hanger M, Levy M, Mealy MA, Brandt AU, et al. Patient perspectives on neuromyelitis optica spectrum disorders: data from the PatientsLikeMe online community. *Mult Scler Relat Disord.* (2017) 17:116–22. doi: 10.1016/j.msard.2017.07.014

24. Seok JM, Choi M, Cho EB, Lee HL, Kim BJ, Lee KH, et al. Fatigue in patients with neuromyelitis optica spectrum disorder and its impact on quality of life. *PLoS One.* (2017) 12:e0177230. doi: 10.1371/journal.pone.0177230

25. Charon R. The patient-physician relationship. narrative medicine: a model for empathy, reflection, profession, and trust. *JAMA.* (2001) 286:1897–902. doi: 10.1001/jama.286.15.1897

26. Fioretti C, Mazzocco K, Riva S, Oliveri S, Masiero M, Pravettoni G. Research studies on patients' illness experience using the narrative medicine approach: a systematic review. *BMJ Open.* (2016) 6:e011220. doi: 10.1136/bmjopen-2016-011220

27. Zaharias G. What is narrative-based medicine? narrative-based medicine 1. *Can Fam Physician.* (2018) 64:176–80.

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 29 September 2022
DOI 10.3389/fimmu.2022.966781

OPEN ACCESS

EDITED BY
Barbara M.P. Willekens,
Antwerp University Hospital, Belgium

REVIEWED BY
Benjamin Victor Ineichen,
University of Zurich, Switzerland
Honghao Wang,
Guangzhou First People's Hospital, China
Yaqing Shu,
Third Affiliated Hospital of Sun Yat-sen University, China

*CORRESPONDENCE
Yang-Tai Guan
yangtaiguan@sina.com

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Immunology

RECEIVED 11 June 2022
ACCEPTED 31 August 2022
PUBLISHED 29 September 2022

CITATION
Yao X-Y, Gao M-C, Bai S-W, Xie L, Song Y-Y, Ding J, Wu Y-F, Xue C-R, Hao Y, Zhang Y and Guan Y-T (2022) Enlarged perivascular spaces, neuroinflammation and neurological dysfunction in NMOSD patients. *Front. Immunol.* 13:966781. doi: 10.3389/fimmu.2022.966781

Enlarged perivascular spaces, neuroinflammation and neurological dysfunction in NMOSD patients

Xiao-Ying Yao[1], Mei-Chun Gao[1], Shu-Wei Bai[1], Li Xie[2], Ya-Ying Song[1], Jie Ding[1], Yi-Fan Wu[1], Chun-Ran Xue[1], Yong Hao[1], Ying Zhang[1] and Yang-Tai Guan[1]*

[1]Department of Neurology, Ren Ji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai, China, [2]Clinical Research Center, School of Medicine, Shanghai Jiao Tong University, Shanghai, China

Background and objectives: Cerebrospinal fluid (CSF) and interstitial fluid exchange along a brain-wide network of perivascular spaces (PVS) termed the 'glymphatic system'. The aquaporin-4 (AQP4) water channels abundantly expressed on astrocytic endfeet play a key role in the CSF circulation in the glymphatic system. Neuromyelitis optica spectrum disorder (NMOSD) is an inflammatory demyelinating autoimmune disease of the central nervous system (CNS) featured with a specific autoantibody directed against AQP4 in most of patients. Anti-AQP4 antibodies are likely resulting in the impairment of the brain glymphatic system and the enlargement of PVS in NMOSD patients. In the current study, we aimed to demonstrate the features of EPVS detected by MRI and its association with the CSF anti-AQP4 antibody titer, CNS inflammatory markers, and disease severity in NMOSD patients.

Methods: We conducted a retrospective review of a consecutive cohort of 110 patients with NMOSD who had brain MRI. We assessed the correlation of EPVS with markers of neuroinflammation, blood-brain barrier (BBB) function and severity of neurological dysfunction in patients. We used multivariate logistic regression analysis to determine the independent variables associated with disease severity.

Results: The median number of total-EPVS was 15.5 (IQR, 11-24.2) in NMOSD patients. The number of total-EPVS was significantly related to EDSS score after correcting for the effects of age and hypertension ($r=0.353$, $p<0.001$). The number of total-EPVS was also significantly associated with the titer of CSF anti-AQP4 antibody, the albumin rate (CSF/serum ratios of albumin), the CSF albumin, IgG and IgA levels. Logistic regression analysis showed that total-EPVS and serum albumin level were two independent factors to predict disease severity in NMOSD patients (OR=1.053, $p=0.028$; OR=0.858, $p=0.009$ respectively). Furthermore, ROC analysis achieved AUC of 0.736 (0.640-0.831, $p<0.001$) for total-EPVS to determine severe NMOSD (EDSS 4.5-9.5).

Discussion: In our cohort, we found a relationship between EPVS and neuroinflammation and BBB function in NMOSD. Moreover, EPVS might independently predict neurological dysfunction in patients with NMOSD.

KEYWORDS

neuromyelitis optica spectrum disorders (not in MeSH), glymphatic circulation, perivascular spaces, neuroinflammation, blood brain barrier (BBB)

Introduction

The brain was long believed to be devoid of a lymphatic vascular system. In 1970s, Cserr and colleagues has suggested a fluid-transport system in the brain; however, only late in 2012, this lymphatic transport system was designated the glial-associated lymphatic system, or the 'glymphatic system (1, 2). The glymphatic system facilitates movement of cerebrospinal fluid (CSF) and interstitial fluid (ISF) in the brain (3). Astrocytes play a key role in the glymphatic system. Astrocytes create with their vascular endfeet the perivascular spaces (PVS) that surround the cerebral vasculature. The PVS are utilized as 'highways' for fast transport of CSF into deep brain regions (4, 5). The movement of CSF into the parenchyma is facilitated by the aquaporin-4 (AQP4) water channels abundantly expressed on astrocytic endfeet (1, 6). The dysfunction of AQP4 will result in reduced CSF influx into the brain parenchyma and might cause the enlargement of PVS (7). However, there are still some controversies regarding the existence of the glymphatic system, its underlying driving force, and the convective versus diffusive nature of the flow (2).

It has been shown that the glymphatic system plays a role in neuroinflammation and immune responses within the brain (3). And the lymphatic impairment aggravates neuroinflammation probably by the accumulation or entrapment of waste and pro-inflammatory cytokines within the brain (8).

Studies demonstrated that the number and size of PVS may change in the course of inflammatory diseases such as multiple sclerosis (MS) (9–13). Histopathological studies have identified tissue perivascular spaces (EPVS) containing leucocyte infiltrates around chronic active inflammatory lesions of MS (14, 15). These evidence support the role of glymphatic system and pathogenic value of EPVS in neuroinflammatory process.

Neuromyelitis optica spectrum disorder (NMOSD) is an inflammatory demyelinating autoimmune disease of the central nervous system (CNS) mainly affected optic nerve, spinal cord and brain (16). The discovery of an NMO-specific autoantibody directed against AQP4 clearly identified NMOSD as a separate disease from MS (17). Patients can presented with recurrent optic neuritis, relapsing transverse myelitis, and some brainstem and encephalitic syndromes (18).

The autoantibodies that attack the glial AQP4 water channels are likely resulting in the impairment of the brain glymphatic system in NMOSD patients (6). We hypothesize that the gymphatic impairment will cause the enlargement of PVS and the aggravation of neuroinflammation; the latter in turn might lead to more severe neurological dysfunction in NMOSD patients.

So far, the clinical significance of the glymphatic system and EPVS in NMOSD is unknown. We suggested they might play key roles in the immune process of NMOSD. However, glymphatic system is difficult to learn *in situ* while EPVS can be easily visualized on magnetic resonance imaging (MRI). And EPVS could partially reflect the function of glymphatic system. Therefore, in the current study, we aimed to demonstrate the features of EPVS detected by MRI and its association with the CSF anti-AQP4 antibody, CNS inflammatory markers, blood-brain barrier (BBB) function, and disease severity in NMOSD patients.

Methods

Study population

The medical records of consecutive patients admitted to our center from April 2013 to January 2022 with the diagnosis of NMOSD were reviewed. Patients who fulfilled the diagnostic criteria established by Wingerchuk et al. in 2015 (19) were included in the current study. The following patients were excluded: (1) those without the head MR images; (2) serum anti-myelin oligodendrocyte glycoprotein (MOG) antibody positive and diagnosed with MOG antibody-associated disease (MOGAD).

Standard protocol approvals, registrations, and patient consents

The study was approved by the Ethics Committee of RenJi Hospital. Informed consent was obtained from all the included patients.

Clinical data collection

The clinical, laboratory, and radiology records of all the included patients were retrospectively reviewed. The clinical information collected including age, sex, age at onset, past medical history, total disease duration, time from last relapse, annualized relapse rate (ARR), Expanded Disability Status Scale (EDSS) score, serum status and titers of anti-AQP4 antibody, cerebrospinal fluid results (including white cell counts, albumin, IgG, IgM, IgA, albumin rate, IgG index and anti-AQP4 antibody titer), presentation of optic neuritis (ON), myelitis, and longitudinally extensive transverse myelitis (LETM). The patient was regarded as being in the acute phase of the attack if the time from last relapse was less than 30 days (20). EDSS score was checked at the time of MRI examination. It is used as a scale to evaluate the severity of neurological dysfunction of NMOSD patients. Mild NMOSD was defined with EDSS 0-4.0; while severe NMOSD was defined with EDSS 4.5-9.5. We define the relapse of NMOSD as a clinical exacerbation presenting with new or worsening symptoms accompanied with a change on the neurologic examination that correlated with a new or enhancing MRI lesion. The interval should be at least 30 days since the previous relapse (21). Serum anti-AQP4 and anti-MOG antibodies were tested in all patients using a cell-based assay. Albumin rate is the quotient of (CSF/Ser)*10^{-3}, which indicates the disruption of blood-brain barrier.

MR protocol

Patients underwent MRI according to a standardized protocol as part of routine clinical assessments. The protocol included T1- and T2-weighted, fluid-attenuated inversion recovery (FLAIR), axial trace diffusion-weighted imaging (DWI) with 2 b-values (0 and 1000), and apparent diffusion coefficient (ADC) sequences. All studies were performed on 3.0 T scanners. Images were 2D sequences. Sequences typically included 20–30 slices of 5-mm thickness. The imaging parameters were as follows: T1 (repetition time [TR] 194 ms; echo time [TE] 3.11 ms; field of view [FOV] 200×220; matrix 224×352; pixel 0.9×0.6); T2 (TR 5000 ms; TE 101 ms; FOV 207×220; matrix 224×352; pixel 0.9×0.6); FLAIR (TR 8000 ms; TE 102 ms; FOV 207×220; matrix 203×320; pixel 1.0×0.7); diffusion tensor imaging (TR 3260 ms; TE 50\83 ms; FOV 220×220; matrix 160×160; pixel 1.4×1.4).

Assessment of EPVS

Enlarged PVS (EPVS) are commonly seen in the centrum semiovale (CSO-EPVS), basal ganglia (BG-EPVS) and midbrain (MB-EPVS) (5).

EPVS were rated on axial T2-weighted MRI using a validated visual rating scale (22–24). EPVS were defined as ≤2 mm round or linear CSF isointense lesions (T2-hyperintense and T1/FLAIR hypointense with respect to brain) along the course of penetrating arteries (Figure 1). They were distinguished from lacunes by the latter's large size (>2 and ≤15mm) and surrounding rim of FLAIR hyperintensity (25).

EPVS were separated and rated in the BG, CSO and MB regions. They were counted in both sides on the brain slice showing the greatest extent of EPVS. A total number of EPVS (total-EPVS) was calculated as the sum of EPVS in the three regions.

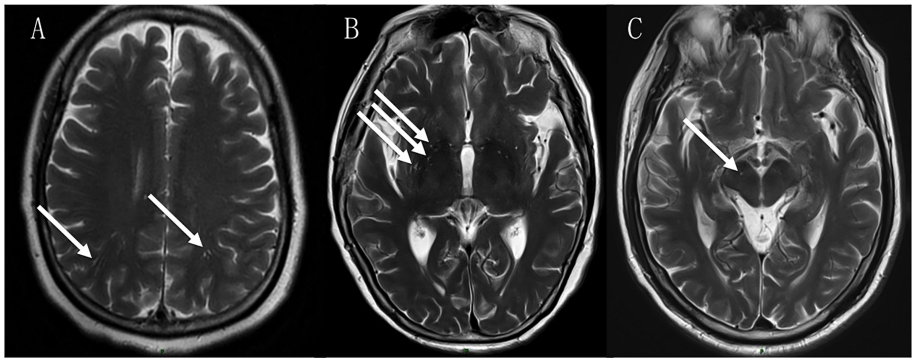


FIGURE 1
EPVS in the centrum semiovale (CSO-EPVS) **(A)**, basal ganglia (BG-EPVS) **(B)** and midbrain (MB-EPVS) **(C)** in NMOSD patients.

The following rating categories were used: 0 = no EPVS, 1 = 1 to 10 EPVS, 2 = 11 to 20 EPVS, 3 = 21 to 40 EPVS, and 4 = more than 40 EPVS. For the purpose of this analysis, EPVS were categorized into 0–2 vs 3–4 grades (mild vs severe EVPS).

Two trained neurologists evaluated MRI scans and when inconsistency existed between the reported results both neurologists discussed the case and reached an accord.

Statistical analysis

Data analysis was performed using SPSS version 21.0 (SPSS Inc., Chicago, Ill., USA). For the statistical analysis on the anti-AQP4 antibody titer X, a logarithmic value of $\log_2(X+1)$ was used. The Kolmogrov-Smirnov Z test was used to verify the normal distribution of the data. Categorical variables were summarized as counts (percentage) and continuous variables as the means (standard deviation, SD) or medians (interquartile ranges, IQR), if not distributed normally. Statistical comparisons between two groups were performed using Student's t-test or the Mann–Whitney U test for continuous variables, as well as the $\chi 2$ and Fisher's exact tests for categorical variables, as deemed appropriate. Correlations between continuous variables were assessed by Spearman's correlation coefficient. Logistic regression analysis was used to detect the independent factors associated with severe EPVS (EPVS grade 3-4) or severe NMOSD (EDSS 4.5-9.5). Receiver operating characteristic (ROC) curve and area under curve (AUC) were used to test the accuracy for EPVS to determine disease severity. A two-tailed probability value < 0.05 was considered significant. We did not correct for multiple comparisons.

Results

Clinical data of patients with NMOSD

As Table 1 shows, a total of 110 patients with NMOSD were enrolled. The mean age was 48.1 years (range 15-83 years); the female patients (n=93) had a dominant percentage of 84.5%; 67 (60.9%) patients were in the acute phase. The serum anti-AQP4 antibody was positive in 93 (84.5%) patients. CSF anti-AQP4 antibody was tested in 72 patients and 46 (63.9%) were positive. The annualized relapse rate (ARR) was 0.5 (IQR 0-1.1). The median score of EDSS was 3.5 (IQR, 2.5-6.1) at the time of MRI examination. Thirty patients were diagnosed comorbidities of autoimmune disorders (including Sjogren's syndrome, systemic lupus erythematosus, rheumatoid arthritis, mixed connective tissue disease, ankylosing spondylitis, and hashimoto thyroiditis). And the rates of hypertension, diabetes mellitus, and hyperlipidemia were 17.3%, 12.7% and 10.9% respectively.

The features of EPVS in patients with NMOSD

The median numbers of BG-EPVS, CSO-EPVS and MB-EPVS were 7.0 (IQR, 5.0-11.0), 8.0 (IQR, 4.0-12.2) and 0 (IQR, 0-1.0) respectively. The median number of total-EPVS was 15.5 (IQR, 11-24.2) in NMOSD patients.

We define EPVS grade 0-2 as mild category of EPVS and grade 3-4 as severe category of EPVS. There were 39 (35.5%) patients had severe EPVS. NMOSD patients with severe EPVS as compared to those with mild EVPS were older (56.1 ± 13.4 vs 43.6 ± 12.9, $p<0.001$) and had a higher percentage of hypertension (28.2% vs 11.3%, $p=0.025$). Severe EPVS patients tended to have higher levels of CSF albumin and CSF IgG, higher titers of CSF anti-AQP4 antibody, and a lower level of serum albumin. The median albumin rate was higher in patients with severe EPVS, which indicates a more severe blood-brain barrier disruption. Moreover, EDSS score was significantly higher in severe EPVS group as compared to mild EPVS group (3.0 vs 6.0, $p<0.001$). (Table 1) And there were more numbers of total-EPVS in patients with CSF anti-AQP4 antibody positive than negative (18.5 vs 13.0, $p=0.034$).

Sensitivity analyses were conducted by comparing lowest EPVS group (EVPS grade 0-1) with highest EPVS group (EPVS grade 4) and similar results were shown. (Supplementary materials: Table 1)

Correlation of EPVS with markers of neuroinflammation, blood-brain barrier function and severity of neurological dysfunction in NMOSD patients

The number of total-EPVS was significantly correlated with EDSS score ($r= 0.444$, $p<0.001$), which means NMOSD patients with more numbers of EPVS had more severe neurological dysfunction. (Figure 2A) The positive relationship between numbers of total-EPVS and EDSS scores still existed when stratified patients by age, serum anti-AQP4 status, disease phase, and first attack or relapse. (Supplementary materials: Table 2)

Previous studies have highlighted associations between EPVS severity and increasing age and vascular risk factors such as hypertension (26). Therefore, in order to exclude the influence of age and hypertension, partial correlation analysis was performed. The result revealed that the number of total-

EPVS was still significantly related to EDSS score after correcting for the effects of age and hypertension (r=0.353, p<0.001).

For different areas of EPVS, the numbers of CSO-, BG-, and MB- EPVS were respectively associated with EDSS score (r=0.352, p<0.001 for CSO; r=0.454, p<0.001 for BG; r=0.200, p=0.036 for MB). However, after correcting for the effects of age and hypertension, only the positive relationship between the numbers of CSO-, BG- EPVS and EDSS score remained significant (r=0.265, p=0.006 for CSO; r=0.385, p<0.001 for BG).

TABLE 1 Clinical data of patients with neuromyelitis optica spectrum disorders (NMOSD).

	NMOSD (N=110)			
	AllN=110	**EPVS grade 0-2N=71**	**EPVS grade 3-4N=39**	**P value**
Age (mean ± SD)	48.1 ± 14.4	43.6 ± 12.9	56.1 ± 13.4	<0.001
Sex (Female, %)	93 (84.5%)	60 (84.5%)	33 (84.6)	0.988
Age at onset (mean ± SD)	44.3 ± 15.7	39.8 ± 13.8	52.5 ± 15.9	<0.001
Acute phase (%)	67 (60.9%)	41 (57.7%)	26 (66.7%)	0.359
Total disease duration (median, IQR, months)	19.5 (2.8-68.2)	23.0 (3.0-72.0)	14.0 (2.0-67.0)	0.597
Time from last relapse (median, IQR, days)	15 (7-60)	21 (7-60)	15 (7-37)	0.338
Numbers of all attacks (median, IQR)	2 (1-3)	2 (1-3)	2 (1-4)	0.730
Annualized relapse rate (ARR) (median, IQR)	0.5 (0-1.1)	0.4 (0-1.0)	0.5 (0-2.0)	0.424
Co-morbidities				
Other Autoimmune Disorders (%)	30 (27.3%)	22 (31.0%)	8 (20.5%)	0.238
Systemic lupus erythematosus (%)	6 (5.5%)	5 (7.0%)	1 (2.6%)	0.420
Hypertension (%)	19 (17.3%)	8 (11.3%)	11 (28.2%)	0.025
Diabetes Mellitus (%)	14 (12.7%)	6 (8.5%)	8 (20.5%)	0.129
Hyperlipidemia (%)	12 (10.9%)	8 (11.3%)	4 (10.3%)	1.000
History of ischemic stroke or Transient ischemic attack (%)	2 (1.8%)	1 (1.4%)	1 (2.6%)	1.000
History of smoking (%)	6 (5.5%)	5 (7.0%)	1 (2.6%)	0.582
History of alcoholism (%)	1 (0.9%)	0 (0%)	1 (2.6%)	0.355
Clinical presentations				
ON [a] (%)	50 (45.5%)	31 (43.7%)	19 (48.7%)	0.610
Myelitis (%)	84 (76.4%)	53 (74.6%)	31 (79.5%)	0.568
LETM [b] (%)	70 (63.6%)	42 (59.2%)	28 (71.8%)	0.187
CSF[d] analysis				
White cell counts (median, IQR)	2.0 (0-7.0)	2.0 (0-7.2)	2.0 (0-6.5)	0.500
Albumin (mg/L, median, IQR)	344 (217-432)	322 (184-394)	398 (302-528)	0.023
Albumin rate [e] (median, IQR)	5.8 (4.0-8.9)	5.0 (3.7-7.8)	6.6 (5.4-10.1)	0.008
IgG (mg/L, median, IQR)	35.7 (24.1-57.0)	30.2 (22.6-54.8)	43.4 (29.8-75.7)	0.028
IgA (mg/L, median, IQR)	3.9 (2.6-7.2)	3.5 (1.9-7.3)	5.2 (3.0-7.2)	0.062
IgM (mg/L, median, IQR)	0.6 (0.3-1.2)	0.6 (0.2-1.0)	0.5 (0.3-1.3)	0.764
IgG index (median, IQR)	0.52 (0.47-0.59)	0.51 (0.47-0.58)	0.54 (0.47-0.59)	0.310
Anti-AQP4 antibody [f] (%)	46 (63.9%)	26 (56.5%)	20 (76.9%)	0.083
Titer of Anti-AQP4 (log_2) [g] (median, IQR)	1.0 (0-2.1)	1.0 (0-2.1)	2.1 (0.75-3.5)	0.033
Serum Anti-AQP4 antibody (%)	93 (84.5%)	58 (81.7%)	35 (89.7%)	0.264
Titer of serum Anti-AQP4 (log_2) [h] (median, IQR)	5.9 (0-8.3)	5.0 (0-8.3)	6.7 (3.5-8.3)	0.599
Serum Albumin (g/L, mean ± SD)	41.2 ± 4.3	42.0 ± 4.1	39.8 ± 4.5	0.01

(Continued)

TABLE 1 Continued

	NMOSD (N=110)			
	AllN=110	**EPVS grade 0-2N=71**	**EPVS grade 3-4N=39**	**P value**
EDSS [i] (median, IQR)	3.5 (2.5-6.1)	3.0 (2.0-4.0)	6.0 (3.0-8.0)	<0.001
EPVS counts [j]				
BG-EPVS [k]	7.0 (5.0-11.0)	6.0 (4.0-8.0)	11.0 (9.0-15.0)	<0.001
CSO-EPVS [l]	8.0 (4.0-12.2)	5.0 (3.0-8.0)	18.0 (13.0-22.0)	<0.001
MB-EPVS [m]	0 (0-1.0)	0 (0-1.0)	1.0 (0-2.0)	0.001
Total-EPVS	15.5 (11-24.2)	13 (8-15)	29 (24-37)	<0.001

[a]ON, optic neuritis.
[b]LETM, longitudinally extensive transverse myelitis.
[c]Brain syndromes of NMOSD refer to area postrema syndrome, brainstem syndrome, diencephalic syndrome and cerebral syndrome.
[d]CSF, cerebrospinal fluid.
[e]Albumin rate = Quotient (CSF/Ser)*10^{-3}.
[f]Anti-AQP4 antibody, anti-aquaporin-4 antibody.
[g]For the analysis of the CSF anti-AQP4 antibody titer X, a logarithmic value of $\log_2$ (X+1) was used.
[h]For the analysis of the serum anti-AQP4 antibody titer X, a logarithmic value of $\log_2$ (X+1) was used.
[i]EDSS, Expanded Disability Status Scale.
[j]EPVS, enlarged perivascular spaces.
[k]CSO, centrum semiovale.
[l]BG, basal ganglia.
[m]MB, Midbrain.

The number of total-EPVS was significantly associated with the titer of CSF, but not serum, anti-AQP4 antibody (r=0.275, p=0.02). (Figure 2B) For different areas of EPVS, only the number of CSO-EPVS was significantly related to the titer of CSF anti-AQP4 antibody (r=0.254, p=0.031).

Moreover, the number of total-EPVS was significantly correlated to the albumin rate (r=0.320, p=0.003), which indicates NMOSD patients with more numbers of EPVS were accompanied with more serious blood-brain barrier disruption. (Figure 2C) And both CSO-EPVS and BG-EPVS were related to the albumin rate (r=0.239, p=0.028 for CSO-EPVS; r=0.396, p<0.001 for BG-EPVS).

Furthermore, total-EPVS was significantly associated with CSF albumin, IgG and IgA levels (r=0.328, p=0.002 for CSF albumin; r=0.275, p=0.009 for CSF IgG; r=0.250, p=0.019 for CSF IgA),

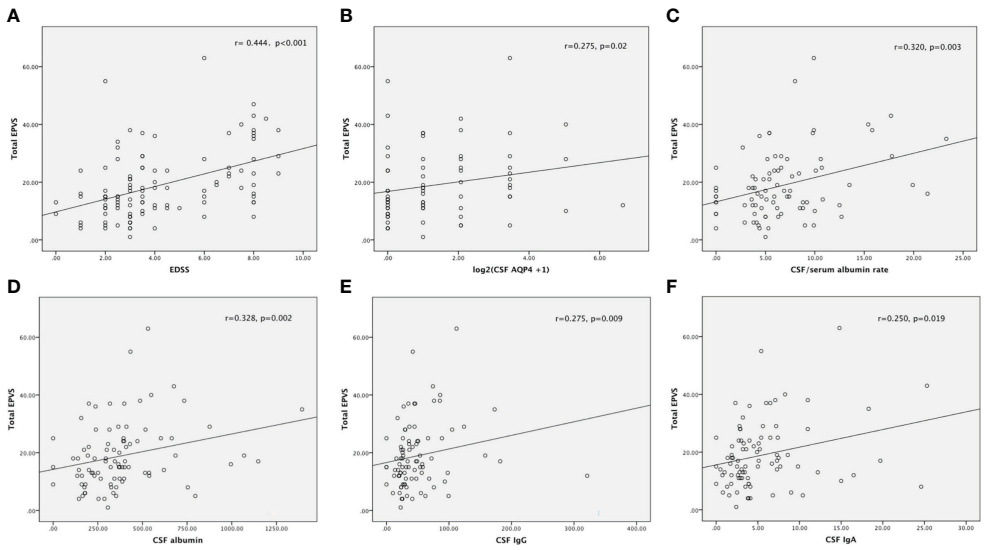


FIGURE 2
The number of total-EPVS was significantly correlated with EDSS score **(A)**, CSF titer of anti-AQP4 antibody **(B)**, CSF/Serum albumin rate **(C)**, the levels of CSF albumin **(D)**, CSF IgG **(E)** and CSF IgA **(F)**.

TABLE 2 Evaluation of independent factors associated with severe EPVS [a] (grade 3-4) in patients with neuromyelitis optica spectrum disorders by logistic regression analysis.

Model 1

Variates	B	Exp(B)/OR	95% confidence interval	P value
Age (years)	0.058	1.059	1.017-1.103	0.005
History of Hypertension	0.366	1.442	0.415-5.008	0.564
EDSS [b]	0.298	1.347	1.107-1.639	0.003
Model 2				
Variates	**B**	**Exp(B)/OR**	**95% confidence interval**	**P value**
Age (years)	0.097	1.102	1.033-1.175	0.003
History of Hypertension	0.983	2.672	0.541-13.201	0.228
CSF titer of Anti-AQP4 [c]	0.559	1.748	1.046-2.922	0.033
CSF IgG (mg/L)	-0.029	0.971	0.936-1.008	0.127
CSF IgA (mg/L)	-0.039	0.971	0.798-1.159	0.679
CSF/Serum albumin rate [d]	0.205	1.228	0.949-1.590	0.119
Serum Albumin (g/L)	0.046	1.047	0.879-1.246	0.607
EDSS	0.054	1.056	0.686-1.626	0.806

[a]EPVS, enlarged perivascular spaces.
[b]EDSS, Expanded Disability Status Scale.
[c]For the analysis of the CSF anti-AQP4 antibody titer X, a logarithmic value of $\log_2(X+1)$ was used.
[d]Albumin rate = Quotient (CSF/Ser)*10^{-3}.

which implied that more numbers of total-EPVS were related to more severe central nervous inflammation. (Figures 2D-F)

Logistic analysis of independent factors associated with severe EPVS in NMOSD patients

Two logistic regression models were conducted to find out the independent factors associated with severe EPVS (EPVS grade 3-4). As Table 2 showed, age and EDSS were independent factors in model 1 (including the covariates of age, history of hypertension and EDSS); while age and CSF titer of anti-AQP4 antibody were independent factors in model 2 (including the covariates of age, history of hypertension, CSF titer of anti-AQP4 antibody, CSF IgG, CSF IgA, CSF/serum albumin rate, serum albumin and EDSS). (Table 2)

Logistic analysis of independent predictors of disease severity in NMOSD patients

In order to clarify the independent predictive effect of total-EPVS with disease severity in NMOSD, patients were divided into two groups: mild NMOSD group (EDSS 0-4.0) (N=72) and severe NMOSD group (EDSS 4.5-9.5) (N=38). The variates of total-EPVS, as well as age, age of onset, numbers of all attacks, and serum albumin were entered into the logistic model. The result showed that total-EPVS and serum albumin level were two independent factors in the model to predict disease severity (OR=1.053, 95%CI 1.006-1.102, p=0.028 for total-EPVS; OR=0.858, 95% CI 0.765-0.962, p=0.009 for serum albumin) (Table 3). Furthermore, ROC analysis achieved AUC of 0.736 (0.640-0.831, p<0.001) for total-EPVS to determine severe NMOSD. (Figure 3)

TABLE 3 Evaluation of independent predictors of severe neuromyelitis optica spectrum disorders (EDSS 4.5-9.5) by logistic regression analysis.

Variates	B	Exp(B)/OR	95% confidence interval	P value
Age (years)	-0.034	0.967	0.841-1.112	0.637
Age of onset (years)	0.054	1.056	0.923-1.207	0.429
Serum Albumin (g/L)	-0.153	0.858	0.765-0.962	0.009
Number of all attacks	0.077	1.080	0.871-1.340	0.483
Total-EPVS	0.051	1.053	1.006-1.102	0.028

EDSS, Expanded Disability Status Scale.
EPVS, enlarged perivascular spaces.

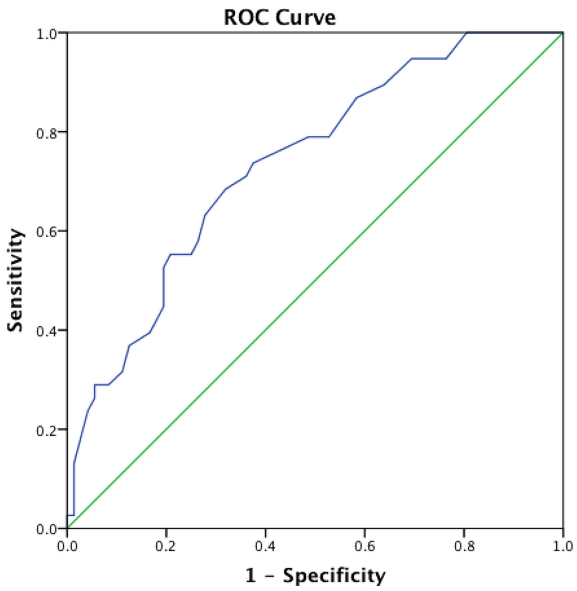


FIGURE 3
The ROC curve for total-EPVS to determine severe NMOSD (EDSS 4.5-9.5).

Discussion

Our study demonstrated that more total-EPVS was significantly correlated with higher CSF anti-AQP4 antibody titer, more severe blood-brain barrier disruption and intenser neuroinflammation. Moreover, total-EPVS was an independent predictor of severe neurological dysfunction in NMOSD.

PVS is part of the structure of glymphatic system. Studies have already proved that PVS are important in immune responses and inflammatory processes within the brain (12, 27). EPVS are believed to be associated with blood-brain barrier leakage and associated infiltration of monocytes, lymphocytes, and macrophages in the PVS (28).

Pathological examination showed abundant antibody-secreting cells were noted in perivascular spaces in autoimmune glial fibrillary acidic protein (GFAP) astrocytopathy, a CNS inflammatory disease targeting astrocytes (29). In a case report of Behçet's disease, the neuropathological examination revealed an acute inflammation consisted of a neutrophilic and eosinophilic infiltration of the perivascular spaces and brain parenchyma (30). It is suggested that during the pathogenesis of MS, T cell activation begins at the periphery of the lymphoid compartment (extracerebral area) and then reaches the CNS, with the T cells circulating in the PVS (10). In an animal model of progressive MS, immunohistological analysis showed that mature and isotype-switched B cells predominately localized to the meninges and perivascular space, with IgG isotype-switched B cells frequently accumulating in the parenchymal space (31).

Astrocytic endfeet and their dense expression of the aquaporin-4 water channels promote fluid exchange between the perivascular spaces and the neuropil. Thus, anti-AQP4 antibody targeting the main water channels on asctrocytes will impair CSF influx from PVS to neuropil in NMOSD (6). Therefore, we presume that the glymphatic system and EPVS might play even more important roles in the pathogenesis of NMOSD than the above-mentioned CNS inflammatory diseases.

Our results showed that patients with higher CSF titer of anti-AQP4 antibody had more EPVS on MRI. Higher level of CSF antibody will impair more water channels on astrocyte endfeet of PVS, which in turn block the influx of CSF and lead to the enlargement of PVS.

We also found that EPVS are associated with neuroinflammation and BBB disruption in NMOSD. As our data showed, severe EPVS are related to higher CSF albumin, IgG and IgA levels, which implied an intense inflammatory reaction in CNS; severe EPVS are also correlated with a greater impairment of BBB indicated by higher CSF/serum ratios for albumin. It has already been proved that glymphatic impairment aggravates CNS inflammation by suppressing cytokine clearance from the brain (8). The BBB is a very efficient barrier formed by the vascular endothelial cells, their tight junctions, and the underlying basement membrane. The BBB can be considered the internal boundary of PVS at the capillary level (32). Therefore, inflammation in the PVS can aggravate the impairment of BBB, and vice versa. It still remains unclear as to whether PVS dilation is a cause, effect, or secondary process of endothelial dysfunction and increased BBB permeability (33). Previous studies also demonstrated

EPVS to be a marker of BBB dysfunction (34), as well as a marker of neuroinflammation (10).

Our study also demonstrated that EPVS was independently associated with neurological dysfunction in NMOSD. Since EPVS can indicate inflammation in CNS, thereby more EPVS are related to more severe clinical presentation. Previous studies in MS have already demonstrated that increased numbers of EPVS are associated with clinical disability (23, 35).

The results of the current study prompt us to raise the following hypothesis. Anti-AQP4 antibodies originate in peripheral and enter the PVS through endothelial transcytosis or at areas of increased BBB permeability. Then anti-AQP4 antibodies in PVS bind selectively to AQP4 on astrocyte endfeet. This interaction results in down-regulation of surface AQP4 and less clearance of waste and pro-inflammatory cytokines in the CNS. Moreover, it activates complement produced locally by astrocytes, which in turn leads to increased BBB permeability and massive infiltration of leukocytes and cytokines. And inflammation in PVS will increase the numbers and volumes of EPVS. The neurological dysfunction will also aggravate in the condition of more severe neuroinflammation.

There are several limitations in the current study. One limitation is the relatively small number of patients included and the retrospective design of the study; although the statistical analyses were significant, the correlations were relatively weak. Therefore, large-scale prospective studies should be conducted with longer periods of follow-up to confirm the results drawn from the current study. Secondly, more biomarkers of neuroinflammation and BBB function such as CSF pro-inflammatory cytokines and radiological studies should be used to further prove the current results. Thirdly, the glymphatic system has been proved to be existed in brain and optic nerves; however, whether it exists in spinal cord need further studies. It is an important question to be explored because spinal cord is an end organ frequently attacked in NMOSD. Finally, we also found a relationship between EVPS and disease severity in anti-AQP4 antibody negative NMOSD patients and it could not be explained by the above-mentioned hypothesis. There might be other mechanisms involved.

Conclusion

In conclusion, we found a relationship between EPVS and neuroinflammation and BBB function in NMOSD. Moreover, EPVS might independently predict neurological dysfunction in patients with NMOSD.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving human participants were reviewed and approved by ethics committee of Ren Ji Hospital. The patients/participants provided their written informed consent to participate in this study.

Author contributions

X-YY: conception, methodology, data analysis, and writing original draft. M-CG: resources, data collection. S-WB: resources, data collection. LX: data analysis. Y-YS: resources, data collection. C-RX: resources, data collection. Y-FW: resources, data collection. YH: data curation. YZ: validation, methodology. Y-TG: conception. supervision, writing – review and editing. All authors contributed to the article and approved the submitted version.

Funding

This study was supported by the National Natural Science Foundation of China (No. 81801211) and the Outstanding Youth Training Funds of Shanghai Ren Ji Hospital (No. PYII-17-001). Innovative research team of high-level local universities in Shanghai (SHSMU-ZDCX20211901).

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fimmu.2022.966781/full#supplementary-material

References

1. Iliff JJ, Wang M, Liao Y, Plogg BA, Peng W, Gundersen GA, et al. A paravascular pathway facilitates CSF flow through the brain parenchyma and the clearance of interstitial solutes, including amyloid β. *Sci Transl Med* (2012) 4 (147):147ra111. doi: 10.1126/scitranslmed.3003748

2. Abbott NJ, Pizzo ME, Preston JE, Janigro D, Thorne RG. The role of brain barriers in fluid movement in the CNS: Is there a 'glymphatic' system? *Acta Neuropathol* (2018) 135(3):387–407. doi: 10.1007/s00401-018-1812-4

3. Mogensen FL, Delle C, Nedergaard M. The glymphatic system (En)during inflammation. *Int J Mol Sci* (2021) 22(14):7491. doi: 10.3390/ijms22147491

4. Hutchings M, Weller RO. Anatomical relationships of the pia mater to cerebral blood vessels in man. *J Neurosurg* (1986) 65(3):316–25. doi: 10.3171/jns.1986.65.3.0316

5. Groeschel S, Chong WK, Surtees R, Hanefeld F. Virchow-robin spaces on magnetic resonance images: Normative data, their dilatation, and a review of the literature. *Neuroradiology* (2006) 48(10):745–54. doi: 10.1007/s00234-006-0112-1

6. Mestre H, Hablitz LM, Xavier AL, Feng W, Zou W, Pu T, et al. Aquaporin-4-dependent glymphatic solute transport in the rodent brain. *Elife* (2018) 7:e40070. doi: 10.7554/eLife.40070

7. Mestre H, Kostrikov S, Mehta RI, Nedergaard M. Perivascular spaces, glymphatic dysfunction, and small vessel disease. *Clin Sci (Lond)* (2017) 131 (17):2257–74. doi: 10.1042/CS20160381

8. Filiano AJ, Gadani SP, Kipnis J. How and why do T cells and their derived cytokines affect the injured and healthy brain? *Nat Rev Neurosci* (2017) 18(6):375–84. doi: 10.1038/nrn.2017.39

9. Tsutsumi S, Ito M, Yasumoto Y, Tabuchi T, Ogino I. The virchow-robin spaces: Delineation by magnetic resonance imaging with considerations on anatomofunctional implications. *Childs Nerv Syst* (2011) 27(12):2057–66. doi: 10.1007/s00381-011-1574-y

10. Wuerfel J, Haertle M, Waiczies H, Tysiak E, Bechmann I, Wernecke KD, et al. Perivascular spaces–MRI marker of inflammatory activity in the brain? *Brain* (2008) 131(Pt 9):2332–40. doi: 10.1093/brain/awn171

11. Zhu YC, Dufouil C, Mazoyer B, Soumaré A, Ricolfi F, Tzourio C, et al. Frequency and location of dilated virchow-robin spaces in elderly people: A population-based 3D MR imaging study. *AJNR Am J Neuroradiol* (2011) 32 (4):709–13. doi: 10.3174/ajnr.A2366

12. Achiron A, Faibel M. Sandlike appearance of virchow-robin spaces in early multiple sclerosis: A novel neuroradiologic marker. *AJNR Am J Neuroradiol* (2002) 23(3):376–80.

13. Granberg T, Moridi T, Brand JS, Neumann S, Hlavica M, Piehl F, et al. Enlarged perivascular spaces in multiple sclerosis on magnetic resonance imaging: A systematic review and meta-analysis. *J Neurol* (2020) 267(11):3199–212. doi: 10.1007/s00415-020-09971-5

14. Vos CM, Geurts JJ, Montagne L, van Haastert ES, Bö L, van der Valk P, et al. Blood-brain barrier alterations in both focal and diffuse abnormalities on postmortem MRI in multiple sclerosis. *Neurobiol Dis* (2005) 20(3):953–60. doi: 10.1016/j.nbd.2005.06.012

15. Greter M, Heppner FL, Lemos MP, Odermatt BM, Goebels N, Laufer T, et al. Dendritic cells permit immune invasion of the CNS in an animal model of multiple sclerosis. *Nat Med* (2005) 11(3):328–834. doi: 10.1038/nm1197

16. Wingerchuk DM, Lennon VA, Pittock SJ, Lucchinetti CF, Weinshenker BG. Revised diagnostic criteria for neuromyelitis optica. *Neurology* (2006) 66(10):1485–9. doi: 10.1212/01.wnl.0000216139.44259.74

17. Lennon VA, Wingerchuk DM, Kryzer TJ, Pittock SJ, Lucchinetti CF, Fujihara K, et al. A serum autoantibody marker of neuromyelitis optica: Distinction from multiple sclerosis. *Lancet* (2004) 364(9451):2106–12. doi: 10.1016/S0140-6736(04)17551-X

18. Wingerchuk DM, Lennon VA, Lucchinetti CF, Pittock SJ, Weinshenker BG. The spectrum of neuromyelitis optica. *Lancet Neurol* (2007) 6(9):805–15. doi: 10.1016/S1474-4422(07)70216-8

19. Wingerchuk DM, Banwell B, Bennett JL, Cabre P, Carroll W, Chitnis T, et al. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. *Neurology* (2015) 85(2):177–89. doi: 10.1212/WNL.0000000000001729

20. Chen ZG, Huang J, Fan R, Weng RH, Shinohara RT, Landis JR, et al. Urinalysis in patients with neuromyelitis optica spectrum disorder. *Eur J Neurol* (2020) 27(4):619–25. doi: 10.1111/ene.14128

21. Kessler RA, Mealy MA, Levy M. Early indicators of relapses vs pseudorelapses in neuromyelitis optica spectrum disorder. *Neurol Neuroimmunol Neuroinflamm* (2016) 3(5):e269. doi: 10.1212/NXI.0000000000000269

22. Potter GM, Chappell FM, Morris Z, Wardlaw JM. Cerebral perivascular spaces visible on magnetic resonance imaging: Development of a qualitative rating scale and its observer reliability. *Cerebrovasc Dis* (2015) 39(3-4):224–31. doi: 10.1159/000375153

23. Cavallari M, Egorova S, Healy BC, Palotai M, Prieto JC, Polgar-Turcsanyi M, et al. Evaluating the association between enlarged perivascular spaces and disease worsening in multiple sclerosis. *J Neuroimaging* (2018) 28(3):273–7. doi: 10.1111/jon.12490

24. Wu B, Yao X, Lei C, Liu M, Selim MH. Enlarged perivascular spaces and small diffusion-weighted lesions in intracerebral hemorrhage. *Neurology* (2015) 85 (23):2045–52. doi: 10.1212/WNL.0000000000002169

25. Wardlaw JM, Smith EE, Biessels GJ, Cordonnier C, Fazekas F, Frayne R, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. *Lancet Neurol* (2013) 12(8):822–38. doi: 10.1016/S1474-4422(13)70124-8

26. Charidimou A, Boulouis G, Pasi M, Auriel E, van Etten ES, Haley K, et al. MRI-Visible perivascular spaces in cerebral amyloid angiopathy and hypertensive arteriopathy. *Neurology* (2017) 88(12):1157–64. doi: 10.1212/WNL.0000000000003746

27. Etemadifar M, Hekmatnia A, Tayari N, Kazemi M, Ghazavi A, Akbari M, et al. Features of virchow-robin spaces in newly diagnosed multiple sclerosis patients. *Eur J Radiol* (2011) 80(2):e104–8. doi: 10.1016/j.ejrad.2010.05.018

28. Wardlaw JM, Benveniste H, Nedergaard M, Zlokovic BV, Mestre H, Lee H, et al. Perivascular spaces in the brain: Anatomy, physiology and pathology. *Nat Rev Neurol* (2020) 16(3):137–53. doi: 10.1038/s41582-020-0312-z

29. Long Y, Liang J, Xu H, Huang Q, Yang J, Gao C, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in Chinese patients: A retrospective study. *Eur J Neurol* (2018) 25(3):477–83. doi: 10.1111/ene.13531

30. Hadfield MG, Aydin F, Lippman HR, Kubal WS, Sanders KM. Neuro-behçet's disease. *Clin Neuropathol* (1996) 15(5):249–55.

31. DiSano KD, Royce DB, Gilli F, Pachner AR. Central nervous system inflammatory aggregates in the theiler's virus model of progressive multiple sclerosis. *Front Immunol* (2019) 10:1821. doi: 10.3389/fimmu.2019.01821

32. Gouveia-Freitas K, Bastos-Leite AJ. Perivascular spaces and brain waste clearance systems: Relevance for neurodegenerative and cerebrovascular pathology. *Neuroradiology* (2021) 63(10):1581–97. doi: 10.1007/s00234-021-02718-7

33. Low A, Mak E, Rowe JB, Markus HS, O'Brien JT. Inflammation and cerebral small vessel disease: A systematic review. *Ageing Res Rev* (2019) 53:100916. doi: 10.1016/j.arr.2019.100916

34. Wardlaw JM, Doubal F, Armitage P, Chappell F, Carpenter T, Muñoz Maniega S, et al. Lacunar stroke is associated with diffuse blood-brain barrier dysfunction. *Ann Neurol* (2009) 65(2):194–202. doi: 10.1002/ana.21549

35. Favaretto A, Lazzarotto A, Riccardi A, Pravato S, Margoni M, Causin F, et al. Enlarged virchow robin spaces associate with cognitive decline in multiple sclerosis. *PloS One* (2017) 12(10):e0185626. doi: 10.1371/journal.pone.0185626

frontiers | Frontiers in Neurology

TYPE Case Report
PUBLISHED 05 October 2022
DOI 10.3389/fneur.2022.1017086

OPEN ACCESS

EDITED BY
Robert Weissert,
University of Regensburg, Germany

REVIEWED BY
Tejas Mehta,
University of Missouri, United States
Ryan Kammeyer,
University of Colorado, United States
Cullen Mark O'Gorman,
Princess Alexandra Hospital, Australia

*CORRESPONDENCE
Chengjuan Xie
xie_chengjuan@126.com

†These authors share first authorship

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

RECEIVED 11 August 2022
ACCEPTED 15 September 2022
PUBLISHED 05 October 2022

CITATION
Li A, Hu Y, Li J, Chen X, Jiang Y and Xie C (2022) Case report: Anti-GAD65 antibody-associated autoimmune encephalitis following HPV vaccination.
Front. Neurol. 13:1017086.
doi: 10.3389/fneur.2022.1017086

Case report: Anti-GAD65 antibody-associated autoimmune encephalitis following HPV vaccination

Aonan Li†, Ying Hu†, Jialu Li, Xingui Chen, Yubao Jiang and Chengjuan Xie*

Department of Neurology, The First Affiliated Hospital of Anhui Medical University, Hefei, China

Human papillomavirus (HPV) infection is a sexually transmitted disease that may lead to cervical cancer. HPV vaccines have been implemented widely to prevent this. While generally few complications of vaccination are reported, there have been occasional reports of adverse reactions post-vaccination. The safety profile of the HPV vaccine is reassuring. However, since its introduction, several serious post-vaccination central nervous system complications have been reported; however, causality has not been established. Herein, we describe a 39-year-old woman who developed seizures and experienced a rapid decline in memory shortly after her first dose of the HPV vaccine. Cranial magnetic resonance imaging and cerebrospinal fluid analysis were performed, and the patient was diagnosed with anti-glutamic acid decarboxylase 65 (anti-GAD65) antibody-associated autoimmune encephalitis. She responded well to high-dose glucocorticoids. Four-month follow-up revealed full recovery and absence of recurrence. Since the HPV vaccine is administered worldwide, this case should raise clinicians' awareness regarding the possible CNS complications related to vaccinations, such as anti-GAD65 antibody-associated AE.

KEYWORDS

anti-glutamic acid decarboxylase 65 (anti-GAD65) antibody, autoimmune encephalitis (AE), seizure, human papillomavirus, vaccination, cervical cancer

Introduction

Cervical cancer is one of the most dangerous malignancies affecting women worldwide. An estimated 604,000 new cases of cervical cancer and 342,000 deaths are reported annually (1). Its occurrence is closely related to persistent infection with high-risk human papilloma virus (HPV). Currently, the HPV vaccine is the most important primary measure to prevent cervical cancer. Various HPV vaccinations currently on the market are safe and effective, with the bivalent HPV vaccine being more than 90% effective against HPV 16/18-related precancerous lesions (2, 3). However, adverse reactions to vaccines require equal attention. Several serious neurological disorders following vaccines have been reported, including autoimmune encephalitis (AE), myelitis and central nervous system (CNS) demyelination thus far (4–6).

Autoimmune encephalitis (AE) is a type of encephalitis mediated by mechanisms that induce an immune response against central nervous system antigens. Different subtypes of AEs are distinguished according to antibodies and have different clinical symptoms and prognoses. Among them, AE associated with anti-glutamate decarboxylase-rich antibodies is a treatable cause of encephalitis. Anti-GAD65 antibody-associated AE is characterized by acute or subacute seizures, psychiatric symptoms, and cognitive impairment. Other manifestations include cerebellar ataxia and stiff person syndrome (7–9). The patient may present with a single symptom or multiple symptoms together. In the absence of a detectable paraneoplastic etiology, it is also regarded as a type of limbic encephalitis. It responds well to immunotherapy, including intravenous human immunoglobulin, glucocorticoids, plasma exchange, and other immunosuppressive agents.

In this case report, we describe a 39-year-old woman with anti-GAD65 antibody-associated AE who had seizures and short-term memory deficits shortly after receiving her first dose of HPV vaccination.

Case description

A healthy 39-year-old female patient suffered a sudden generalized tonic-clonic seizure during sleep 12 days after receiving the first dose of bivalent HPV vaccine (Cecolin®, Xiamen Innovax, Xiamen, China). The patient complained of tinnitus, auditory hypersensitivity, dizziness, and memory loss, all of which began after the first seizure. Additionally, the patient complained of difficulty falling asleep, poor continuity of sleep at night, and feeling tired and weak after waking up. There are also symptoms of dysautonomia, such as excessive sweating.

Fever, psychiatric symptoms, or involuntary movements were not observed. The patient did not have any previous physical or mental illnesses, was not under any medication, and had no family history of genetic disorders. There were no abnormalities noted during the pre-HPV vaccination screening for cervical cancer, indicating that the patient did not have an HPV infection prior to vaccination.

After her third seizure, the patient was hospitalized for further examination and treatment. She was prescribed oral levetiracetam (1.0 g/day) to control seizures. Neurological examination was normal except for short-term memory impairment. Further assessment of the patient's overall cognitive and memory functions revealed a score of 24/30 (normal range 26–30) on the Montreal Cognitive Assessment (MoCA), showing impaired orientation, attention, and short-term memory. Results of the Auditory Vocabulary Learning Test showed a score of 6 for immediate recall, 9 for delayed recall, and 13 for recognition, indicating impaired memory function. Laboratory tests showed that blood cell count, blood biochemistry, thyroid function, rheumatic disease screening were within the normal range. Tumor marker detection revealed an elevated CA125 level (34.79 U/mL, normal range 0–30.2 U/mL), while the levels of other tumor markers were normal. Semi-quantitative detection of paraneoplastic neuron antibody showed that the GAD65 antibody was positive, with a titer of 32 AU; other antibodies were negative. Thoraco-abdomino-pelvic computed tomography showed a few foci of fibrosis in the middle lobe of the right lung and ovarian cysts on the left side, without other abnormal findings. Pelvic ultrasound and further examination of the bilateral adnexal showed no abnormalities. Subsequently, the findings of pelvic computed tomography were considered a menstrual-related ovarian cyst, and teratoma was excluded. Ultrasound of the thyroid, breast, bilateral neck, and axillary lymph nodes showed a benign right breast nodule and a left thyroid cyst, with no change in size or morphology compared with that of the patient's physical examination conducted 2 years ago. Magnetic resonance imaging of the brain showed abnormal signals in both medial temporal lobes, predominantly on the right side (Figure 1). Video electroencephalogram monitoring revealed widespread diffuse slow waves during wakefulness.

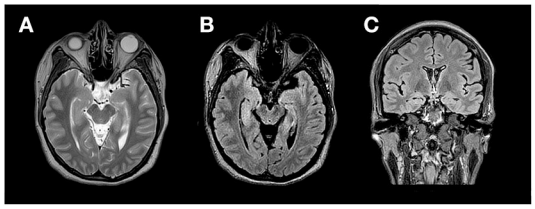


FIGURE 1
MRI (3.0T) at the time of presentation. Axial T2 Weighted Image **(A)**, Axial Fluid attenuated inversion recovery (FLAIR) sequence **(B)** and coronal FLAIR **(C)** show cortical thickening and hyper intense signal in the medial aspect of bilateral temporal lobes Right > Left.

Cerebrospinal fluid (CSF) analysis showed elevated leukocytes (35 leukocytes, 100% mononuclear cells), normal protein, glucose, and chloride, and no CSF infection. Cell-based and tissue-based assays revealed that the CSF was positive for GAD65 antibody (titer, 1:100++), mainly distributed in the hippocampus, striatum, cerebral cortex, and cerebellum (Figure 2). All other AE-associated antibodies tested negative.

Based on these results, the patient was diagnosed with anti-GAD65 antibody-associated AE. Pulse dose steroid therapy was initiated—high dose glucocorticoids (methylprednisolone, 500 mg daily for 5 days) initially and tapered to 250 mg daily for 5 days, with a good response. Oral prednisone treatment was

Abbreviations: AE, autoimmune encephalitis; anti-GAD65, anti-glutamic acid decarboxylase 65; CSF, cerebrospinal fluid; GABA, gamma-aminobutyric acid; HPV, human papilloma virus.

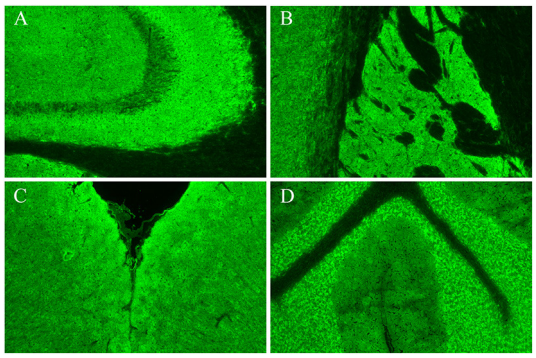


FIGURE 2
Results of cerebrospinal fluid using tissue-based assays (TBA). Hippocampus **(A)**, striatum **(B)**, cerebral cortex **(C)**, and cerebellum **(D)**.

continued after discharge, initially 60 mg per day then gradually tapered to 5 mg for maintenance. On follow-up, the patient reported no further seizures after discharge, and her memory and sleep quality have improved. After 4 months, the Video EEG retest showed normal results. The MoCA score improved to 30/30, representing an improvement in her overall cognitive function. The immediate recall, delayed recall, and recognition scores in the Auditory Vocabulary Learning Test increased to 10, 13, and 15, respectively, indicating improved memory. Nonetheless, she complained of a persistent subtle decline in memory relative to her premorbid state.

Discussion

In this report, we describe a woman with generalized tonic-clonic seizure during sleep and short-term memory impairment 12 days after her first dose of the bivalent HPV vaccine. After excluding other causes, the patient's clinical presentation, positive CSF GAD65 antibody, neuroimaging, and video electroencephalogram monitoring findings, led to the diagnosis of anti-GAD65 antibody-associated AE, which meets Graus' criteria for autoimmune encephalitis (10). In patients with AE with seizures as the first symptom, the differential diagnosis should include diseases such as viral encephalitis, metabolic encephalopathy, central nervous system demyelination, etc.

The HPV vaccine is essential for reducing cervical cancer incidence and mortality. Symptoms such as headaches, dizziness, nausea, and muscle pain are common and self-limiting adverse reactions after the HPV vaccination (11, 12). Although the safety and efficacy of HPV vaccines have been demonstrated in clinical trials (13), post-marketing surveillance has turned up multiple reports of serious neurological adverse effects (e.g., such as acute disseminated encephalomyelitis, Guillain-Barré syndrome, multiple sclerosis, optic neuritis, and encephalitis) following vaccination (4, 6, 14–17). Therefore, based on previous reports of CNS adverse reactions following HPV vaccination, clinicians were quick to consider HPV vaccination as a possible trigger for anti-GAD65 antibody-associated AE.

In a study of 1,396 cases of encephalitis after vaccination (hepatitis B, influenza, Haemophilus influenza type B, and measles-mumps-rubella), the onset of encephalitis was within 2 weeks of vaccination in 708 patients (18). This patient had symptoms ∼12 days after her vaccination; she had no illness such as fever or respiratory infections before the onset of the disease. Hence, based on the compatible temporal relationship between the onset of symptoms and the presumed trigger, physical examination and diagnostic tests, and the exclusion of an HPV infection through cervical cancer virus screening before vaccination. This patient was diagnosed with anti-GAD65 antibody-associated AE, the first case of autoimmune encephalitis linked to the timing of HPV vaccination.

The association between HPV vaccination and autoimmune encephalitis relates to various factors, including genetic susceptibility, immune dysfunction after viral infection, and the type of vaccine adjuvant. Susceptibility to infection, inflammation, and autoimmune responses vary among individuals, and individual heterogeneity in the immune response has a significant impact on the response after vaccination.

The pathophysiological mechanisms that trigger neuroinflammation after vaccination need to be further investigated. Strong expression of pro-inflammatory cytokines and T-cell responses may be responsible for the neuroinflammation triggered after vaccination. The ChAdOx1 nCoV-19 vaccine has demonstrated this in clinical trials (19). Many target genes are induced and transcribed, leading to the synthesis and release of pyrogenic cytokines into the circulation (12), imitating the body's reaction to a natural infection. Following stimulation, the body produces a complicated set of immune cascade responses. These cytokines and inflammatory mediators in the blood can induce a strong immune response in the nervous system. In some, microglia activation leads to the development of neuroinflammation (12, 20). In addition, there are hypotheses that the link between vaccination and encephalitis may be due to the opening of the blood-brain barrier and disruption of immune tolerance caused by CNS infection, or through molecular mimicry, such as a shared pathogenic epitope between the vaccine (antigen) and CNS structures (21). Nevertheless, this may be the explanation for the possible association between vaccination and autoimmune encephalitis.

Investigations on adverse reactions to vaccines have found that aluminum adjuvants used in vaccines can also trigger autoimmune reactions through specific molecular patterns and non-specific mechanism (22). In addition, aluminum adjuvants can also trigger an acute encephalopathic state by enhancing the immune response to antigens (23). And

aluminum adjuvants are also themselves antigens; they can pass through the blood-brain barrier and deposit in the brain, generating neurotoxic effects and compromising cognitive function. Genetic susceptibility matters to the immune response following vaccination. Human leukocyte antigen and non-human leukocyte antigen genes drive the differences in immune responses following vaccination. Previous studies demonstrated an association between different human leukocyte antigen genes and hyper- or hyporesponsiveness to vaccination (24). Therefore, the development of autoimmune-related diseases is more likely among patients with a genetic predisposition to an enhanced response to vaccination (25). However, additional mechanisms may be implicated.

Large-scale and long-term safety data shows that HPV vaccination has not led to an increased incidence of autoimmune disease (26–28). It is possible that no direct association between vaccination and disease exists in this patient. Post-vaccination, some cases of neurological disease may arise only by chance. Because this is a case report, we admit that a causal relationship between vaccination and encephalitis cannot be proven. Observational data or animal models will be needed to determine causality. Moreover, we continue to believe that the advantages of vaccination significantly outweigh the possible hazards of an ongoing immunization program.

Conclusion

We report the first case of anti-GAD65 antibody-associated AE following HPV vaccination in a patient who recovered well without severe cognitive impairment due to prompt diagnosis and treatment. However, the etiology of autoimmune encephalitis after HPV vaccination is not completely clear and several possible mechanisms may be involved and must be further investigated. For now, it is important to collect reports of autoimmune encephalitis after vaccination. Therefore, this case report provides clinicians with the opportunity to identify potentially vaccine-associated anti-GAD65 antibody-associated AE. In the meantime, clinicians should be vigilant in inquiring about the vaccination history of patients with AE and may need to pay particular attention to patients with anti-GAD65 antibody-associated AE who were injected with the HPV vaccine.

Patient perspective

When the patient had her first seizure, she and her family were very worried. Thorough investigation post admission revealed the actual cause of seizure, which brought relief to the patient and her family. After several days of glucocorticoid pulse therapy, seizure recurrence ceased, and her memory gradually recovered. The patient learned that her encephalitis was most likely a rare adverse reaction related to vaccination and was happy to share her case. At the same time, she hopes that people with the same condition will be as fortunate to be diagnosed and treated promptly.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary material, further inquiries can be directed to the corresponding author/s.

Ethics statement

The studies involving human participants were reviewed and approved by Ethics Committee of Anhui Medical University. The patients/participants provided their written informed consent to participate in this study. Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

Author contributions

AL compiled background information and wrote the manuscript. YH, JL, and XC acquired and analyzed data. YH, YJ, and XC were treating physicians. AL and CX revised the manuscript. All authors approved the final manuscript.

Funding

This study was supported by the Anhui Provincial Natural Science Foundation (No. 1908085MH249).

Acknowledgments

The authors highly appreciate the understanding and support from the patient and her family.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those

of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.1017086/full#supplementary-material

References

1. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin.* (2021) 71:209–49. doi: 10.3322/caac.21660

2. Centers for Disease Control and Prevention (CDC). FDA licensure of bivalent human papillomavirus vaccine (HPV2, Cervarix) for use in females and updated HPV vaccination recommendations from the Advisory Committee on Immunization Practices (ACIP). *MMWR Morb Mortal Wkly Rep.* (2010) 59:626–9.

3. Paavonen J, Naud P, Salmerón J, Wheeler CM, Chow SN, Apter D, et al. Efficacy of human papillomavirus (HPV)-16/18 AS04-adjuvanted vaccine against cervical infection and precancer caused by oncogenic HPV types (PATRICIA): final analysis of a double-blind, randomised study in young women. *Lancet.* (2009) 374:301–14. doi: 10.1016/S0140-6736(09)61248-4

4. Hviid A, Svanstrom H, Scheller NM, Gronlund O, Pasternak B, Arnheim-Dahlstrom L. Human papillomavirus vaccination of adult women and risk of autoimmune and neurological diseases. *J Intern Med.* (2018) 283:154–65. doi: 10.1111/joim.12694

5. Martin S, Azzouz B, Morel A, Trenque T. Anti-NMDA receptor encephalitis and vaccination: a disproportionality analysis. *Front Pharmacol.* (2022) 13:940780. doi: 10.3389/fphar.2022.940780

6. Sekiguchi K, Yasui N, Kowa H, Kanda F, Toda T. Two cases of acute disseminated encephalomyelitis following vaccination against human papilloma virus. *Intern Med.* (2016) 55:3181–4. doi: 10.2169/internalmedicine.55.5472

7. Graus F, Saiz A, Dalmau J. GAD antibodies in neurological disorders - insights and challenges. *Nat Rev Neurol.* (2020) 16:353–65. doi: 10.1038/s41582-020-0359-x

8. Honnorat J, Plazat LO. Autoimmune encephalitis and psychiatric disorders. *Rev Neurol.* (2018) 174:228–36. doi: 10.1016/j.neurol.2017.11.004

9. Vrillon A, Carle G, Berzero G, Honnorat J, Huberfeld G, Psimaras D, et al. Psychiatric symptoms in anti glutamic acid decarboxylase associated limbic encephalitis in adults: a systematic review. *Neurosci Biobehav Rev.* (2020) 119:128–37. doi: 10.1016/j.neubiorev.2020.08.015

10. Graus F, Titulaer MJ, Balu R, Benseler S, Bien CG, Cellucci T, et al. A clinical approach to diagnosis of autoimmune encephalitis. *Lancet Neurol.* (2016) 15:391–404. doi: 10.1016/S1474-4422(15)00401-9

11. Bonaldo G, Vaccheri A, D'Annibali O, Motola D. Safety profile of human papilloma virus vaccines: an analysis of the US Vaccine Adverse Event Reporting System from 2007 to 2017. *Br J Clin Pharmacol.* (2019) 85:634–43. doi: 10.1111/bcp.13841

12. Herve C, Laupeze B, Del Giudice G, Didierlaurent AM, Tavares Da Silva F. The how's and what's of vaccine reactogenicity. *NPJ Vaccines.* (2019) 4:39. doi: 10.1038/s41541-019-0132-6

13. Wheeler CM, Castellsagué X, Garland SM, Szarewski A, Paavonen J, Naud P, et al. Cross-protective efficacy of HPV-16/18 AS04-adjuvanted vaccine against cervical infection and precancer caused by non-vaccine oncogenic HPV types: 4-year end-of-study analysis of the randomised, double-blind PATRICIA trial. *Lancet Oncol.* (2012) 13:100–10. doi: 10.1016/S1470-2045(11)70287-X

14. Grimaldi-Bensouda L, Rossignol M, Kone-Paut I, Krivitzky A, Lebrun-Frenay C, Clet J, et al. Risk of autoimmune diseases and human papilloma virus (HPV) vaccines: Six years of case-referent surveillance. *J Autoimmun.* (2017) 79:84–90. doi: 10.1016/j.jaut.2017.01.005

15. Mouchet J, Salvo F, Raschi E, Poluzzi E, Antonazzo IC, De Ponti F, et al. Human papillomavirus vaccine and demyelinating diseases-A systematic review and meta-analysis. *Pharmacol Res.* (2018) 132:108–18. doi: 10.1016/j.phrs.2018.04.007

16. Martinez-Lavin M. HPV vaccine: adverse event signals were minimised or ignored. *BMJ.* (2019) 366:l4508. doi: 10.1136/bmj.l4508

17. Anamnart C, Tisavipat N, Owattanapanich W, Apiwattanakul M, Savangned P, Prayoonwiwat N, et al. Newly diagnosed neuromyelitis optica spectrum disorders following vaccination: case report and systematic review. *Mult Scler Relat Disord.* (2022) 58:103414. doi: 10.1016/j.msard.2021.103414

18. Al Qudah Z, Abukwaik W, Patel H, Souayah N. Encephalitis after Vaccination in United States. A Report from the CDC/FDA Vaccine Adverse Event Reporting System. [1990–2010]. *Neurology.* (2012) 78:P03.151. doi: 10.1212/WNL.78.1_MeetingAbstracts.P03.151

19. Ewer KJ, Barrett JR, Belij-Rammerstorfer S, Sharpe H, Makinson R, Morter R, et al. T cell and antibody responses induced by a single dose of ChAdOx1 nCoV-19 (AZD1222) vaccine in a phase 1/2 clinical trial. *Nat Med.* (2021) 27:270–8. doi: 10.1038/s41591-020-01194-5

20. Giannotta G, Giannotta N. Vaccines and neuroinflammation. *Int J Pub Heal Safe.* (2018) 3:1000163.

21. Huynh W, Cordato DJ, Kehdi E, Masters LT, Dedousis C. Post-vaccination encephalomyelitis: literature review and illustrative case. *J Clin Neurosci.* (2008) 15:1315–22. doi: 10.1016/j.jocn.2008.05.002

22. Salemi S, D'Amelio R. Could autoimmunity be induced by vaccination? *Int Rev Immunol.* (2010) 29:247–69. doi: 10.3109/08830181003746304

23. Khan Z, Combadière C, Authier FJ, Itier V, Lux F, Exley C, et al. Slow CCL2-dependent translocation of biopersistent particles from muscle to brain. *BMC Med.* (2013) 11:99. doi: 10.1186/1741-7015-11-99

24. Ovsyannikova IG, Poland GA. Vaccinomics: current findings, challenges and novel approaches for vaccine development. *AAPS J.* (2011) 13:438–44. doi: 10.1208/s12248-011-9281-x

25. Posteraro B, Pastorino R, Di Giannantonio P, Ianuale C, Amore R, Ricciardi W, et al. The link between genetic variation and variability in vaccine responses: systematic review and meta-analyses. *Vaccine.* (2014) 32:1661–9. doi: 10.1016/j.vaccine.2014.01.057

26. Willame C, Rosillon D, Zima J, Angelo MG, Stuurman AL, Vroling H, et al. Risk of new onset autoimmune disease in 9- to 25-year-old women exposed to human papillomavirus-16/18 AS04-adjuvanted vaccine in the United Kingdom. *Hum Vaccin Immunother.* (2016) 12:2862–71. doi: 10.1080/21645515.2016.1199308

27. Lehtinen M, Eriksson T, Apter D, Hokkanen M, Natunen K, Paavonen J, et al. Safety of the human papillomavirus (HPV)-16/18 AS04-adjuvanted vaccine in adolescents aged 12–15 years: Interim analysis of a large community-randomized controlled trial. *Hum Vaccin Immunother.* (2016) 12:3177–85. doi: 10.1080/21645515.2016.1183847

28. Phillips A, Hickie M, Totterdell J, Brotherton J, Dey A, Hill R, et al. Adverse events following HPV vaccination: 11 years of surveillance in Australia. *Vaccine.* (2020) 38:6038–46. doi: 10.1016/j.vaccine.2020.06.039

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 05 October 2022
DOI 10.3389/fneur.2022.990553

OPEN ACCESS

EDITED BY
Harry Alexopoulos,
National and Kapodistrian University of Athens, Greece

REVIEWED BY
Wei Shan,
Capital Medical University, China
Stefan Blum,
The University of Queensland, Australia

*CORRESPONDENCE
Hongzhi Guan
pumchghz@126.com

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

RECEIVED 10 July 2022
ACCEPTED 09 September 2022
PUBLISHED 05 October 2022

CITATION
Bai L, Ren H, Liang M, Lu Q, Lin N, Liu M, Fan S, Cui R and Guan H (2022) Neurological disorders associated with glutamic acid decarboxylase 65 antibodies: Clinical spectrum and prognosis of a cohort from China. *Front. Neurol.* 13:990553. doi: 10.3389/fneur.2022.990553

Neurological disorders associated with glutamic acid decarboxylase 65 antibodies: Clinical spectrum and prognosis of a cohort from China

Lin Bai[1], Haitao Ren[1], Menglin Liang[2], Qiang Lu[1], Nan Lin[1], Mange Liu[1], Siyuan Fan[1], Ruixue Cui[2] Hongzhi Guan[1]* on behalf of the Encephalitis Collaborative Group

[1]Department of Neurology, Peking Union Medical College Hospital, Peking Union Medical College and Chinese Academy of Medical Sciences, Beijing, China, [2]Department of Nuclear Medicine, Peking Union Medical College Hospital, Peking Union Medical College and Chinese Academy of Medical Sciences, Beijing, China

Objective: To describe clinical phenotypes and prognosis of neurological autoimmunity related to glutamic acid decarboxylase 65 (GAD65) antibodies in China.

Method: In this retrospective observational study from Peking Union Medical College Hospital, we identified patients with neurological disorders related to GAD65 antibodies (cell-based assay) from May 2015 to September 2021. Clinical manifestations, immunotherapy responsiveness, and outcomes were collected after obtaining informed consent from all patients.

Results: Fifty-five patients were included: 40 (72.73%) were women and initial neurological symptoms developed at 42(34-55) years of age. The median time to the nadir of the disease was 5 months (range from 1 day to 48 months). The clinical syndromes included limbic encephalitis (LE) or epilepsy (Ep) (n = 34, 61.82%), stiff-person syndromes (SPS) (n = 18, 32.73%), autoimmune cerebellar ataxia (ACA) (n = 11, 20%), and overlap syndrome in eight (14.55%) patients. Thirty-two (58.2%) patients had comorbidities of other autoimmune diseases, including Hashimoto thyroiditis (n = 17, 53.13%), T1DM (n = 11, 34.78%), vitiligo (n = 6, 18.75%), and others (n=5, 15.63%). Two (3.64%) patients had tumors, including thymoma and small cell lung cancer. Fifty-one (92.7%) patients received first-line immunotherapy (glucocorticoids and/or IV immunoglobulin), and 4 (7.3%) received second-line immunotherapy (rituximab). Long-term immunotherapy (mycophenolate mofetil) was administered to 23 (41.8%) patients. At the median time of 15 months (IQR 6–33.75 month, range 3–96 month) of follow-up, the patients' median modified Rankin Score (mRS) had declined from 2 to 1. Thirty-eight (70.4%) patients experienced clinical improvement (mRS declined $\geq$1), 47 (87%) had favorable clinical outcomes (mRS $\leq$2), and nine were symptom-free (16.7%). The sustained response to immunotherapy ranged from 7/15 (63.63%) in ACA patients and 22/34 (64.7%) in LE/Ep patients to 14/17 (82.35%) in SPS patients.

Conclusions: LE/Ep was the most common neurological phenotype of GAD65 antibody neurological autoimmunity in our cohort. Most patients had comorbidities of other autoimmune diseases, but underlying tumors were rare. Most patients responded to immunotherapy. However, the long-term prognosis varied among different clinical phenotypes.

KEYWORDS

encephalitis, autoimmune disease, glutamic acid decarboxylases 65, antibody, immunotherapy

Introduction

Glutamic acid decarboxylase (GAD) is a rate-limiting enzyme in the synthesis of the inhibitory neurotransmitter gamma-aminobutyric acid (GABA). It consists of two isoforms — GAD65 and GAD67. GAD65 is highly enriched in nerve terminals (1) and mediates activity-dependent GABA synthesis when postsynaptic inhibition is needed (2). While GAD67 produces foundational neuronal cytosolic GABA (3). Autoantibodies against GAD may disrupt the synthesis of GABA and impair GABAergic inhibitory circuits.

GAD65 antibodies are associated with diabetes mellitus type 1 (T1DM) and diverse neurologic disorders. They were initially characterized in a patient with stiff-person syndrome (SPS) and T1DM in 1988 (4). Subsequently, GAD65 antibodies were also identified in patients with autoimmune cerebellar ataxia (ACA), limbic encephalitis (LE) and epilepsy (Ep).The complexity of the disease is influenced by diverse clinical phenomena and different prognoses. It is challenging for physicians to diagnose and treat. Recently, Muñoz-Lopetegi et al. (5) and Budhram et al. (6) reported case series of Caucasian patients. However, few large cohorts of GAD65 antibodies associated disorders have been reported in East Asia (7). In this study, we reported a case series in China to offer further insights into the clinical phenotypes and prognosis of GAD65 antibodies associated disorders.

Methods

Patients

Patients with GAD65 antibodies and neurologic symptoms (encephalopathy, epilepsy, psychiatric symptoms, rigidity, movement disorders, gait disturbances, diplopia, and sleep disorders) were enrolled between May 2015 and September 2021 in Peking Union Medical College Hospital (PUMCH) Encephalitis and Paraneoplastic Syndrome Project. GAD65 antibodies were detected by a cell-based assay (CBA). Clinical information was obtained from the patients' medical files. The data included age, gender, CSF test, MRI, EEG, therapeutic regimens, and treatment outcomes.

Standard protocol approvals, registrations, and patient consent

The institutional review board of PUMCH approved the study protocol (JS-891). Written informed consent was obtained from all patients.

Definition of the clinical phenotypes, immunotherapy regimen, and follow-up

LE was identified as subacute onset (rapid progression of fewer than 3 months) of working memory deficits, seizures, or psychiatric symptoms with medial temporal lobe T2-hyperintensity. Ep was classified by the International League Against Epilepsy 2017 (8). Overlap syndromes were identified when patients present with more than one neurologic syndrome. LE with Ep alone was not classified as an overlap syndrome.

The immunotherapy responses were assessed from the medical files. Clinical improvement was defined as a decrease in the modified Rankin score (mRS) ($\geq$1 point) from that at the previous visit. For patients with Ep, at least 50% seizure frequency reduction was considered an improvement. A favorable outcome was defined as an mRS $\leq$2, and a poor outcome was defined as an mRS $>$2 at the end of follow-up.

Laboratory tests

The cerebrospinal fluid (CSF) and serum samples were tested using a CBA (EUROIMMUN, Lübeck, Germany; REF: FA 1022-1005-50) in the neurological immunology laboratory of PUMCH. The antibody titers were measured using serial dilutions of serum and CSF until the reactivity was no longer visible.

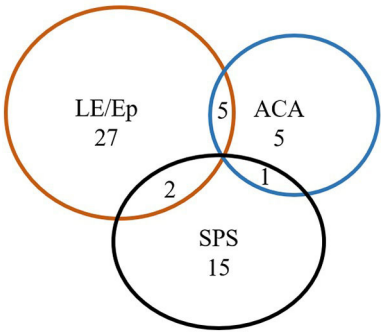


FIGURE 1
Neurological autoimmunity associated with anti-GAD65 antibodies. LE, limbic encephalitis; Ep, epilepsy; SPS, stiff person syndromes; ACA, autoimmune cerebellar ataxia.

Statistics

The median with the interquartile range (IQR) was used in continuous variables. Categorical variables are reported as numbers (percentages). We used Pearson's χ^2 or Fisher's exact test for multiple categories and a *t*-test or the Mann–Whitney U test for continuous variables. A two-sided $p < 0.05$ was considered statistically significant. We used SPSS 24.0 for analysis.

Results

Clinical characteristics and syndromes

Fifty-five patients were enrolled. The median age at onset was 42(34-55) years of age. Forty (72.7%) patients were females. The median time nadir was 5 months (range 1 day-6 months). Most common clinical manifestations were seizures ($n = 30$, 54.5%), progressive proximal limb rigidity ($n = 18$, 32.7%), working memory deficits ($n = 17$, 31.9%), and ataxia ($n = 11$, 20%). One patient manifested rapid-eye-movement sleep behavior disorder (RBD). All patients had typical neurological syndromes related to GAD65 antibody: Ep ($n = 8$, 14.5%), LE ($n = 26$, 47.3%), SPS ($n = 18$, 32.7%), and ACA ($n = 11$, 20%), and 8 (14.5%) patients had overlap syndromes (Figure 1, Table 1).

TABLE 1 Characteristics of the 55 patients with GAD65-ab associated with neurological autoimmunity.

Characteristics	Values
Female sex, *n* (%)	40 (72.73%)
Age, y (IQR)	42 (34-55)
Time to nadir, median (IQR; range)	5 months
	(1 d-6months; 1 d-48month)
Clinical symptoms, *n* (%)	
Seizures	30/55 (54.5%)
Muscle rigidity	18/55 (32.7%)
Memory deficits	17/55 (31.9%)
Ataxia	11/55 (20%)
Diplopia	7/55 (12.7%)
Psychosis	6/55 (10.9%)
Dysarthria	4/55 (7.3%)
RBD	1/55 (1.8%)
Autoimmune Comorbidities *n* (%)	32/55 (58.18%)
Hashimoto thyroiditis	17/32 (53.12%)
T1DM	11/32 (34.38%)
Vitiligo	6/32 (18.75%)
Rheumatoid arthritis	1/32 (3.13%)
Grave's disease	1/32 (3.13%)
Psoriasis	1/32 (3.13%)
Thrombocytopenia	1/32 (3.13%)
Myasthenia gravis	1/32 (3.13%)
Tumor found within 5 years of symptom onset	2/55 (3.34%)
Thymoma	1/2 (50%)
Small cell lung cancer	1/2 (50%)

RBD, rapid-eye-movement sleep behavior disorder; T1DM, type 1 diabetes mellitus; Time to nadir. The time from symptoms onset to attend a hospital.

LE and Ep

Thirty-four patients were involved, with 23 (67.6%) females. The patients had either an acute or a subacute onset (2 months vs. 10 months, $P = 0.001$). Fifty patients (91%) presented with tonic-clonic epileptic seizures, and all had focal onset, including aware motor onset (7/34, 23.3%) (such as lip-smacking, wandering, or other automatic activities), non-motor onset (18/34, 60%) (such as emotional seizures and sensory seizures), or impaired awareness (5/34, 16.7%). Seventeen (31.9%) patients were disturbed by memory decline: 10 of them acutely started, and seven patients had symptoms that occurred approximately half a year later.

SPS

Eighteen patients were involved, with 14 (77.8%) females. The median duration was 13 months, which was longer than the other phenotypes (13 m vs. 3 m, $P = 0.000$). The majority of the patients (15/18, 83.3%) had symptoms that began from the lower back or bilateral proximal limb. One patient had symptoms that began from the left lower limb (1/18, 5.56%), and two patients began from the neck and shoulders (2/18, 11.1%). Four patients manifested mild upper motor neuron (UMN) signs (brisk reflexes and Babinski sign).

TABLE 2 Different clinical syndromes of GAD65-ab-associated neurological autoimmunity.

Clinical syndrome	LE/Ep ($N = 34$)	SPS ($N = 18$)	ACA ($N = 11$)
Age, y, median (IQR, range)	41 (31–55.5, 6–75)	43(37-49,13-62)	54 (43–63, 29–75)
Female sex, n (%)	23 (67.6%)	14 (77.8%)	9 (81.8%)
Time to nadir, month, median (IQR; range)	2 (0.34–6, 0.03-48)	13 (3.75–48, 1–48)	6 (2–6, 0.03–15)
Autoimmune Comorbidities, *n* (%)	19 (55.9%)	11 (61.1%)	4 (36.4%)
Oncology	0	2	1
CSF WBC>5×106/L	3/22 (13.6%)	3/13 (23.1%)	2/9 (22.2%)
CSF OCB (+)	10/15 (66.7%)	9/10 (90%)	4/6 (66.7%)
Favorable outcome	22/34 (64.7%)	16/17 (94.1%)	7/11 (63.6%)

CSF, cerebrospinal fluid; OCB, oligoclonal bands; LE, limbic encephalitis; Ep, epilepsy; SPS, stiff person syndromes; ACA, autoimmune cerebellar ataxia; Time to nadir, The time from symptoms onset to attend a hospital; Favorable outcome, A favorable outcome was defined as an mRS≤2 at the end of follow-up.

ACA

There were 11 patients with a median onset age of 54 years, which was older than the patients with other GAD65 antibody related neurologic disorders (54 years vs. 41 years, $p = 0.039$). Gait ataxia was most frequently documented (11/11, 100%), followed by dizziness or diplopia (7/11, 63.6%), and dysarthria (4/11, 63.4%) (Table 2).

Overlap

Eight patients had combined syndromes, including LE/Ep combined with CA ($n = 5$, 62.5%), LE/Ep plus SPS ($n = 2$, 25%), and SPS plus CA ($n = 1$, 12.5%), with an interval mediation time of 10.5 months (4.5–22.5). LE/Ep was most likely to merge with other syndromes. Details were provided in the supplementary materials (Supplementary Table 1).

Oncology

Two (3.6%) female patients had a tumor within 5 years of symptom onset. One patient with SPS was diagnosed with thymoma (B1 type) at the onset, and her symptoms progressed after the thymoma removal. The other patient with small cell lung cancer (cTxN1M0, IIa) felt neck and shoulder stiffness at the onset and began to have diplopia and stumbling 6 months later. She received antitumor therapy and IVIG treatment. Neck and Shoulder stiffness disappeared, but ataxia was not approved (mRS 4).

Ancillary test results

Neuroimage

All patients underwent brain magnetic resonance imaging (MRI), and 36.36% (20/55) of the patients showed abnormal results, including 16 medial temporal lobe abnormalities, two multifocal cortical lesions, and two mild cerebellar atrophy.

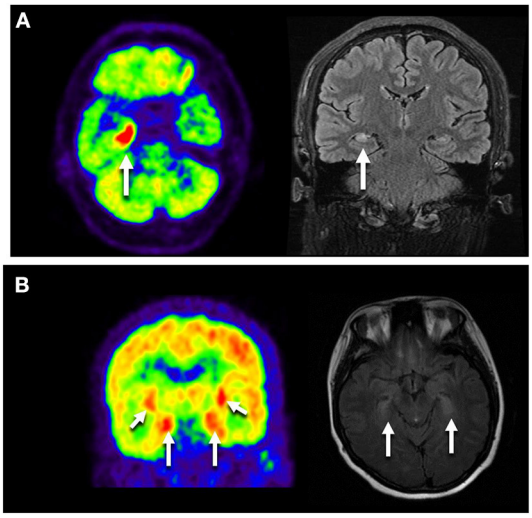


FIGURE 2
Neuroimages of the GAD65 antibody associated neurological disorders. **(A)** Images of NO.9 who presented with Ep. Axial FLAIR MRI showed right temporal hyperintensity, and Axial FDG-PET/CT scan showed hypermetabolic right mesiotemporal spot (white arrow). **(B)** Images of NO.41 who manifested as LE plus ACA. 18F-FDG PET/CT showed increased FDG uptake in the bilateral medial temporal lobes. MRI showed bilateral medial temporal lobe T2 hypertension. LE, limbic encephalitis; Ep, epilepsy; ACA, autoimmune cerebellar ataxia.

Thirteen patients completed 18F-FDG PET/CT scans: one presented with multicortical hypermetabolism, four with temporal lobe hypermetabolism, and one with cerebellar hypometabolism (Figure 2).

Electrophysiology

EEG was available for patients with LE/Ep. There were 16 patients with abnormal EEG findings, showing abnormal discharges in the temporal lobe. Ten patients with SPS

underwent multichannel surface electromyography, and 9/10 showed continuous motor unit activity in at least one axial muscle. One was normal because of clonazepam usage.

CSF

CSF white blood cell (WBC) count data were available for 37 patients. The median WBC count was 1 (IQR 0–4) $\times$ 10^6/L. Seven (18.9%) patients had pleocytosis. Twenty-six patients underwent CSF oligoclonal immunoglobulin G (IgG) bands (OCB) test. Seven patients had no oligoclonal bands despite having positive GAD65 antibodies in the CSF.

Treatment outcomes

Therapy

Fifty-one (92.7%) patients received immunotherapy. Forty-two (77.8%) patients received intravenous immunoglobulins (IVIG) (0.4 g/kg for 5 days), and 44 patients (81.5%) received intravenous methylprednisolone (IVMP). Four patients additionally received rituximab for second-line immunotherapy. Long-term immunotherapy with Mycophenolate Mofetil (MMF) was given in 23 (41.8%) patients as a maintenance therapy to prevent and manage relapses. Four patients refused immunotherapy, including two with SPS and two with Ep. They received treatment with clonazepam or antiepileptic drugs because of mild clinical symptoms.

Outcome

The median follow-up time was 15 months (IQR 6–33.75month, range 3–96 month). One patient with tumor-negative SPS failed to follow up. Thirty-eight (70.4%) patients experienced clinical improvement, and 9 (16.7%) were symptom-free. Forty-seven (87%) patients attained satisfactory neurologic function (mRS 0–2). The median mRS declined from 2 (IQR 2–3) to 1 (IQR 1–2). There were no deaths in our group, but one patient failed to attend the follow-up.

The median mRS in patients with SPS dropped from 3 (IQR 2.5-3) to 1 (IQR 0.5–2), with 14 (82.35%) patients showing clinical improvement, and 4 (22.2%) completely recovered. For CA, the median mRS score declined from 3 (IQR 2–4) to 2 (IQR 1–3). For LE or Ep patients, the median mRS changed from 2 (IQR 2-2) to 1 (IQR 1–2). 22 (64.7%) patients achieved a >50% seizure frequency reduction, and 5 (14.7%) were symptom-free. Sustained response to immunotherapy ranged from 63.6% in ACA and 64.7% in LE/Ep to 82.35% in SPS (Figure 3).

Most patients with LE converted to chronic epilepsy during the follow-up. The patients with LE or Ep had the same median mRS change after treatment, from two (IQR 2-2) to 1 (IQR 1–2). There was no statistically significant difference in the prognosis between the two groups ($P = 0.681$) when a clinical improvement was defined as a seizure frequency reduction $\geq$50% (Table 3).

Twenty-eight (50.9%) serum samples were available before and after the treatment. Seventeen (60.7%) patients' serum GAD65-ab titers were unchanged, while 13/28 (46.4%) had decreased. GAD65-ab did not turn seronegative. There was no significant correlation between the serum GAD65 antibody titer variation and prognosis ($P = 0.671$).

Discussion

This study aims to describe the clinical characteristics and prognosis of GAD65 antibodies related neurological disorders. To the best of our knowledge, this is the largest single-center cohort from East Asia. Our study provides several relevant findings: (1) The disorder predominately involves middle-aged females. (2) Patients present with several specific neuroimmune phenotypes, including CA, SPS, LE, and Ep. Patients with LE tend to get chronic seizures. (3) Most patients have autoimmune comorbidities, but underlying tumors are unusual. (4) Most patients experienced clinical improvement after immunotherapy, but only a minority remain symptom-free.

GAD65 antibodies related neurological disorders present with a set of well-established symptoms, including SPS, ACA, LE, and Ep. SPS usually involves the axial muscles and proximal limb muscles. Some patients have UMN manifestations that support spinal cord involvement (6, 9). In patients with epilepsy, abnormal discharges usually originate from the temporal lobe (10–12). We found that patients with LE can present with chronic seizures during years of follow-up and there was no significant difference in prognosis between LE and Ep ($P = 0.681$). This finding may indicate that the two clinical phenotypes have some common clinical features and underlying pathophysiology.

Autoimmune comorbidities are frequent in patients with GAD65-ab related neurological disorders, including autoimmune thyroid disease (30–48%), T1DM (11–30%), vitiligo (2–16%), and rheumatic disorders (6–7%) (10, 13–15). GAD antibodies were positive in over 50 % of patients with T1DM, and these patients had a higher prevalence of autoimmune thyroiditis than anti-GAD-negative patients with T1DM (16). Besides, A previous study has indicated that vitiligo may be a diagnostic clue to an autoimmune cause of encephalitis (17). The autoantibodies induced by the exposure antigen triggered by vitiligo are more likely to attack the mimic extracellular epitopes of neurons because both the skin and the nervous system are derived from the external germ layer. Further studies are needed to investigate the relationship between these diseases.

Approximately 4–11% of patients have underlying tumors (5–7, 18). In our group, 3.64% ($n = 2$) of the patients had manifestations of atypical SPS syndrome and overlap

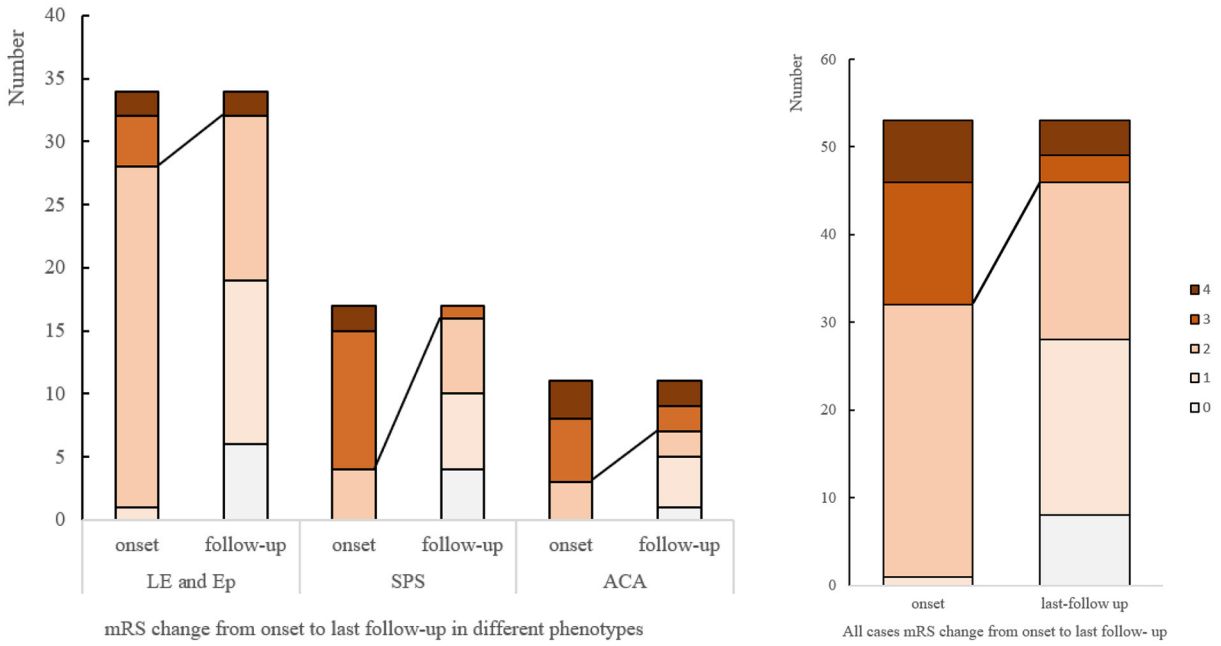


FIGURE 3
mRS changes from onset to last follow-up. mRS, modified Rankin score; LE, limbic encephalitis; Ep, epilepsy; SPS, stiff person syndromes; ACA, autoimmune cerebellar ataxia.

(SPS+CA) syndrome combined with tumors. This finding is lower than that of a previous study and suggests that patients presenting with CA, SPS, or atypical syndromes should be vigilant.

CSF tests and neuroimages are important for disease diagnosis. The CSF cell counts and protein levels are usually normal, but 40–70% of patients have oligoclonal immunoglobulin G (IgG) bands (19). Brain MRI shows parenchymal atrophy, cortical/subcortical parenchymal T2 hyperintensity, and abnormal hippocampal signals in LE/Ep (20). 18F-FDG PET/CT shows FDG uptake in the parietotemporal lobes (21). For patients with chronic epilepsy, hypometabolism in the mesial temporal lobe areas, together with hypometabolism in the insulae and medial inferior frontal-hypothalamus, may be characteristic of patients with GAD65-ab (22).

Immunotherapy is the main treatment strategy for anti-GAD65-related neurological disorders (23). However, there is a lack of consensus on the immunotherapy regimen. A study showed that IVIG had a better therapeutic effect than IVMP for patients with SPS and ACA (24). Another study found that corticosteroids were the best regimen for ACA (25). Some research indicates that there was no significant difference in the effectiveness between the two regimens (26, 27). Studies found that rituximab lacked efficacy in patients with SPS (28, 29). However, there were opposite standpoints reported (30, 31). Researchers found that tocilizumab was helpful for super-refractory status epilepticus (32). There are possible benefits from epilepsy surgery in some anti-GAD65-LE (14). Saidha et al. reported that MMF was effective in the treatment of anti-GAD65-associated limbic encephalitis (9, 33). Clinical treatment should take the disease severity, comorbidities, and patient economic situation into account to make an appropriate strategy.

Most patients show clinical improvement after immunotherapy. Previous studies reported that 63.6%-95% of patients experience clinical improvement, but only 0–1% of patients who present with ACA or Ep are symptom-free (6). This indicates that anti-GAD65-related neurological autoimmunity is a chronic disease. SPS has a better response to immunotherapy than epilepsy and ACA (34–36). The different prognoses may indicate that the target antigens are different among clinical phenotypes. In patients with pre-and post-treatment samples, serum GAD-Ab titers became lower after initial improvement and unchanged during follow-up (5, 37). In our cohort, most patients had satisfactory neurologic function (mRS≤2), but complete recovery only occurred in a minority of patients. About 40% of patients received MMF as long-term immunotherapy in this study. We found that MMF ($p = 0.306$) did not improve the clinical outcomes, possibly because of the relatively small sample size, and the role of long-term immunotherapy needs further investigation.

TABLE 3 Comparisons of the clinical data of the patients with GAD65-ab-associated neurological disorders.

	Favorable outcome	Poor outcome	*P*-value
Age (y)	42.5(12-71)	42.5(6-75)	0.766
Sex (*n*, %)			
Male	11/15	4/15	1.00
Female	27/39	12/39	
Diagnosis duration (month)	3 (0.03–48)	6.5 (0.03–21)	0.371
Symptoms (*n*, %)			
SPS	14/17	3/17	0.191
LE/Ep	22	12	0.279
ACA	7	4	0.716
Immunotherapy (*n*, %)			
First-line	18/27	9/27	0.551
First-line + MMF	15/19	4/19	0.309
Second line	1/4	3/4	0.073
Baseline mRS score >2 (*n*, %)	17/22	5/22	0.357

LE, limbic encephalitis; Ep, epilepsy; SPS, stiff person syndromes; ACA, autoimmune cerebellar ataxia; mRS, modified Rankin Score; MMF mycophenolate mofetil; IVIG, Intravenous Immunoglobulin; MP, methylprednisolone; First-line, IVIG and IVMP alone or combine; Second line, Rituximab.

There are limitations to this study. (1) As the national referral center for complicated diseases, our cohort may be biased by more complicated and refractory cases, which may introduce bias in the characterization of the entity we aim to describe in this review. (2) We collected clinical data from medical records, which may lead to an overestimation of some symptoms. (3) We need to find more suitable evaluation plans for each syndrome rather than mRS. (4) This study is a single-center cohort. More accurate prognostic evaluation warrants further large-size cohort investigation and extended follow-up.

To summarize, GAD65 antibody related neurological disorders present with various clinical symptoms, and LE/Ep are the most common phenotypes in Chinese patients. The majority of patients have clinical improvement after immunotherapy, but full recovery only occurs in a small proportion of patients. Therefore, this condition seems to be a chronic disease. Multicenter, large-cohort studies are needed to reach a consensus for standardizing the immunotherapy regimens and to obtain further insights into the prognosis.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary material, further inquiries can be directed to the corresponding author/s.

Author contributions

LB: data curation, writing the original draft, and submitting. HR: data collection and data analysis. ML: writing the orginal draft and supervision. SF and RC: supervision. HG: supervision, writing-reviewing, and editing. QL and NL: article supervision in the process of manuscript revision. All authors agree to be accountable for the content of the work. All authors contributed to the article and approved the submitted version.

Funding

CAMS Innovation Fund for Medical Sciences (CIFMS 2021-I2M-1-003).

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.990553/full#supplementary-material

References

1. Sheikh SN, Martin SB, Martin DL. Regional distribution and relative amounts of glutamate decarboxylase isoforms in rat and mouse brain. *Neurochem Int.* (1999) 35:73–80. doi: 10.1016/s0197-0186(99)00063-7

2. Patel AB, de Graaf RA, Martin DL, Battaglioli G, Behar KL. Evidence that GAD 65 mediates increased GABA synthesis during intense neuronal activity *in vivo*: GAD 65 mediated GABA synthesis during

seizures. *J Neurochem.* (2006) 97:385–96. doi: 10.1111/j.1471-4159.2006.03741.x

3. Kaufman DL, Houser CR, Tobin AJ. Two forms of the gamma-aminobutyric acid synthetic enzyme glutamate decarboxylase have distinct intraneuronal distributions and cofactor interactions. *J Neurochem.* (1991) 56:720–3. doi: 10.1111/j.1471-4159.1991.tb08211.x

4. Solimena M, Folli F, Denis-Donini S, Comi GC, Pozza G, De Camilli P, et al. Autoantibodies to glutamic acid decarboxylase in a patient with stiff-man syndrome, epilepsy, and type I diabetes mellitus. *N Engl J Med.* (1988) 318:1012–20. doi: 10.1056/NEJM198804213181602

5. Muñoz-Lopetegi A, de Bruijn MAAM, Boukhrissi S, Bastiaansen AEM, Nagtzaam MMP, Hulsenboom ESP, et al. Neurologic syndromes related to anti-GAD65: Clinical and serologic response to treatment. *Neurol Neuroimmunol Neuroinflamm.* (2020) 7:e696. doi: 10.1212/NXI.0000000000000696

6. Budhram A, Sechi E, Flanagan EP, Dubey D, Zekeridou A, Shah SS, et al. Clinical spectrum of high-titre GAD65 antibodies. *J Neurol Neurosurg Psychiatry.* (2021) 92:645–54. doi: 10.1136/jnnp-2020-325275

7. Kuo YC, Lin CH. Clinical spectrum of glutamic acid decarboxylase antibodies in a Taiwanese population. *Eur J Neurol.* (2019) 26:1384–90. doi: 10.1111/ene.14005

8. Fisher RS. The New classification of seizures by the international league against epilepsy 2017. *Curr Neurol Neurosci Rep.* (2017) 17:48. doi: 10.1007/s11910-017-0758-6

9. McKeon A, Robinson MT, McEvoy KM, Matsumoto JY, Lennon VA, Ahlskog JE, et al. Stiff-man syndrome and variants: clinical course, treatments, and outcomes. *Arch Neurol.* (2012) 69:230–8. doi: 10.1001/archneurol.2011.991

10. Falip M, Carreño M, Miró J, Saiz A, Villanueva V, Quílez A, et al. Prevalence and immunological spectrum of temporal lobe epilepsy with glutamic acid decarboxylase antibodies. *Eur J Neurol.* (2012) 19:827–33. doi: 10.1111/j.1468-1331.2011.03609.x

11. Tizazu E, Ellis CA, Reichert J, Lancaster E. Low rate of glutamic acid decarboxylase 65 (GAD-65) antibodies in chronic epilepsy. *Seizure.* (2020) 80:38–41. doi: 10.1016/j.seizure.2020.05.008

12. Daif A, Lukas RV, Issa NP, Javed A, VanHaerents S, Reder AT, et al. Antiglutamic acid decarboxylase 65 (GAD65) antibody-associated epilepsy. *Epilepsy & Behavior.* (2018) 80:331–6. doi: 10.1016/j.yebeh.2018.01.021

13. Lacruz Ballester L, Fernandez-Fournier M, Puertas Muñoz I, Rodriguez Fraga O, Lastras Fernandez-Escandon C, Rodriguez de. Rivera Garrido FJ, Alba Suarez EM, Tallon Barranco A. Serum glutamate decarboxylase antibodies and neurological disorders: when to suspect their association? *Neurol Sci.* (2022) 43:633–41. doi: 10.1007/s10072-021-05281-4

14. Dimova P, Minkin K. Case report: multisystem autoimmune and overlapping GAD65-antibody-associated neurological disorders with beneficial effect of epilepsy surgery and rituximab treatment. *Front Neurol.* (2021) 12:756668. doi: 10.3389/fneur.2021.756668

15. Pittock SJ, Yoshikawa H, Ahlskog JE, Tisch SH, Benarroch EE, Kryzer TJ, et al. Glutamic acid decarboxylase autoimmunity with brainstem, extrapyramidal, and spinal cord dysfunction. *Mayo Clin Proc.* (2006) 81:1207–14. doi: 10.4065/81.9.1207

16. Bárová H, Perusicová J, Hill M, Sterzl I, Vondra K, Masek Z. Anti-GAD-positive patients with type 1 diabetes mellitus have higher prevalence of autoimmune thyroiditis than anti-GAD-negative patients with type 1 and type 2 diabetes mellitus. *Physiol Res.* (2004) 53:279–86.

17. Haitao R, Huiqin L, Tao Q, Xunzhe Y, Xiaoqiu S, Wei L, et al. Autoimmune encephalitis associated with vitiligo? *J Neuroimmunol.* (2017) 310:14–6. doi: 10.1016/j.jneuroim.2017.05.019

18. Ariño H, Höftberger R, Gresa-Arribas N, Martínez-Hernández E, Armangue T, Kruer MC, et al. Paraneoplastic neurological syndromes and glutamic acid decarboxylase antibodies. *JAMA Neurol.* (2015) 72:874–81. doi: 10.1001/jamaneurol.2015.0749

19. Graus F, Saiz A, Dalmau J, GAD. antibodies in neurological disorders — insights and challenges. *Nat Rev Neurol.* (2020) 16:353–65. doi: 10.1038/s41582-020-0359-x

20. Fredriksen JR, Carr CM, Koeller KK, Verdoorn JT, Gadoth A, Pittock SJ, et al. findings in glutamic acid decarboxylase associated autoimmune epilepsy. *Neuroradiology.* (2018) 60:239–45. doi: 10.1007/s00234-018-1976-6

21. Seniaray N, Verma R, Ranjan R, Belho E, Mahajan H. 18F-FDG PET/CT in initial diagnosis and treatment response evaluation of anti-NMDAr and anti-GAD dual antibody autoimmune encephalitis. *Clin Nucl Med.* (2021) 46:e63–4. doi: 10.1097/RLU.0000000000003379

22. Mongay-Ochoa N, Sala-Padró J, Reynés-Llompart G, Rodríguez-Bel L, Jaraba S, Morandeira F, et al. Brain FDG-PET findings in glutamic acid decarboxylase antibody-associated epilepsy. *Journal of Neuroimaging.* (2021) 31:869–73. doi: 10.1111/jon.12874

23. Alexopoulos H, Dalakas MC. The immunobiology of autoimmune encephalitides. *J Autoimmun.* (2019) 104:102339. doi: 10.1016/j.jaut.2019.102339

24. Czempik PF, Gawryluk J, Wiórek A, Krzystanek E, Krzych ŁJ. Efficacy and safety of therapeutic plasma exchange in stiff person syndrome. *Open Medicine.* (2021) 16:526–31. doi: 10.1515/med-2021-0220

25. Jones AL, Flanagan EP, Pittock SJ, Mandrekar JN, Eggers SD, Ahlskog JE, et al. Responses to and outcomes of treatment of autoimmune cerebellar ataxia in adults. *JAMA Neurol.* (2015) 72:1304–12. doi: 10.1001/jamaneurol.2015.2378

26. Li T-R, Zhang Y-D, Wang Q, Shao X-Q, Li Z-M, Lv R-J. Intravenous methylprednisolone or immunoglobulin for anti-glutamic acid decarboxylase 65 antibody autoimmune encephalitis: which is better? *BMC Neurosci.* (2020) 21:13. doi: 10.1186/s12868-020-00561-9

27. Gagnon MM, Savard M. Limbic encephalitis associated with GAD65 antibodies: brief review of the relevant literature. *Can J Neurol Sci.* (2016) 43:486–93. doi: 10.1017/cjn.2016.13 Epub 2016 Mar 31.

28. Dalakas MC, Rakocevic G, Dambrosia JM, Alexopoulos H, McElroy B, A. double-blind, placebo-controlled study of rituximab in patients with stiff person syndrome. *Ann Neurol.* (2017) 82:271–7. doi: 10.1002/ana.25002

29. Thaler FS, Zimmermann L, Kammermeier S, Strippel C, Ringelstein M, Kraft A, et al. Rituximab treatment and long-term outcome of patients with autoimmune encephalitis: real-world evidence from the generate registry. *Neurol Neuroimmunol Neuroinflamm.* (2021) 8:e1088. doi: 10.1212/NXI.0000000000001088

30. Bacorro EA, Tehrani R. Stiff-person syndrome: persistent elevation of glutamic acid decarboxylase antibodies despite successful treatment with rituximab. *J Clin Rheumatol.* (2010) 16:237–9. doi: 10.1097/RHU.0b013e3181e931fa

31. Brunker L, Hirst P, Schlesinger JJ. New-onset refractory status epilepticus with underlying autoimmune etiology: a case report. *SN Compr Clin Med.* (2020) 2:103–7. doi: 10.1007/s42399-019-00185-z

32. Jaafar F, Haddad L, Koleilat N, Sharara-Chami R, Shbarou R. Super refractory status epilepticus secondary to anti-GAD antibody encephalitis successfully treated with aggressive immunotherapy. *Epilepsy Behav Rep.* (2020) 14:100396. doi: 10.1016/j.ebr.2020.100396

33. Saidha S, Murphy S, Ronayne A, McCarthy P, Hennessy MJ, Counihan T. Treatment of anti-glutamic acid decarboxylase antibody-associated limbic encephalitis with mycophenolate mofetil. *J Neurol.* (2010) 257:1035–8. doi: 10.1007/s00415-010-5476-9

34. Joubert B, Belbezier A, Haesebaert J, Rheims S, Ducray F, Picard G, et al. Long-term outcomes in temporal lobe epilepsy with glutamate decarboxylase antibodies. *J Neurol.* (2020) 267:2083–9. doi: 10.1007/s00415-020-09807-2

35. Chengyu L, Weixiong S, Chao C, Songyan L, Lin S, Zhong Z, et al. Clinical features and immunotherapy outcomes of anti-glutamic acid decarboxylase 65 antibody-associated neurological disorders. *J Neuroimmunol.* (2020) 345:577289. doi: 10.1016/j.jneuroim.2020.577289

36. Honnorat J, Saiz A, Giometto B, Vincent A, Brieva L, de Andres C, et al. Cerebellar ataxia with anti–glutamic acid decarboxylase antibodies: study of 14 patients. *Arch Neurol.* (2001) 58:225. doi: 10.1001/archneur.58.2.225

37. Blanc F, Ruppert E, Kleitz C, Valenti MP, Cretin B, Humbel RL, et al. Acute limbic encephalitis and glutamic acid decarboxylase antibodies: a reality? *J Neurol Sci.* (2009) 287:69–71. doi: 10.1016/j.jns.2009.09.004

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 25 October 2022
DOI 10.3389/fneur.2022.970168

OPEN ACCESS

EDITED BY
Ilka Kleffner,
University Hospital Bochum GmbH, Germany

REVIEWED BY
Erum Khan,
B.J. Medical College and Civil Hospital, India
Merja Hannele Soilu-Hänninen,
University of Turku, Finland
Francesca Gobbin,
University of Verona, Italy

*CORRESPONDENCE
Vincent van Pesch
vincent.vanpesch@saintluc.uclouvain.be

†These authors have contributed equally to this work and share last authorship

SPECIALTY SECTION
This article was submitted to Neuroepidemiology, a section of the journal Frontiers in Neurology

RECEIVED 15 June 2022
ACCEPTED 30 September 2022
PUBLISHED 25 October 2022

CITATION
Sambon P, Sellimi A, Kozyreff A, Gheysens O, Pothen L, Yildiz H and van Pesch V (2022) Epidemiology, clinical presentation, treatment, and outcome of neurosarcoidosis: A mono-centric retrospective study and literature review. *Front. Neurol.* 13:970168. doi: 10.3389/fneur.2022.970168

Epidemiology, clinical presentation, treatment, and outcome of neurosarcoidosis: A mono-centric retrospective study and literature review

Pauline Sambon[1], Amina Sellimi[2], Alexandra Kozyreff[3], Olivier Gheysens[4], Lucie Pothen[1], Halil Yildiz[1†] and Vincent van Pesch[2*†]

[1]Department of Internal Medicine and Infectious Diseases, Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Brussels, Belgium, [2]Department of Neurology, Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Brussels, Belgium, [3]Department of Ophthalmology, Cliniques Universitaires Saint-Luc, Université Catholique de Louvain, Brussels, Belgium, [4]Department of Nuclear Medicine, Cliniques Universitaires Saint-Luc and Institute of Clinical and Experimental Research (IREC), Université Catholique de Louvain, Brussels, Belgium

Introduction: Neurosarcoidosis is a rare granulomatous disorder, and treatment guidelines are mainly based on retrospective studies.

Materials and methods: This retrospective study was performed to provide a detailed description of the clinical characteristics and treatment outcomes of patients with neurosarcoidosis followed at Cliniques Universitaires Saint Luc in Belgium. The second objective of our study was to perform a comparative literature review of neurosarcoidosis, with a focus on treatment outcomes with the use of TNF-α antagonist.

Results: Among 180 patients with sarcoidosis followed in our hospital, 22 patients with neurosarcoidosis were included in the final analysis. Our literature research identified 776 articles of which 35 articles met our inclusion criteria, including 1,793 patients diagnosed with neurosarcoidosis. In our cohort, the majority of patients (86%) were diagnosed with systemic sarcoidosis which was similar to that reported in the literature (83%). Serum CRP and calcemia were elevated only in 33 and 18% of patients, respectively. Serum lysozyme and angiotensin-converting enzyme were elevated in 79 and 16% of patients, respectively. Lumbar puncture and CSF fluid analysis were performed in 15/22 patients and were abnormal in all patients. Brain MRI was performed in 21/22 patients and showed abnormalities in 16 patients consisting of parenchymal lesions in 63%, hypothalamic-pituitary axis lesions in 38%, and meningeal enhancement in 31%. In both cohort patients, methotrexate was the most frequently used treatment (>45% of cases) with a favorable outcome in an average of 50% of patients. A TNF-α antagonist was administered in 9% of patients in our cohort and in 27% of patients in the literature review. The proportion of favorable outcomes in literature research was significantly higher in patients treated with TNF-α antagonists compared to methotrexate ($p < 0.0001$), mycophenolate mofetil ($p < 0.0001$), or azathioprine ($p < 0.0001$).

Conclusion: The results of our cohort and literature review confirm that neurosarcoidosis occurred most frequently in the context of systemic sarcoidosis. Methotrexate is the most frequent second-line therapy. The effectiveness of therapy with TNF-α antagonists is well-demonstrated and associated with a better outcome. Their earlier use during the disease course among aggressive and/or refractory neurosarcoidosis should be considered.

KEYWORDS

sarcoidosis, neurosarcoidosis, methotrexate, azathioprine, TNF-α antagonist, outcome

Introduction

Sarcoidosis is a systemic inflammatory disorder characterized by non-caseating granulomatous lesions. Although all organs may be affected, it occurs most frequently (>90% of cases) in lymph nodes, particularly mediastinal; but also in lungs, skin, and eyes (e.g., uveitis) (1). Skin involvement (lupus pernio, cutaneous granuloma, erythema nodosum, and subcutaneous nodules) occurs in 30% of patients (2, 3). Liver and spleen lesions are found in 5–15% of patients undergoing computed tomography (4, 5). Cardiac, bone, and neurological involvement are also possible but less frequent. However, cardiac involvement can be life-threatening and is the second cause of death from sarcoidosis after pulmonary involvement (1). Neurosarcoidosis is also an important cause of morbidity and mortality, especially in young patients (6–9), and occurs in 5–20% of patients (10). Neurosarcoidosis may affect cranial/peripheral nerves, brain, leptomeninges, spinal cord, and muscles (10–12). Clinical presentations are various; including facial nerve palsy, optic neuritis, aseptic meningitis, and lesions of the central nervous system inducing focal neurological deficits, hydrocephalus, encephalopathy, psychosis, peripheral neuropathy, and myopathy (9–12). Neurosarcoidosis is often associated with systemic sarcoidosis but isolated neurosarcoidosis is also described (6–10).

The diagnosis of sarcoidosis and especially neurosarcoidosis is challenging. There are many alternative causes of granulomatosis such as infection (e.g., mycobacterium tuberculosis), inflammatory diseases (e.g., inflammatory bowel diseases, granulomatosis with polyangiitis), and lymphoma (e.g., Hodgkin's lymphoma) which must be ruled out (9). A comprehensive diagnostic workup is necessary and a tissue biopsy is often required to confirm the diagnosis (9, 13). The diagnostic criteria of sarcoidosis, which have been recently updated (14), are based on the combination of a compatible clinical presentation, the presence of non-necrotizing granulomatous inflammation, and the exclusion of other causes of granulomatous diseases. Recently, the Neurosarcoidosis Consortium Consensus Group (NCCC) proposed new diagnostic criteria, to optimize the diagnosis of neurosarcoidosis and to enhance the clinical care of patients with suspected neurosarcoidosis (13). According to these criteria, the diagnosis of neurosarcoidosis is classified as follows: (i) possible when there are compatible clinical and radiological features without pathologic confirmation, (ii) probable when there is a pathologic confirmation of systemic granulomatous disease, and (iii) definite when there is a nervous system biopsy consistent with neurosarcoidosis (with or without systemic sarcoidosis) (13).

Treatment guidelines for neurosarcoidosis are mainly based on small cohort studies and non-randomized clinical trials, as there is a lack of robust randomized clinical trials. First-line treatment consists of corticosteroid therapy followed by methotrexate, azathioprine, or mycophenolate mofetil as corticosteroid-sparing second-line therapy. Cyclophosphamide has been used in the past for treating refractory sarcoidosis but is nowadays less considered due to its potential heavy side effects (bone marrow suppression, infection, infertility, hemorrhagic cystitis, and malignancy). Cyclosporine A has also been used but should not be preferred due to its safety profile (high blood pressure, renal impairment, and tremor) (15–17).

Based on the role of tumor necrosis factor-alpha (TNF-α) in autoimmune disease, anti-TNF-α monoclonal antibodies have been used as a novel therapeutic approach and are associated with favorable results in many diseases such as rheumatoid arthritis, psoriatic arthritis, inflammatory bowel disease, and non-infectious uveitis (18), as well as in systemic sarcoidosis (19–23). Recent studies provided class IV evidence that TNF-α antagonists are also beneficial in neurosarcoidosis (16, 24–26). They are currently proposed as third-line therapy in the management of aggressive and/or refractory neurosarcoidosis (10, 15, 27–38).

The objectives of our study are to describe the clinical and paraclinical features of neurosarcoidosis patients followed in a single Belgian academic center and to perform a comparative literature review of neurosarcoidosis, with a focus on treatment outcomes, in particular with the use of TNF-α antagonists.

Materials and methods

Patient selection and inclusion criteria

The study was conducted at Cliniques Universitaires Saint-Luc, UCLouvain (Belgium). All files of adult sarcoidosis

followed in the departments of Internal Medicine and Neurology until March 2022 were retrospectively reviewed. Only patients diagnosed with possible, probable, or definite neurosarcoidosis (both central and peripheral neurosarcoidosis) according to the Neurosarcoidosis Consortium Consensus Group's 2018 Diagnostic Criteria (13) were included for final analysis.

Data collection

Data were extracted from each patient's clinical records and reviewed by HY and PS to confirm the diagnosis of neurosarcoidosis, according to Neurosarcoidosis Consortium Consensus Group's criteria. Data on baseline characteristics, demographic features, clinical manifestations, history of systemic and neurologic sarcoidosis, biological (serum and cerebrospinal fluid), radiological (spinal cord MRI, brain MRI, [18F]FDG-PET/CT, thoraco-abdominal CT scan, chest x-ray), histological and electromyography results, treatment regimens, disease course and outcome were systematically collected for all patients.

The baseline was defined as the date of neurosarcoidosis diagnosis. Biopsy-confirmed sarcoidosis was defined by the presence of non-caseating granulomas (13, 24). Duration of follow-up was defined as the time between neurosarcoidosis diagnosis and the most recent clinical assessment. Therapies were classified as first-, second-, and third-lines. First-line therapy consists of corticosteroid treatment, second-line therapy consists of immunosuppressive therapy with methotrexate, azathioprine, mycophenolate mofetil, cyclosporine A, or (hydroxy)-chloroquine, and third-line therapy either consists of cyclophosphamide or monoclonal antibodies (TNF-α inhibitors or B-cell targeted therapy) (10, 24).

Based on clinical and/or radiological features, treatment response and outcomes were classified as ≪ complete remission ≫, ≪ partial remission ≫, ≪ clinically and/or radiologically active disease≫, ≪ progressive disease ≫, ≪ relapse ≫ or ≪ mortality ≫. Favorable outcomes include complete and partial remission which were defined, respectively, by the absence or conversely the presence of residual symptoms, without the need for alternative immunosuppressive therapy (10). Relapse or progression was defined as clinical and/or radiological worsening, either subacute or chronic, due to either neurological or systemic manifestations of sarcoidosis requiring a therapeutic modification (25). Relapse was defined as reoccurrence during a stable phase or appearance of a new localization, while progression as slow worsening of residual symptoms (4).

Ethical consideration

This study was approved by the local Ethics Committee of the Cliniques Universitaires Saint-Luc (Brussels, CEHF 2021/29OCT/452-SARCO2). No written consent form was required given the retrospective nature of the study.

Statistical analysis

Quantitative variables were reported as median values with ranges while qualitative values were shown as numbers and percentages. Treatment responses were compared using Fisher's exact test for categorical variables.

Literature review

A comprehensive literature search was manually performed by searching the Pubmed/MEDLINE databases until 20 April 2022. We used the following terms: neurosarcoidosis OR (nervous system AND (sarcoidosis OR granulomatous disease OR sarcoid granuloma) AND (tumor necrosis factor OR TNF-alpha OR infliximab OR adalimumab OR certolizumab OR golimumab OR etanercept OR azathioprine OR methotrexate OR mycophenolate mofetil OR chloroquine OR cyclosporine OR cyclophosphamide OR rituximab OR thalidomide OR chlorambucil). Studies written in English or French were considered for inclusion, without date range restrictions. Original research articles were included if they reported at least five cases of possible, probable, or definite neurosarcoidosis, treated with second-line or third-line therapies. Studies were excluded if they reported pediatric cases or patients only treated with first-line therapy consisting of corticosteroid treatment. We conducted a second and manual search in the reference lists of the included articles. The title and abstract of the studies were independently screened by two reviewers (HY and PS) to ensure eligibility for inclusion. The flow diagram of included studies is shown in Figure 1. A pooled analysis of all available data was performed. The results are presented as the number for which the data are present out of the total number of patients for which the data were described [n/N *(%)*].

Results

Baseline characteristics

Among the 180 adult sarcoidosis patients followed in our tertiary center, 25 were identified as having neurosarcoidosis and 22 were included in the final analysis. Three patients were excluded: one patient had ocular and lymph node sarcoidosis with myopathy explained by concomitant myasthenia confirmed by neuromuscular biopsy, one had a diffuse glioneuronal tumor on brain biopsy, and one had altered consciousness explained by hypercalcemia without evidence

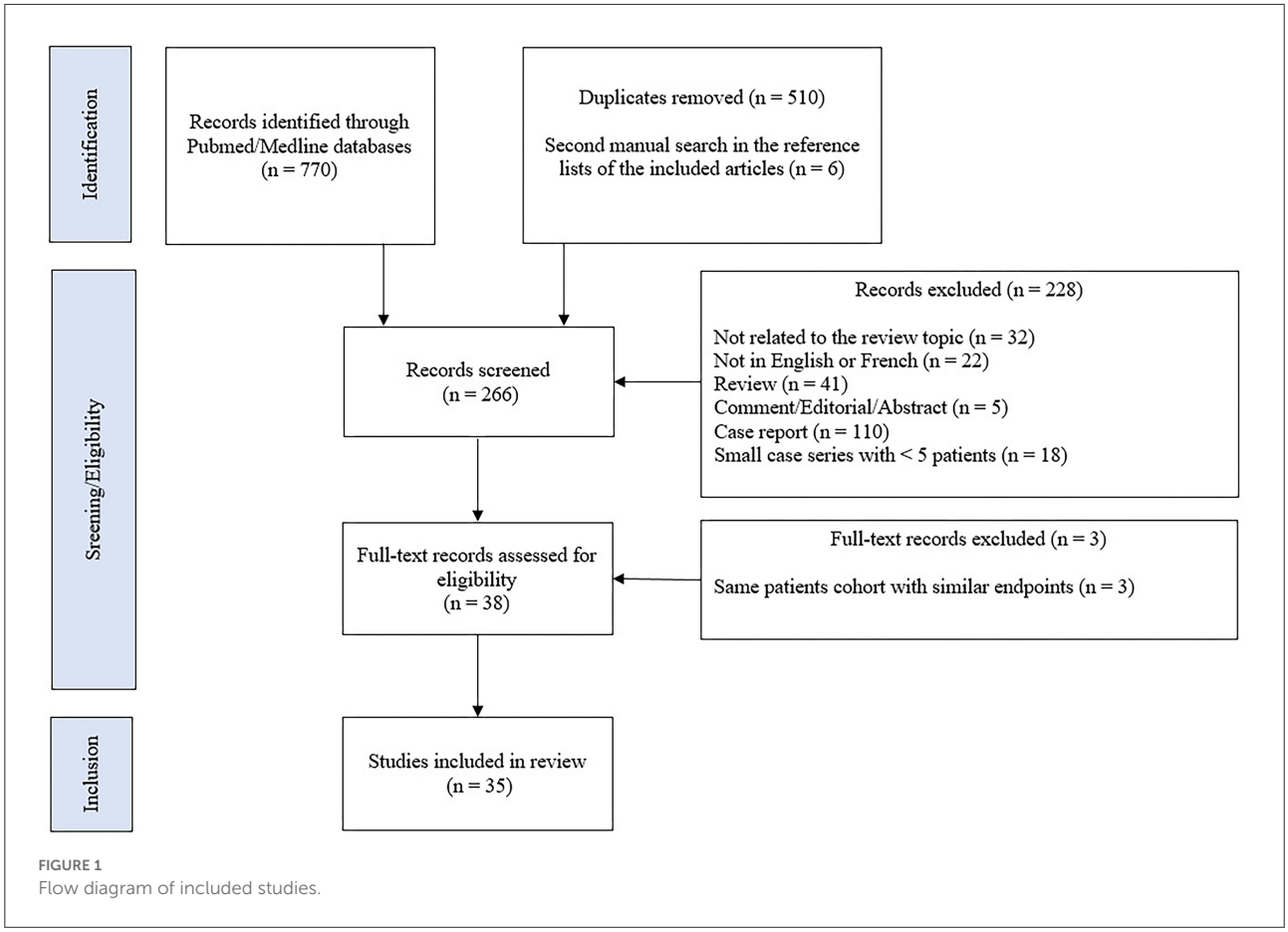


FIGURE 1
Flow diagram of included studies.

of neurosarcoidosis. Patient characteristics are presented in Table 1 and Supplementary Table 1. Fourteen (64%) were male. The median age at the time of neurosarcoidosis diagnosis was 40.5 years (range 22–67) and the median time from onset of symptoms to diagnosis was 4 months (range 1–23). Except for one patient who met possible diagnostic criteria, all patients had histologically proven sarcoidosis from lymph nodes ($n = 16$), salivary glands ($n = 1$), spleen ($n = 1$), liver ($n = 1$), pituitary glands ($n = 2$), or brain parenchyma ($n = 3$) biopsy. Five (23%) patients were classified as having definite neurosarcoidosis and 16 (73%) with probable neurosarcoidosis. The median duration of follow-up was 3.6 years (range 0.2–17.4).

Clinical characteristics

Clinical characteristics are reported in Table 2. Nineteen patients (86%) were diagnosed with systemic sarcoidosis, either before ($n = 3$) or concomitantly ($n = 16$) to the neurological involvement. Systemic sarcoidosis mainly consisted of lymph node, lung, and articular involvement; followed by ocular, splenic, salivary gland, skin, hepatic, and bone involvement (Figure 2). Ten (45%) patients had involvement of at least three systemic organs, while 5 (23%) had only lymph node involvement. Systemic symptoms mostly consisted of fatigue (36%) and arthralgia (36%), followed by weight loss, visual symptoms (diplopia, blurred vision), dyspnea, cough, and fever. Presenting neurological symptoms varied largely and consisted of a majority of headache (41%) and gait abnormalities (41%); followed by sensory abnormalities, including hypoesthesia, paresthesia, and neuropathic pain; and micturition abnormalities. The most commonly affected neurological site consisted of meningeal involvement (64%), including aseptic meningitis, leptomeningitis, and pachymeningitis, as well as parenchymal disease (45%), cranial nerve neuropathy (36%), and spinal cord involvement (32%) (Figure 3). Other neurosarcoidosis sites included the hypothalamic-pituitary axis, peripheral neuropathy, myopathy, and vascular disease, including ischemic and hemorrhagic stroke. Seventeen (77%) patients had multiple neurological involvement sites. Hypothalamic-pituitary axis involvement and aseptic meningitis were the unique manifestation in one and three patients, respectively, whereas isolated cranial neuropathy and hydrocephalus were not observed.

TABLE 1 Baseline characteristics of our patient cohort diagnosed with neurosarcoidosis.

Cases	Sex, age (years)	Ethnicity	History of sarcoidosis	Systemic involvement	Neurological involvement	Neuro-sarcoidosis	Abnormal Brain MRI	Abnormal Spinal cord MRI	Abnormal FDG-PET/CT	Biopsy site	Biopsy results
1	M, 53	C	No	LN, E, J, S	M, P, NE, SC, PN	Probable	Yes	Yes	Yes	Spleen	+
2	M, 27	N-A	No	LN	NE	Definite	Yes	NA	Yes	Pituitary gland	+
3	M, 67	A	No	L, LN, J	M, P, NE	Probable	Yes	No	Yes	LN	+
4	M, 40	C	No	No	CN, P	Definite	Yes	No	No	Brain parenchyma	+
5	M, 38	N-A	No	L, LN, J	M	Probable	No	NA	Yes	LN	+
6	M, 35	N-A	No	LN	SC	Probable	NA	Yes	Yes	LN	+
7	M, 55	A	No	L, LN, E, SG	CN, M, P	Possible	Yes	NA	NA	LN/Salivary glands	–/–
8	F, 22	A	No	LN, E	CN, M	Probable	Yes	NA	Yes	LN	+
9	M, 42	C	Yes	L, LN, C, B, H, S	NE	Probable	Yes	NA	Yes	LN/Liver	+/+
10	F, 26	C	No	No	NE	Definite	Yes	NA	No*	LN/Pituitary gland	Lymphoma/+
11	M, 32	C	No	No	M, P, NE	Definite	Yes	NA	No	Brain parenchyma	+
12	M, 41	C	No	LN	SC, PN, V	Probable	Yes	Yes	Yes	LN	+
13	M, 38	C	No	LN	M, P, SC	Probable	Yes	Yes	NA	LN	+
14	M, 49	C	Yes	L, LN, J	M, SC, My	Probable	No	Yes	Yes	LN	+
15	F, 40	C	Yes	L, LN, E, SG	CN, M, P	Probable	No	No	Yes	LN/Salivary glands	+/–
16	F, 39	N-A	No	LN, S	CN, P, V	Probable	Yes	NA	Yes	LN	+
17	F, 43	C	No	L, LN	M, P	Definite	Yes	No	Yes	LN/Brain parenchyma	+/+
18	F, 59	C	No	L, LN, C, E, J, S	CN, M, PN, My	Probable	Yes	NA	Yes	LN	+
19	F, 47	C	No	LN, J	M, P, SC	Probable	Yes	Yes	Yes	LN	+
20	F, 58	C	No	LN, E, J	CN, M, NE, SC	Probable	Yes	Yes	Yes	LN	+
21	M, 42	A	No	LN	PN, My	Probable	No	No	Yes	LN/Salivary gland/Skin	+/–/–
22	M, 25	C	No	LN, SG, H	CN, M, PN	Probable	No	No	Yes	LN/Salivary glands	+/+

NA, Not available; F, Female; M, Male; C, Caucasian; A, African; N-A, North-African; MRI, magnetic resonance imaging; EMG, Eletromyography; CSF, Cerebrospinal fluid; +, biopsy consistent with sarcoidosis; −, biopsy inconsistent with sarcoidosis; ACE, angiotensin-converting enzyme; L, Lungs; LN, Lymph nodes; C, Cutaneous; E, Eye; SG, Salivary glands; J, Joints; B, Bones; H, Hepatic; S, Spleen; H, Heart; CN, Cranial neuropathy; M, Meningeal involvement; P, Parenchymal disease; Hy, Hydrocephalus; NE, Neuro-endocrine; V, Vascular disease; SP, Spinal cord disease; PN, Peripheral neuropathy, My, Myopathy.
*Axillary and mediastinal lymph nodes attributed to lymphoma (confirmed by biopsy).

10.3389/fneur.2022.970168

10.3389/fneur.2022.970168

TABLE 2 Baseline and clinical characteristics of patients from our cohort and the literature.

		Our cohort	Literature review
Number of cases		22	1,793
Age at neurosarcoidosis diagnosis (years), median (range)		40.5 (22–67)	41.5 (26–70)*
Sex			
	Male, n/N (%)	14 (64)	777/1,682 (46)
	Female, n/N (%)	8 (36)	905/1,682 (54)
Ethnicity			
	Caucasian, n/N (%)	14 (64)	802/1,239 (65)
	African/North-African, n/N (%)	8 (36)	264/1,239 (21)
	Other, n/N (%)	0 (0)	135/1,239 (11)
	Unknown, n/N (%)	0 (0)	38/1,239 (3)
Neurosarcoidosis classification			
	Possible, n/N (%)	1 (5)	187/1,385 (13)
	Probable, n/N (%)	16 (73)	853/1,291 (66)
	Definite, n/N (%)	5 (23)	354/1,622 (23)
Isolated neurosarcoidosis, n/N (%)		3 (14)	220/1,331 (17)
Systemic sarcoidosis, n/N (%)		19 (86)	1,111/1,331 (83)
	History of systemic sarcoidosis, n/N (%)	3 (16)	214/637 (34)
	Systemic sarcoidosis at baseline, n/N (%)	16 (84)	179/578 (31)
	Primary neurological presentation, n/N (%)	0 (0)	220/1,331 (17)
Site of systemic involvement			
	Lymph nodes, n/N (%)	19 (86)	485/907 (53)
	Lungs, n/N (%)	8 (36)	568/907 (62)
	Ear-Nose-Throat, n/N (%)	0 (0)	69/907 (8)
	Salivary glands, n/N (%)	3 (14)	14/907 (2)
	Eye, n/N (%)	6 (27)	216/907 (24)
	Heart, n/N (%)	0 (0)	113/907 (12)
	Joints, n/N (%)	7 (32)	105/907 (12)
	Bones, n/N (%)	1 (5)	12/907 (1)
	Skin, n/N (%)	2 (9)	147/907 (16)
	Spleen, n/N (%)	4 (18)	47/907 (5)
	Liver, n/N (%)	2 (9)	89/907 (10)
	Kidney, n/N (%)	0 (0)	18/907 (2)
	Digestive tract, n/N (%)	0 (0)	12/907 (1)
	Scrotal, n/N (%)	0 (0)	4/907 (0.4)
Site of neurological involvement			
	Cranial neuropathy, n/N (%)	8 (36)	498/1,518 (33)
	Meningeal involvement, n/N (%)	14 (64)	722/1,507 (48)
	Parenchymal disease, n/N (%)	10 (45)	622/1334 (47)
	Hydrocephalus, n/N (%)	0 (0)	40/1,237 (3)
	Hypothalamic/pituitary axis, n/N (%)	6 (27)	162/1,237 (13)
	Vascular disease, n/N (%)	2 (9)	24/1,237 (2)
	Myelopathy/spinal cord involvement, n/N (%)	7 (32)	426/1,237 (34)
	Myopathy, n/N (%)	3 (14)	94/1237 (8)
	Peripheral neuropathy, n/N (%)	5 (23)	159/1,352 (12)

n/N represents the number for which the data are present out of the total number of patients for which the data were described.
*Available data (n/N = 1,545/1,793).

Biological and radiological characteristics

Results of ancillary investigations at diagnosis are summarized in Table 3. Serum CRP and calcemia were elevated in 33 and 18% of patients, respectively. Serum lysozyme was elevated in 79% of patients compared to 16% with increased serum angiotensin-converting enzyme (ACE) levels. Lumbar puncture and CSF fluid analysis were performed in 15/22 patients and were abnormal in all patients. Pleocytosis (cell count $> 5/mm^3$) was found in 80%, increased proteinorrhachia (>40 mg/dl) indicating blood-brain barrier dysfunction in 71%, low glucose levels in 9%, and CSF-specific IgG oligoclonal bands in 38% of patients. Brain MRI showed abnormalities in 16/21 (76%) patients mainly consisting of parenchymal lesions (63%), hypothalamic-pituitary axis lesions (38%), and meningeal enhancement (31%). Lesions were localized in the temporal lobe in 25% of patients. Spinal cord MRI showed abnormalities in 7/13 (54%) patients, revealing either longitudinally extensive myelitis or multiple disseminated spinal cord lesions. Lesions were predominantly located in the thoracic (86%), followed by the cervical (57%) and lumbar (29%) spine. [18F]FDG-PET/CT revealed systemic or neurosarcoidosis in 17/20 (85%) patients. Forty-one percent of these patients had a previously negative chest X-ray or thoracic CT scan.

Treatment

Detailed treatments of patients are reported in Table 4. Initial therapy consisted of corticosteroids in all except in

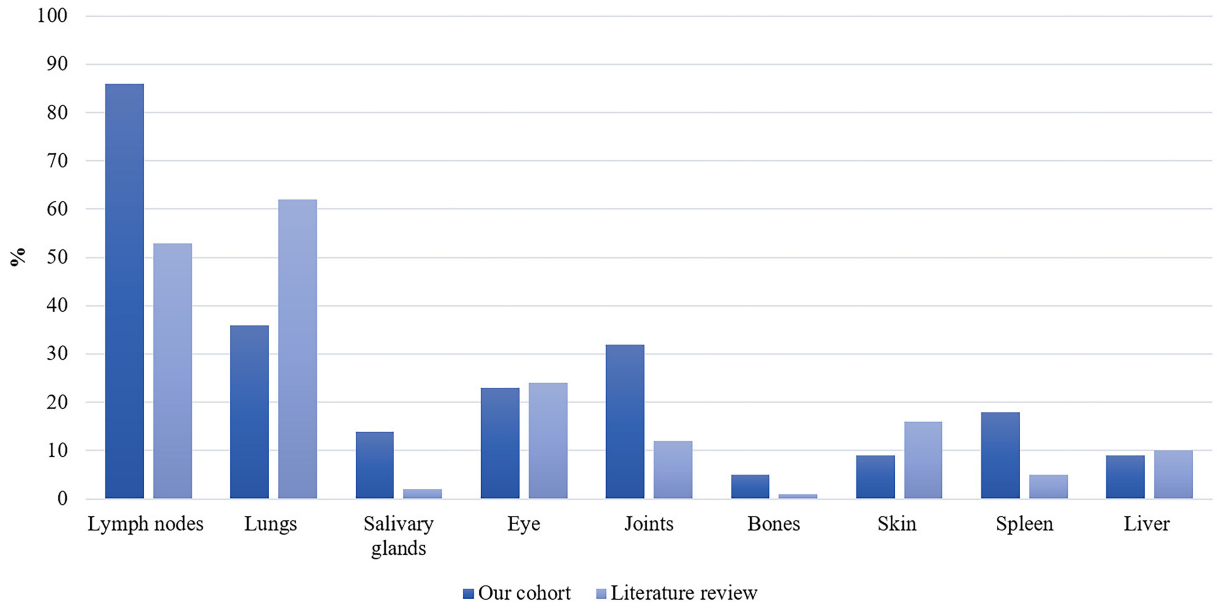


FIGURE 2
Proportion of systemic sarcoidosis involvement in our patient cohort (n = 22) and the literature (n/N = 907/1,793) expressed as percentages.

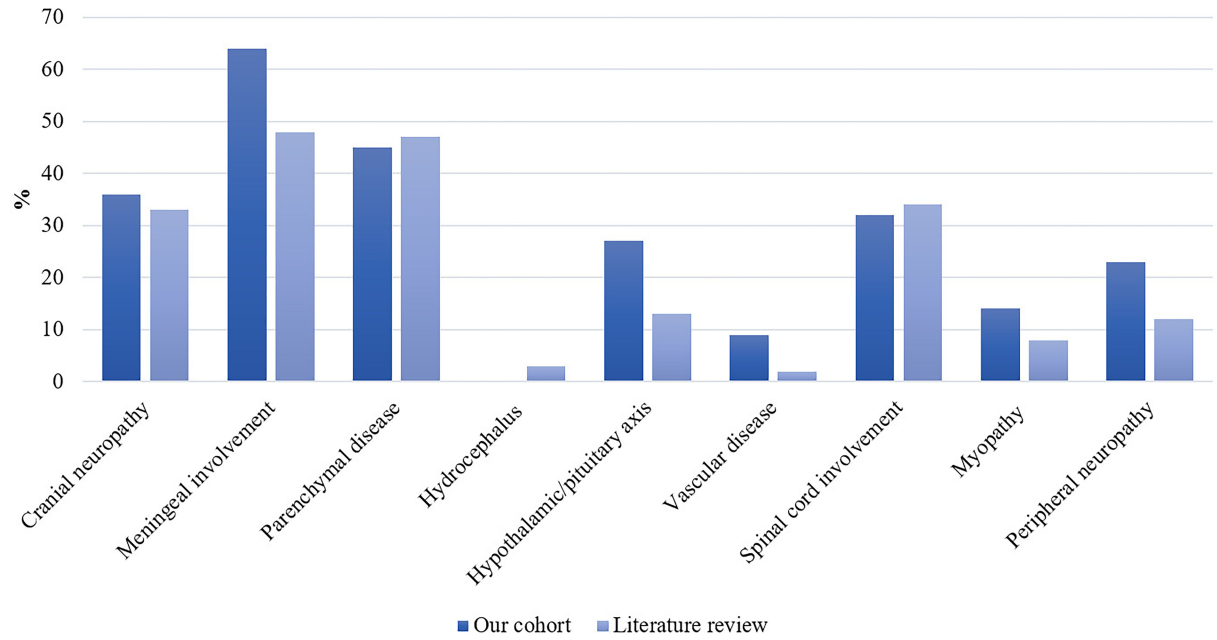


FIGURE 3
Distribution of neurological site involvement of neurosarcoidosis in our patient cohort (n = 22) and the literature as expressed as percentages. In the literature: n/N = 498/1,518 for cranial neuropathy; n/N = 722/1,507 for meningeal involvement; n/N = 622/1,334 for parenchymal disease; n/N = 40/1,237 for hydrocephalus; n/N = 162/1,237 for hypothalamic/pituitary axis; n/N = 24/1,237 for vascular disease; n/N = 426/1,237 for spinal cord involvement; n/N = 94/1,237 for myopathy; and n/N = 159/1,353 for peripheral neuropathy.

two patients; one was treated with methotrexate alone and one with ABVD (Doxorubicin, Bleomycin, Vinblastine, and Dacarbazine) for concomitant mediastinal lymphoma. Sixteen (73%) patients received second-line and 5 (23%) required intensification of treatment to third-line therapies (Table 5). Second-line therapy consisted of methotrexate in more

10.3389/fneur.2022.970168

TABLE 3 Paraclinical features of patients from our cohort and the literature at the time of neurological disease onset.

		Our cohort	Literature review
Serum analysis			
	CRP (mg/dl), median (range)	3.85 (0.1–71)	NA
	CRP increased, n/N (%)	7/21 (33)	7/21 (33)
	ACE (UECA), median (range)	51 (26–75)	73 (12–293)
	ACE increased, n/N (%)	3/19 (16)	311/839 (37)
	Lysozyme (mg/l), median (range)	23.9 (8.4–66)	35 (28–48)
	Lysozyme increased, n/N (%)	11/14 (79)	12/26 (46)
	Calcium (mmol/l), median (range)	2.45 (2.13–2.71)	2.39 (2.31–2.47)
	Calcium increased, n/N (%)	4/22 (18)	17/299 (59)
	Abnormal protein electrophoresis, n/N (%)	2/13 (15)	NA
Cerebrospinal fluid analysis			
	Lumbar puncture performed, n/N (%)	15/22 (68)	–
	White cell count (cells/mm^3), median (range)	15 (2–33)	40 (0–648)
	Pleiocytosis, n/N (%)	12/15 (80)	528/832 (63)
	Protein (mg/dl), median (range)	58 (26–1,186)	105 (41–980)
	Proteinorachy, n/N (%)	10/14 (71)	563/807 (70)
	Hypoglycorachy, n/N (%)	1/11 (9)	123/371 (33)
	Increased IgG index, n/N (%)	1/8 (13)	18/49 (37)
	Oligoclonal bands present, n/N (%)	5/13 (38)	78/339 (23)
	Normal, n/N (%)	0/15 (0)	14/143 (10)
Abnormal imaging investigation			
	Chest X-ray, n/N (%)	2/9 (22)	130/267 (49)
	Thoracic CT scan, n/N (%)	6/7 (86)	58/118 (49)
	Abdnominal CT scan, n/N (%)	3/5 (60)	NA
	Brain CT scan, n/N (%)	0/1 (0)	20/37 (54)
	Brain MRI, n/N (%)	16/21 (76)	570/752 (76)
	Spinal MRI, n/N (%)	7/13 (54)	326/538 (61)
	[F18]FDG-PET CT, n/N (%)	17/20 (95)	137/319 (43)
	EMG, n/N (%)	4/10 (40)*	NA
Detailed abnormal brain MRI			
	Parenchymal lesions, n (%)	10/16 (63)	135/291 (46)
	Meningeal enhancement, n (%)	5/16 (31)	102/264 (39)
	Mass lesion, n (%)	2/16 (13)	7/82 (9)
	Cranial nerve enhancement, n (%)	3/16 (19)	21/151 (14)
	Hypothalamus/pituiary axis lesions, n (%)	6/16 (38)	39/169 (23)
	Vascular lesions, n (%)	2/16 (13)	NA
	Parietal location, n (%)	1/16 (6)	NA
	Temporal location, n (%)	4/16 (25)	NA
	Gadolinium enhancement, n (%)	8/16 (50)	69/132 (52)
Detailed abnormal spinal cord MRI			
	Longitudinally extensive myelitis, n (%)	3/7 (43)	66/137 (48)
	Multiple separated spinal cord lesions, n (%)	2/7 (29)	14/60 (23)
	Lesion location: cervical spine, n (%)	4/7 (57)	43/84 (51)
	Lesion location: thoracic spine, n (%)	6/7 (86)	38/67 (57)
	Lesion location: lumbar spine, n (%)	2/7 (29)	5/27 (19)
	Gadolinium enhancement, n (%)	7/7 (100)	101/140 (72)

n/N represents the number for which the data are present out of the total number of patients for which the data are described. NA, Not available; ACE, Angiotensin-converting enzyme; MRI, magnetic resonance imaging; EMG, electromyography.
*Abnormal EMG due to concomitant myasthenia (n = 1).

than 50% of patients, followed by mycophenolate mofetil (18%), azathioprine (9%), and hydroxychloroquine (9%) (Figure 4). Methotrexate had a favorable outcome in 67% and azathioprine in 50% of patients, while hydroxychloroquine and mycophenolate mofetil in 100% of cases (Figure 5). A TNF-α antagonist was administered in 2 (9%) patients with 100% favorable outcomes. It was discontinued in 1/2 of patient because of remission and no relapse occurred following TNF-α antagonist discontinuation. Other treatment modalities consisted of hormonal substitution, anti-epileptic medication, and cervical decompression neurosurgery. The median cumulative dose of corticosteroids was 10.4 g, ranging from 2.9 to 33 g. At last follow-up, 75% of patients were corticosteroid-free. Eight patients (36%) experienced adverse events.

TABLE 4 Treatment and clinical outcomes of our patient cohort.

Cases	Initial therapy	Maintenance immunosuppressive therapy	Cumulative corticosteroids dose (mg)	Relapse and/or deterioration during disease course	Adverse events	Other treatment modalities	Outcomes at last follow-up visite	Follow-up (years)
1	Bolus CS	CYC, MMF	21,880	Yes	Yes	Hormonal substitution	Mortality**	1.4
2	Bolus CS	MTX	15,570	Yes	No	Hormonal substitution	Complete remission	3.1
3	Bolus CS	None	14,181	No	No	Hormonal substitution	Complete remission	4.2
4	Bolus CS	MTX, INF	21,109	Yes	Yes	Anti-epileptic medication	Partial remission	3.3
5	Bolus CS	HDQ, MTX	5,214	Yes	No	None	Complete remission	4.4
6	CS	None	NA	Yes	No	None	Partial remission	4.2
7	Bolus CS	None	10,314	No	Yes	None	Complete remission	17.4
8	CS	AZA, MMF, RTX	3,912	Yes	No	None	Complete remission	4
9	CS	None	3,922	No	No	None	Complete remission	4.3
10	ABVD*	None	NA	No	No	Hormonal substitution	Complete remission	3
11	CS	MTX	2,880	Yes	No	None	Partial remission	2.2
12	Bolus CS	MTX	33,050	Yes	Yes	Cervical decompression surgery	Progressive disease	6.3
13	Bolus CS	MTX	NA	No	No	None	Partial remission	8.6
14	Bolus CS	MTX, INF	11,952	Yes	Yes	None	Partial remission	7.8
15	CS	HDQ	9,768	Yes	No	None	Complete remission	6.1
16	CS	MTX	NA	Yes	Yes	None	Partial remission	2.4
17	MTX	MTX, RTX	NA	Yes	No	Pyridostigmin***	Partial remission	1.6
18	Bolus CS	MTX	10,415	Yes	Yes	None	Partial remission	1.3
19	CS	AZA, MMF	7,037	No	Yes	None	Partial remission	1
20	CS	MMF, MTX	18,079	Yes	Yes	None	Partial remission	3.8
21	CS	MTX	Lost to follow-up	Lost to follow-up	No	None	Lost to follow-up	0.2
22	CS	None	Lost to follow-up	Lost to follow-up	No	None	Lost to follow-up	0.3

NA, Not available; CS, Corticosteroids; MTX, Methotrexate; CYC, Cyclophosphamide; MMF, Mycophenolate Mofetil; INF, Infliximab; HCQ, Hydroxychloroquine; AZA, Azathioprine; RTX, Rituximab.
*ABVD (Doxorubicin, Bleomycin, Vinblastine, Dacarbazine) for concomitant lymphoma.
**Mortality unrelated to neurosarcoidosis.
***For concomitant myasthenia.

10.3389/fneur.2022.970168

TABLE 5 Detailed treatment of our patient cohort.

No treatment, *n* (%)	0 (0)
First line therapy, *n* (%)	21 (95)
Second line therapy, *n* (%)	16 (73)
Third line therapy, *n* (%)	5 (23)
Detailed treatment	
Corticosteroids, *n* (%)	21 (95)
Methotrexate, *n* (%)	12 (55%)
Azathioprine, *n* (%)	2 (9)
Hydroxychloroquine, *n* (%)	2 (9)
Mycophenolate Mofetil, *n* (%)	4 (18)**
Ciclosporine, *n* (%)	0 (0)
Cyclophosphamide, *n* (%)	1 (5)
Rituximab, *n* (%)	2 (9)
TNA-α antagonist, *n* (%)	2 (9)*
Treatment switches	
First to second or third line, *n* (%)	16 (73)
Second to third line, *n* (%)	4 (18)
Third to second line, *n* (%)	1 (5)
Between second line, *n* (%)	4 (18)
Between third line, *n* (%)	0 (0)
Other treatment modalidites, *n* (%)	6 (27)
Hormonal substitution, *n* (%)	4 (18)
Anti-epileptic medication, *n* (%)	1 (5)
Neurochirurgical intervention, *n* (%)	1 (5)
Corticosteroids-free at last-follow-up, *n* (%)	15 (75)
Corticosteroids cumulative dose (g), *n* (%)	10.4 (2.9–33)
Adverse events, *n* (%)	8 (36)
Follow-up (years), median (range)	3.6 (0.2–17.4)

*Infliximab ($n = 2$).
**One patient was treated twice with mycophenolate mofetil.

Outcome

Two patients were lost to follow-up, and thus were not included in the outcome analysis. Seventy percent of patients experienced at least one relapse and/or progression during their disease course. At the last follow-up, 18 of 20 (82%) patients achieved complete ($n = 8$) or partial ($n = 10$) remission while one patient experienced progressive disease (Table 4). In patients with partial remission, residual symptoms were peripheral neuropathy, gait disorders, and cognitive impairment. One patient died during follow-up with a cause of death not related to neurosarcoidosis.

Literature review

Our literature search identified 776 articles of which 741 were excluded after abstracts and full-text records screening (Figure 1). Thirty-five articles met our inclusion criteria, including 1,793 patients diagnosed with neurosarcoidosis from 1995 to 2021 (7, 16, 19–53). Only three (9%) studies were prospective (33, 36, 48) and 14 (40%) were multicentric (16, 19, 22, 24–26, 29, 30, 32, 35, 39, 40, 43, 45) (Supplementary Table 2). As illustrated in Table 2, patient characteristics are consistent with those in our cohort, except for male predominance. The Neurosarcoidosis Consortium Consensus Group's 2018 Diagnostic Criteria (13) were applied to all studies to consistently identify possible (13%), probable (66%), and definite (23%) neurosarcoidosis. Eighty-three percent of patients were diagnosed with systemic sarcoidosis, mainly consisting of lungs (62%) and lymph nodes (53%) involvement. In contrast to our patient cohort, cardiac, ear-nose-throat, kidney, digestive tract, and scrotal localizations were also described. Similar proportions in systemic and neurological symptoms as well as site of neurological involvement were observed in our cohort and in the literature review except for hydrocephalus described in 3% of patients in the literature (Figure 3). Results of ancillary investigations at diagnosis are summarized in Table 3. Serum lysozyme and ACE were elevated in 46 and 37% of patients, respectively. Pleocytosis was found in 63%, increased proteinorrhachia in 70%, low glucose levels in 33%, increased IgG index in 37%, and CSF-specific IgG oligoclonal bands in 23% of patients. Brain MRI showed abnormalities in 76% of patients mainly consisting of parenchymal lesions (46%), meningeal enhancement (39%), and hypothalamic-pituitary axis lesions (23%), similar to our cohort. Spinal cord MRI showed abnormalities in 61% of evaluated patients, while [18F]FDG-PET/CT revealed systemic or neuro-sarcoidosis in 43% of patients. Second-line therapy mainly consisted of methotrexate (45%), followed by azathioprine (14%) and mycophenolate mofetil (12%) (Figure 4). In most cases, these therapies were initiated for treating a progressive or relapsing disease as well as in association with a TNF-α antagonist (Table 6). A favorable outcome was reported in 40% of patients treated with methotrexate, in 56% with azathioprine, and in 45% with mycophenolate mofetil, as illustrated in Figure 5, Tables 6, 7. Relapse or progressive disease occurred most frequently with mycophenolate mofetil (58%) and azathioprine (46%) compared to methotrexate (39%). Third-line therapy consisted of TNF-α antagonists in 27% of patients with a high rate of favorable outcome (80%), similarly to our cohort. The proportion of favorable outcomes was significantly higher in patients treated by the TNF-α antagonist compared to those treated by methotrexate ($p < 0.0001$), mycophenolate mofetil ($p < 0.0001$), or azathioprine ($p < 0.0001$) (Table 8). The final outcome in the literature was reported in 1,446/1,793 patients, with favorable outcomes in 65% of cases.

Focus on neurosarcoidosis treatment with TNF-α antagonists

Including our study, we identified 25 studies reporting 406 patients diagnosed with neurosarcoidosis and treated with

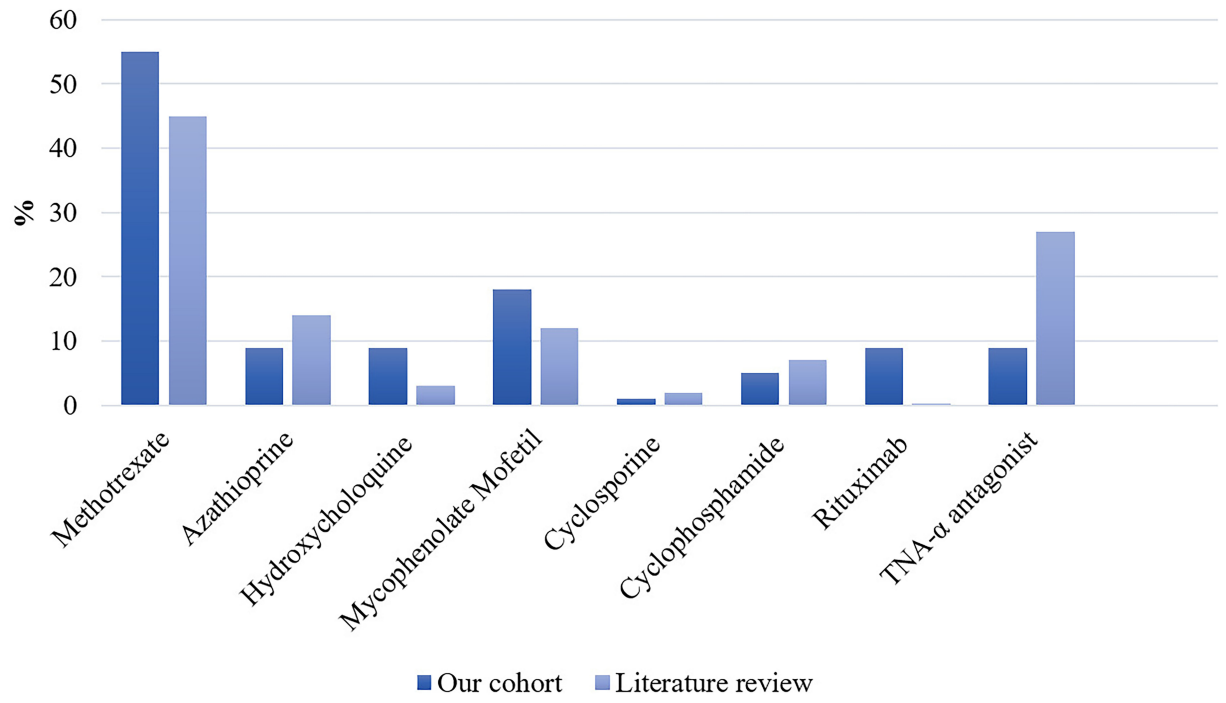


FIGURE 4
Proportion of second- and third-line therapies in our patient cohort ($n = 22$) and the literature expressed as percentages. In the literature: n/N = 557/1,248 for methotrexate; n/N = 185/1,363 for azathioprine; n/N = 44/1,431 for hydroxychloroquine; n/N = 178/1,431 for mycophenolate mofetil; n/N = 20/1,425 for cyclosporine; n/N = 99/1,431 for cyclophosphamide; n/N = 5/1,431 for Rituximab; and n/N = 405/1,494 for TNF-α antagonist.

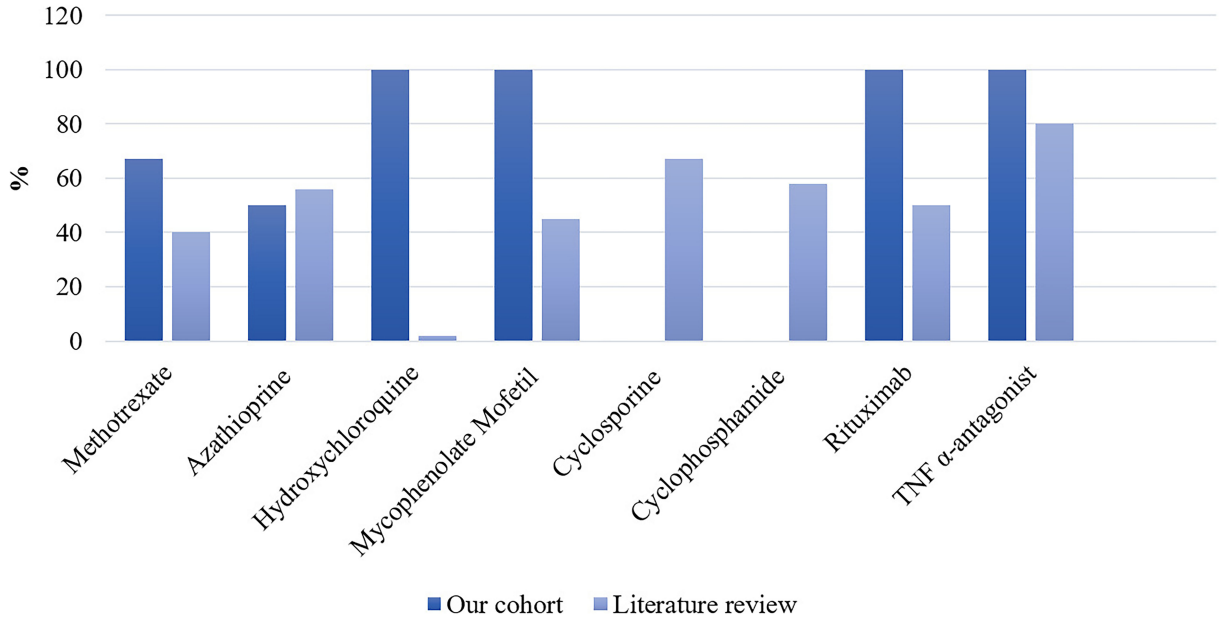


FIGURE 5
Proportion of favorable outcomes in patients from our cohort ($n = 22$) and the literature expressed as percentages. In the literature: n/N = 56/140 for methotrexate; n/N = 31/56 for azathioprine; n/N = 1/60 for hydroxychloroquine; n/N = 27/60 for mycophenolate mofetil; n/N = 10/15 for cyclosporine; n/N = 19/33 for cyclophosphamide; n/N = 2/4 for rituximab; and n/N = 280/352 for TNF-α antagonist.

TABLE 6 Indication, outcome, reasons for discontinuation, and adverse events according to the second-line therapy at baseline and during follow-up in patients from our cohort and the literature.

		Second line therapy							
		Methotrexate		Azathioprine		Mycophenolate mofetil		Cyclosporine	
		Our cohort ($n = 12$) n (%)	Literature review ($n = 557$) n/N (%)	Our cohort ($n = 2$) n (%)	Literature review ($n = 185$) n/N (%)	Our cohort ($n = 5$)* n (%)	Literature review ($n = 178$) n/N (%)	Our cohort ($n = 0$) n (%)	Literature review ($n = 20$) n/N (%)
Indication									
	Since baseline	2 (17)	16/356 (4)	0 (0)	8/143 (6)	1 (20)	6/90 (7)	0 (0)	0/16 (0)
	Active, progressive or relapsing disease	5 (42)	35/356 (10)	1 (50)	36/143 (25)	3 (60)	18/90 (20)	0 (0)	16/16 (100)
	Corticosteroids sparing	3 (25)	20/356 (7)	1 (50)	8/143 (6)	0 (0)	13/90 (14)	0 (0)	0/16 (0)
	Intolerance to other treatment	1 (8)	12/356 (3)	0 (0)	6/143 (4)	1 (20)	6/90 (7)	0 (0)	0/16 (0)
	Associated with TNF-alpha antagonists	1 (8)	83/356 (23)	0 (0)	32/143 (22)	0 (0)	28/90 (31)	0 (0)	0/16 (0)
	Unknown	0 (0)	190/356 (53)	0 (0)	53/143 (37)	0 (0)	19/90 (21)	0 (0)	0/16 (0)
Outcomes									
	Favorable outcome	8 (67)	56/140 (40)	1 (50)	31/56 (56)	5 (100)	27/60 (45)	0 (0)	10/15 (67)
	Active, progressive or relapsing disease	4 (33)	45/140 (32)	1 (50)	13/56 (23)	0 (100)	25/60 (42)	0 (0)	3/15 (20)
	Mortality	0 (0)	0/140 (0)	0 (0)	3/56 (5)	0 (100)	0/60 (0)	0 (0)	1/15 (7)
	Unknown	0 (0)	39/140 (28)	0 (0)	9/56 (16)	0 (100)	8/60 (13)	0 (0)	0/15 (0)
Discontinuation		5/12 (42)	57/127 (45)	2 (100)	24/87 (26)	4 (80)	38/64 (59)	0 (0)	11/15 (73)
	Intolerance or adverse events	0/5 (0)	14/57 (25)	1 (50)	4/24 (17)	2 (50)	3/38 (8)	0 (0)	2/11 (18)
	Relapse or progressive disease	2/5 (40)	22/57 (39)	1 (50)	11/24 (46)	0 (0)	22/38 (58)	0 (0)	0/11 (0)
	Complete or partial remission	3/5 (60)	15/57 (26)	0 (0)	5/24 (21)	2 (0)	9/38 (24)	0 (0)	0/11 (0)
	Study protocol	0 (0)	0 (0)	0 (0)	0/24 (0)	0 (0)	0/38 (0)	0 (0)	6/11 (55)
	Unknown	0/5 (0)	6/57 (10)	0 (0)	4/24 (17)	0 (0)	4/38 (10)	0 (0)	3/11 (27)
Adverse events		0 (0)	21/246 (9)	1 (50)	5/62 (8)	1 (25)	1/18 (6)	0 (0)	5/15 (33)

n/N represents the number for which the data are present out of the total number of patients for which the data are described.
NA, Not available.
*One patient was treated two times with mycophenolate mofetil.
Table adapted from Gavoille et al. (25).

TNF-α antagonists (7, 16, 19–28, 36, 39, 41, 42, 44–53) (Supplementary Table 3). Detailed patient characteristics and treatment modalities are reported in Table 9. Ninety-seven percent of patients received intravenous infliximab at a dose of 5 mg/kg (ranging from 3.5 to 7 mg/kg) initially given at 2- or 4-week intervals followed by every 6 or 8 weeks. Three percent of patients received 40 mg of subcutaneous adalimumab every 1 or 2 weeks. Anti-TNF-α treatment indication mainly consisted of relapse or progression under other therapy (66%). Eighty-eight percent of patients were concomitantly treated with corticosteroids. Other accompanying treatments included methotrexate (28%), azathioprine (11%), and mycophenolate mofetil (10%) in most cases. Seven percent of patients had no concomitant treatment. The median treatment duration was 23

TABLE 7 Indication, outcome, reasons for discontinuation, and adverse events according to the third-line therapy at baseline and during follow-up in patients from our cohort and the literature.

	Cyclophosphamide		Rituximab		TNF-alpha antagonist	
	Our cohort ($n = 1$) n (%)	Literature review ($n = 99$) n/N (%)	Our cohort ($n = 2$) n (%)	Literature review ($n = 5$) n/N (%)	Our cohort ($n = 2$) n (%)	Literature review ($n = 404$) n/N (%)
Indication						
Since baseline	0 (0)	22/70 (31)	0 (0)	0/1 (0)	0 (0)	8/260 (3)
Active, progressive or relapsing disease	1 (100)	41/70 (59)	2 (100)	0/1 (0)	1 (50)	173/260 (66)
Corticosteroids sparing	0 (0)	0/70 (0)	0 (0)	1/1 (100)	0 (0)	15/260 (6)
Intolerance to other treatment	0 (0)	1/70 (1)	0 (0)	0/1 (0)	0 (0)	12/260 (5)
Associated with TNF-alpha antagonists	0 (0)	1/70 (1)	0 (0)	0/1 (0)	–	–
Unknown	0 (0)	5/70 (7)	0 (0)	0 (0)	1 (50)	52/260 (20)
Outcomes						
Favorable outcome	0 (0)	19/33 (58)	2 (100)	2/4 (50)	2 (100)	280/352 (80)
Active, progressive or relapsing disease	1 (100)	9/33 (27)	0 (0)	2/4 (50)	0 (0)	39/352 (11)
Mortality	0 (0)	0/33 (0)	0 (0)	0/4 (0)	0 (0)	1/352 (0,3)
Unknown	0 (0)	5/33 (15)	0 (0)	0/4 (0)	0 (0)	32/352 (9)
Discontinuation	1 (100)	12/58 (21)	1 (50)	NA	1 (50)	69/221 (31)
Intolerance or adverse events	0 (0)	3/12 (25)	0 (0)	–	0 (0)	11/69 (16)
Relapse or progressive disease	1 (100)	2/12 (17)	0 (0)	–	0 (0)	4/69 (6)
Complete or partial remission	0 (0)	0/12 (0)	1 (100)	–	1 (100)	48/69 (69)
Study protocol	0 (0)	0/12 (0)	0 (0)	–	0 (0)	0/69 (0)
Unknown	0 (0)	7/12 (58)	0 (0)	–	0 (0)	6/69 (9)
Adverse events	0 (0)	5/11 (45)	0 (0)	NA	1 (50)	59/204 (29)

n/N represents the number for which the data are present out of the total number of patients for which the data are described.
NA, Not available.
Table adapted from Gavoille et al. (25).

TABLE 8 Comparison of treatment outcomes of neurosarcoidosis according to treatment between azathioprine, methotrexate, and mycofenolate mofetil, respectively, and TNF-alpha antagonist.

	Methotrexate	Anti-TNF alpha	Odds ratio	95% CI	p-value
Favorable outcome	56/140	280/352	4.68	3.10–7.03	<0.0001
	Mycophenolate mofetil	**Anti-TNF alpha**	**Odds ratio**	**95% CI**	**p-value**
Favorable outcome	27/60	280/352	5.89	3.58–9.86	<0.0001
	Azathioprine	**Anti-TNF alpha**	**Odds ratio**	**95% CI**	**p-value**
Favorable outcome	31/56	280/352	5.26	2.84–9.36	<0.0001

Proportions are indicated and were compared using Fisher's exact test. Reciprocal odds ratios for a favorable outcome on anti-TNF-a therapy are indicated, together with a 95% confidence interval and p-value.
Patients from the published cohort and ours are combined.

months (1–93) with a median follow-up of 29 months (1–123 months). Eighty percent presented favorable outcomes following anti-TNF-α therapy, while mortality related to neurosarcoidosis was reported in only one patient (24). Corticosteroids could be, respectively, tapered or stopped in 36% and 34% of patients. Two studies, including a total of 38 patients, reported a significant decrease in the daily dose of corticosteroids ($p < 0.0001$) (45, 46). Anti-TNF-α treatment was discontinued in 31% of patients because of stable disease (70%), intolerance or adverse events (16%), and relapse or progression (6%). Data post-treatment discontinuation was available in 28 patients. Among these, 50% of patients presented a relapse after stopping anti-TNF-α therapy. Twelve patients were rechallenged with either infliximab ($n = 11$) or adalimumab ($n = 1$) and all showed favorable outcomes. Seven studies reported switches from Infliximab to Adalimumab ($n = 8$), Adalimumab to Infliximab ($n = 1$), and Etanercept to Infliximab ($n = 1$). Indications were mainly adverse events (28%) and relapse or progression (33%). A favorable outcome was noted in 94% of these patients. Overall, adverse events were reported in 29% of patients, including infection, infusion reaction, and headache in most cases.

TABLE 9 Combined disease characteristics, management, and outcomes of neurosarcoidosis cases treated with the anti-TNF-α antagonist from our cohort ($n = 2$) and the literature ($n = 404$).

	406
Cases, n	
Sex	
Male, n/N (%)	90/183 (49%)
Female, n/N (%)	93/183 (51%)
Systemic sarcoidosis, n/N (%)	121/132 (92%)
Site of neurological involvement	
Meningeal involvement, n/N (%)	87/160 (54%)
Parenchymal disease, n/N (%)	45/160 (28%)
Cranial neuropathy, n/N (%)	38/160 (24%)
Spinal cord involvement, n/N (%)	69/160 (43%)
TNF-α antagonist treatment	
Infliximab, n/N (%)	368/379 (97%)
Adalimimumab, n/N (%)	10/379 (3%)
Both infliximab and adalimumab, n/N (%)	1/379 (0,3%)
TNF-α antagonist indication	
Relapse/progression under other therapy, n/N (%)	173/262 (66%)
Maintenance or corticosteroid-sparing therapy, n/N (%)	16/262 (6%)
Baseline therapy due to severe disease phenotype, n/N (%)	8/262 (3%)
Intolerance to other treatment, n/N (%)	13/262 (5%)
Unknown, n/N (%)	52/262 (20%)
Prior treatment	
Corticosteroids, n/N (%)	185/289 (64%)
Methotrexate, n/N (%)	78/289 (27%)
Azathioprine, n/N (%)	42/289 (15%)
Mycophenolate mofetil, n/N (%)	50/289 (17%)
Cyclophosphamide, n/N (%)	32/289 (11%)
Hydroxychloroquine, n/N (%)	6/289 (2%)
Cyclosporine, n/N (%)	2/289 (0.7%)
Rituximab, n/N (%)	1/289 (0.3%)
Etanercept, n/N (%)	1/289 (0.3%)
None, n/N (%)	8/289 (3%)
Concomitant treatment	
Corticosteroids, n/N (%)	242/275 (88%)
Methotrexate, n/N (%)	78/275 (28%)
Azathioprine, n/N (%)	30/275 (11%)
Mycophenolate mofetil, n/N (%)	28/275 (10%)
Hydroxychloroquine, n/N (%)	1/275 (0.3%)
Cyclophosphamide, n/N (%)	1/275 (0.3%)
None, n/N (%)	19/275 (7%)
Outcome	
Favorable outcome n/N (%)	282/354 (80)
Relapse or progression, n/N (%)	39/354 (11)
Mortality, n/N (%)	1/354 (0,3%)
Unknown, n/N (%)	32/354 (9%)
Corticosteroids tapering or stopping, n/N (%)	77/110 (70%)
Discontinuation of TNF-α antagonist, n/N (%)	70/223 (31%)

(Continued)

Discussion

We retrospectively described clinical features, ancillary investigations, and treatment in a cohort of patients with neurosarcoidosis treated in a tertiary academic hospital in Belgium and compared our results with the existing evidence published so far in the literature, with a focus on treatment outcomes with TNF-α antagonists.

Clinical characteristics

More than 80% of patients with neurosarcoidosis have associated systemic sarcoidosis, mainly consisting of lungs and lymph nodes involvement. Neurologic manifestations are the initial clinical symptoms in 50–70% of patients and systemic sarcoidosis is subsequently detected during the diagnostic workup. Our data confirm the large diversity and heterogeneity in the clinical presentation of neurosarcoidosis. Meninges are the most frequently affected neurological site and may be complicated by cranial nerve dysfunction and seizures as well as hydrocephalus in case of chronic meningitis (9). The involvement of brain parenchyma can explain acute or chronic cognitive dysfunction, headache, seizures, gait disturbances, stroke, and hydrocephalus (54). Cranial nerve neuropathy is also part of the commonly reported manifestation of neurosarcoidosis, either affecting the optic, the facial, or the vestibulocochlear nerves (9, 54). Spinal cord is involved in one-third of patients and may lead to motor or sensory deficits, bowel and bladder dysfunction, as well as sexual dysfunction (9, 54). Other

TABLE 9 (Continued)

Intolerance or adverse events, n/N (%)	11/70 (16%)
Relapse or progression, n/N (%)	4/70 (6%)
Stable disease, n/N (%)	49/70 (70%)
Unknown, n/N (%)	6/70 (8%)
Relapse post-TNF-α antagonist discontinuation, n/N (%)	14/28 (50%)
Duration of treatment (months), median (range)	23 (1–93)
Adverse events, n/N (%)	60/206 (29%)
Follow-up (months), median (range)	29 (1–123)

n/N represents the number for which the data are present out of the total number of patients for which the data are described.

neurosarcoidosis sites reported to be involved included the hypothalamic-pituitary axis resulting in hormonal deficiencies (e.g., syndrome of inappropriate antidiuretic hormone, hypothyroidism, hyperprolactinemia, hypoadrenalism, and diabetes insipidus) (9, 54), the peripheral nervous system, and muscles (9).

Ancillary investigations

The diagnosis of neurosarcoidosis is challenging, largely due to heterogeneous clinical presentations and low sensitivity of ancillary investigations (10). The diagnostic criteria of neurosarcoidosis have been updated in 2018 and categorized patients into definite, probable, and possible neurosarcoidosis based on suggestive clinical presentation, results of ancillary investigations, histopathological confirmation of non-caseating granulomas, and rigorous exclusion of other causes (9, 10, 13).

In accordance with previous data (9, 10, 42, 53, 54), we confirm the low sensitivity of serum analysis except for lysozyme levels which were increased in almost 80% of our cohort, while it was reported abnormal in only half of patients with neurosarcoidosis in the literature (42). Serum testing is therefore mainly useful to exclude alternative diagnoses such as autoimmune and infectious diseases (tuberculosis, syphilis) and systemic complications in the context of sarcoidosis (liver and kidney impairment as well as hypercalcemia and hematological abnormalities). Therefore, the initial biological workup is classically characterized by: CRP, calcium, antinuclear antibody, antineutrophil cytoplasmic antibody, HIV, and syphilis serologies, as well as screening for tuberculosis. In some cases (history of traveling, immunosuppression, and contact with animals such as cat and sheep), fungal and bacterial serologies (bartonella and brucella) may be indicated according to clinical suspicion (9). Anti-aquaporine-4 and anti-myelin oligodendrocyte glycoprotein IgG antibodies should be measured especially in the context of myelitis. There remains an unmet need to define novel biomarkers to help in establishing the diagnosis of neurosarcoidosis. In 2019, serum soluble IL-2 receptor was proposed to be a sensitive diagnostic biomarker (55) but its cut-off levels have not been precisely defined yet (56).

Although unspecific in neurosarcoidosis (9, 10, 41, 53), lumbar puncture should be considered to evaluate intrathecal inflammation and to exclude alternative diagnoses (9). Many patients with neurosarcoidosis have an abnormal cerebrospinal fluid (CSF) analysis, including pleocytosis (mostly mild to moderate, with lymphocytic predominance), increased protein, and rarely low glucose levels (10, 57). As neurosarcoidosis is a rare non-infectious disease that can cause hypoglycorrhachia (9), it may have a diagnostic value after the exclusion of lymphoma mycobacterial and fungal infection (41, 57). It could be particularly relevant in sarcoidosis with spinal cord involvement as hypoglycorrhachia is not observed in other cases of inflammatory myelopathies (57). An elevated immunoglobulin G index and IgG oligoclonal bands are described in about one-third of patients with neurosarcoidosis but should be interpreted with caution as it occurs in 95% to 98% of patients with multiple sclerosis (50).

In the evaluation of neurosarcoidosis, brain and spinal MRI with gadolinium injection is the gold-standard imaging modality (9, 54, 57) due to its high sensitivity (82–97%) for active inflammation (54). The value of the [18F]FDG-PET/CT is particularly well-illustrated in our cohort as 85% of patients had abnormalities, although a significant proportion of these had a normal chest X-Ray or CT scan. Its usefulness is based on the detection of extra-neurologic localizations and the identification of hypermetabolic target lesions easily accessible for biopsy (9, 25, 58).

Neurosarcoidosis is fundamentally a diagnosis made by histopathology, although there is also a histological differential diagnosis to make (9). The definite diagnostic criteria of neurosarcoidosis are met in a minority of patients as it requires relatively high-risk invasive procedures such as brain or leptomeningeal biopsy (13). Diagnosis of probable neurosarcoidosis is therefore preferentially obtained by less invasive extraneural biopsy, such as pulmonary, lymph node, salivary gland, or skin biopsy (9, 54). Actually, there are many alternative causes of granulomatosis such as infection (e.g., mycobacterium tuberculosis), inflammatory diseases (e.g., inflammatory bowel diseases and granulomatosis with polyangiitis), and lymphoma (e.g., Hodgkin's lymphoma), which must be ruled out (9).

Treatment

Treatment guidelines for neurosarcoidosis are principally based on expert opinion and observations from small cohort studies and non-randomized clinical trials (9, 10, 54). Treatment of neurosarcoidosis should therefore be patient-tailored and take into consideration other concomitant systemic involvement (9). Early and aggressive treatment is required in the majority

of neurosarcoidosis cases to prevent morbidity and mortality (9, 38, 54), except in cases of isolated facial nerve palsy or aseptic meningitis, in which moderate and shorter treatment courses may be sufficient (27, 28, 38, 48).

Corticosteroids remain the cornerstone and first-line treatment in neurosarcoidosis (9, 28, 54, 59). However, due to incomplete response, disease progression, recurrence, or corticosteroid-induced toxicity, second- and/or third-line therapies are required in a majority of patients as was the case in our patient cohort and the literature review. Methotrexate is the most frequently used second-line treatment. Azathioprine, mycophenolate mofetil, and hydroxychloroquine are usual alternatives to methotrexate but are associated with lesser efficacy in relapse prevention (7, 25, 28, 43, 47). Cyclosporine and cyclophosphamide are less considered due to their significant side effects and should therefore be used as a last resort (16, 17, 28).

Third-line treatments such as TNF-α antagonists are increasingly used in the management of neurosarcoidosis. Infliximab and adalimumab are monoclonal antibodies that inactivate TNF-α, a pro-inflammatory cytokine critical for the formation and maintenance of sarcoid granulomas (9, 16, 27, 52). They are commonly used in combination with corticosteroids and other immunosuppressive therapy such as methotrexate and azathioprine, although their effectiveness as monotherapy in neurosarcoidosis is also reported (26). In addition to potential synergistic immunosuppressive benefits, the combination of a TNF-α antagonist with low-dose second-line therapy may be useful to attenuate the risk of anti-drug antibody formation (16, 26, 27, 36, 37). Most patients (80%) achieve a favorable outcome with anti-TNF-α therapy. The proportion of favorable outcomes was significantly higher in patients treated with TNF-α antagonist compared to those treated with methotrexate, mycophenolate mofetil, or azathioprine. In up to 70% of patients, corticosteroids could be tapered or even stopped, confirming the role of anti-TNF-α as efficient corticosteroid-sparing agents even in cases of refractory or aggressive neurosarcoidosis (9, 27). Although rare, relapses can occur during anti-TNF-α therapy. In this circumstance, it is important to verify the presence of anti-drug-neutralizing antibodies (24). When anti-TNF-α therapy is discontinued, patients should be monitored clinically and by MRI since relapse occurs in 50% of cases, particularly during the first year following therapy withdrawal (9, 48) and typically within the same neurological localization (9, 27). The reintroduction of anti-TNF-α therapy resulted in a favorable outcome in 100% of patients. Adverse events are common but rarely require permanent discontinuation. Infections are the most important adverse effects, accounting for approximately one-third of cases, but only one death related to unspecified infectious disease was reported in the literature (24). The risk of infectious complications is higher in patients already treated for a longer duration with corticosteroids and immunosuppressive therapy before the introduction of TNF-α antagonist therapy (21). These results highlight the benefits of TNF-α inhibitors in neurosarcoidosis and suggest that they should be prescribed earlier in the disease course. However, these drugs are currently not licensed nor reimbursed by the Belgian healthcare system for treating neurosarcoidosis.

B cell-Targeted therapy (rituximab) seems to have some efficacy in sarcoidosis especially in systemic sarcoidosis and even in neurosarcoidosis. However, this is based on small cohort studies, and there is insufficient data to support the use of rituximab over TNF inhibitors (47, 60).

Janus Kinase inhibitors (Jak inhibitors) are new drugs targeting the JAK/STAT pathways and are used in several diseases such as rheumatoid arthritis, inflammatory bowel disease, graft vs. host disease, and hemophagocytic lymphohistiocytosis (61). JAK/STAT plays a key role in the signaling pathways of several pro-inflammatory cytokines and thus may be a good therapeutic option. Tofacitinib and baricitinib have been used in refractory cutaneous and systemic sarcoidosis but data on neurosarcoidosis are lacking (62, 63).

Outcome

In our cohort, favorable outcome was reported in up to 81% of patients, compared to 65% of cases in the literature. The higher favorable outcome in our cohort could be attributed to several factors. First, the majority of our patients were diagnosed between 2015 and 2021 and have therefore benefited from the most recent treatment strategies. Second, our cohort did not include patients with hydrocephalus which is known to have a worse outcome (7, 28). Despite the large proportion of favorable outcomes at the last follow-up, ∼70% of patients experienced relapse and/or progression during their disease course. Moreover, some patients, even in case of stable inactive disease or remission, will experience a significant loss of autonomy due to neurological sequelae, especially in the case of spinal cord involvement (19, 53).

Limitations

Our study has several limitations. First, our study and most studies included in the literature search were retrospective in nature with inherent limitations and were performed in tertiary centers leading possibly to selection bias. To maximize case ascertainment, we carefully and systematically reviewed all patients' medical records of our cohort and all available patient data from studies included in the literature review. Although possible neurosarcoidosis cases were not excluded in our study, as recommended since 2018 (13), it allowed us to include a larger number of patients and better reflect daily clinical practice. Second, we did not perform a systematic review. However, the scope and depth of our manuscript are extensive enough to render a review piece. Third, pooled analysis of literature

data must be interpreted with caution due to the heterogeneity of inclusion criteria, neurological manifestations, treatment outcome definition, immunosuppressive therapy strategies, and their evolution over time, as well as the possible inclusion of some patients two times despite rigorous review of each study by the authors, inclusion date and centers, and contact of several corresponding authors. Moreover, all items were not reported for every patient. To compensate for this bias, results were presented as the percentage of patients for which the data were available [n/N (%)].

Conclusion

Sarcoidosis is the most common non-infectious granulomatous disease affecting the nervous system. Its diagnosis remains challenging due to heterogeneity in clinical presentation and results of ancillary investigations. The results of our cohort and literature review provide relevant results regarding treatment with TNF-α antagonists and confirm their effectiveness in neurosarcoidosis. Additional studies, in particular multicenter clinical trials designed for rare diseases (64), are needed to confirm their safety, efficacy, and potential earlier place in the therapeutic armamentarium of neurosarcoidosis, as well as to determine the duration, tapering, and timing for the eventual interruption.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary material, further inquiries can be directed to the corresponding author/s.

Ethics statement

The studies involving human participants were reviewed and approved by Comité d'Éthique Hospitalo-Facultaire (CEHF). Written informed consent for participation was not required for this study in accordance with the national legislation and the institutional requirements.

Author contributions

PS, HY, and VP designed the study, take care of the patients, and wrote the manuscript. AK, LP, and AS corrected the manuscript and helped in the management of the patients. OG corrected the manuscript and helped with the interpretation of the radiological exam. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.970168/full#supplementary-material

References

1. Drent M, Crouser ED, Grunewald J. Challenges of sarcoidosis and its management. *N Engl J Med*. (2021) 385:1018–32. doi: 10.1056/NEJMra2101555

2. Iannuzzi MC, Rybicki BA, Teirstein AS. Sarcoidosis. *N Engl J Med*. (2007) 357:2153–65. doi: 10.1056/NEJMra071714

3. Yanardag H, Pamuk ON, Pamuk GE. Lupus pernio in sarcoidosis: clinical features and treatment outcomes of 14 patients. *J Clin Rheumatol*. (2003) 9:72–6. doi: 10.1097/01.RHU.0000062509.01658.d1

4. Scott GC, Berman JM, Higgins JL Jr. CT patterns of nodular hepatic and splenic sarcoidosis: a review of the literature. *J Comput Assist Tomogr*. (1997) 21:369–72. doi: 10.1097/00004728-199705000-00006

5. Judson MA. Hepatic, splenic, and gastrointestinal involvement with sarcoidosis. *Semin Respir Crit Care Med*. (2002) 23:529–41. doi: 10.1055/s-2002-36517

6. Baughman RP, Winget DB, Bowen EH, Lower EE. Predicting respiratory failure in sarcoidosis patients. *Sarcoidosis Vasc Diffuse Lung Dis*. (1997) 14:154–8.

7. Joubert B, Chapelon-Abric C, Biard L, Saadoun D, Demeret S, Dormont D, et al. Association of prognostic factors and immunosuppressive treatment with long-term outcomes in neurosarcoidosis. *JAMA Neurol*. (2017) 74:1336–44. doi: 10.1001/jamaneurol.2017.2492

8. Affan M, Mahajan A, Rehman T, Kananeh M, Schultz L, Cerghet M. The effect of race on clinical presentation and outcomes in neurosarcoidosis. *J Neurol Sci*. (2020) 417:117073. doi: 10.1016/j.jns.2020.117073

9. Bradshaw MJ, Pawate S, Koth LL, Cho TA, Gelfand JM. Neurosarcoidosis: pathophysiology, diagnosis, and treatment. *Neurol Neuroimmunol Neuroinflamm*. (2021) 8:e1084. doi: 10.1212/NXI.0000000000001084

10. Fritz D, van de Beek D, Brouwer MC. Clinical features, treatment and outcome in neurosarcoidosis: systematic review and meta-analysis. *BMC Neurol*. (2016) 16:220. doi: 10.1186/s12883-016-0741-x

11. Judson MA, Boan AD, Lackland DT. The clinical course of sarcoidosis: presentation, diagnosis, and treatment in a large white and black cohort in the United States. *Sarcoidosis Vasc Diffuse Lung Dis*. (2012) 29:119–27.

12. Caruana LB, Redwine GD, Rohde RE, Russian CJ. A prospective study of patients diagnosed with sarcoidosis: factors - environmental exposure, health assessment, and genetic outlooks. *Sarcoidosis Vasc Diffuse Lung Dis*. (2019) 36:228–42. doi: 10.36141/svdld.v36i3.7112

13. Stern BJ, Royal W III, Gelfand JM, Clifford DB, Tavee J, Pawate S, et al. Definition and consensus diagnostic criteria for neurosarcoidosis: from the Neurosarcoidosis Consortium Consensus group. *JAMA Neurol*. (2018) 75:1546–53. doi: 10.1001/jamaneurol.2018.2295

14. Crouser ED, Maier LA, Wilson KC, Bonham CA, Morgenthau AS, Patterson KC, et al. Diagnosis and detection of sarcoidosis. An official American thoracic society clinical practice guideline. *Am J Respir Crit Care Med*. (2020) 201:e26–51. doi: 10.1164/rccm.202002-0251ST

15. Baughman RP, Valeyre D, Korsten P, Mathioudakis AG, Wuyts WA, Wells A, et al. ERS clinical practice guidelines on treatment of sarcoidosis. *Eur Respir J*. (2021) 58:2004079. doi: 10.1183/13993003.04079-2020

16. Gelfand JM, Bradshaw MJ, Stern BJ, Clifford DB, Wang Y, Cho TA, et al. Infliximab for the treatment of CNS sarcoidosis: a multi-institutional series. *Neurology*. (2017) 89:2092–100. doi: 10.1212/WNL.0000000000004644

17. Gangemi AJ, Myers CN, Zheng M, Brown J, Butler-LeBair M, Cordova F, et al. Mortality for sarcoidosis patients on the transplant wait list in the Lung Allocation Score era: experience from a high volume center. *Respir Med*. (2019) 157:69–76. doi: 10.1016/j.rmed.2019.09.001

18. Jang DI, Lee AH, Shin HY, Song HR, Park JH, Kang TB, et al. The role of tumor necrosis factor alpha (TNF-α) in autoimmune disease and current TNF-α inhibitors in therapeutics. *Int J Mol Sci*. (2021) 22:2719. doi: 10.3390/ijms22052719

19. Sakkat A, Cox G, Khalidi N, Larche M, Beattie K, Renzoni EA, et al. Infliximab therapy in refractory sarcoidosis: a multicenter real-world analysis. *Respir Res*. (2022) 23:54. doi: 10.1186/s12931-022-01971-5

20. Russell E, Luk F, Manocha S, Ho T, O'Connor C, Hussain H. Long term follow-up of infliximab efficacy in pulmonary and extra-pulmonary sarcoidosis refractory to conventional therapy. *Semin Arthritis Rheum*. (2013) 43:119–24. doi: 10.1016/j.semarthrit.2012.10.008

21. Chapelon-Abric C, Saadoun D, Biard L, Sene D, Resche-Rigon M, Hervier B, et al. Long-term outcome of infliximab in severe chronic and refractory systemic sarcoidosis: a report of 16 cases. *Clin Exp Rheumatol*. (2015) 33:509–15.

22. Jounieaux F, Chapelon C, Valeyre D, Israel Biet D, Cottin V, Tazi A, et al. Infliximab et sarcoïdose chronique. L'expérience française à propos de 31 cas [Infliximab treatment for chronic sarcoidosis–a case series]. *Rev Mal Respir*. (2010) 27:685–92. doi: 10.1016/j.rmr.2010.06.011

23. Hostettler KE, Studler U, Tamm M, Brutsche MH. Long-term treatment with infliximab in patients with sarcoidosis. *Respiration*. (2012) 83:218–24. doi: 10.1159/000328738

24. Fritz D, Timmermans WMC, van Laar JAM, van Hagen PM, Siepman TAM, van de Beek D, et al. Infliximab treatment in pathology-confirmed neurosarcoidosis. *Neurol Neuroimmunol Neuroinflamm*. (2020) 7:e847. doi: 10.1212/NXI.0000000000000847

25. Gavoille A, Desbois AC, Joubert B, Durel CA, Auvens C, Berthoux E, et al. Prognostic factors and treatment efficacy in spinal cord sarcoidosis: an observational cohort with long-term follow-up. *Neurology*. (2022) 98:e1479–88. doi: 10.1212/WNL.0000000000200020

26. Hilezian F, Maarouf A, Boutiere C, Rico A, Demortiere S, Kerschen P, et al. TNF-α inhibitors used as steroid-sparing maintenance monotherapy in parenchymal CNS sarcoidosis. *J Neurol Neurosurg Psychiatry*. (2021) 92:890–6. doi: 10.1136/jnnp-2020-325665

27. Riancho-Zarrabeitia L, Delgado-Alvarado M, Riancho J, Oterino A, Sedano MJ, Rueda-Gotor J, et al. Anti-TNF-α therapy in the management of severe neurosarcoidosis: a report of five cases from a single centre and literature review. *Clin Exp Rheumatol*. (2014) 32:275–84.

28. Arun T, Palace J. Effects of immunotherapies and clinical outcomes in neurosarcoidosis: a retrospective cohort study. *J Neurol*. (2021) 268:2466–72. doi: 10.1007/s00415-021-10421-z

29. Agbogu BN, Stern BJ, Sewell C, Yang G. Therapeutic considerations in patients with refractory neurosarcoidosis. *Arch Neurol*. (1995) 52:875–9. doi: 10.1001/archneur.1995.00540330053014

30. Sharma OP. Neurosarcoidosis: a personal perspective based on the study of 37 patients. *Chest*. (1997) 112:220–8. doi: 10.1378/chest.112.1.220

31. Lower EE, Broderick JP, Brott TG, Baughman RP. Diagnosis and management of neurological sarcoidosis. *Arch Intern Med*. (1997) 157:1864–8. doi: 10.1001/archinte.157.16.1864

32. Zajicek JP, Scolding NJ, Foster O, Rovaris M, Evanson J, Moseley IF, et al. Central nervous system sarcoidosis–diagnosis and management. *QJM*. (1999) 92:103–17. doi: 10.1093/qjmed/92.2.103

33. Doty JD, Mazur JE, Judson MA. Treatment of corticosteroid-resistant neurosarcoidosis with a short-course cyclophosphamide regimen. *Chest*. (2003) 124:2023–6. doi: 10.1378/chest.124.5.2023

34. Scott TF, Yandora K, Valeri A, Chieffe C, Schramke C. Aggressive therapy for neurosarcoidosis: long-term follow-up of 48 treated patients. *Arch Neurol*. (2007) 64:691–6. doi: 10.1001/archneur.64.5.691

35. Joseph FG, Scolding NJ. Neurosarcoidosis: a study of 30 new cases. *J Neurol Neurosurg Psychiatry*. (2009) 80:297–304. doi: 10.1136/jnnp.2008.151977

36. Moravan M, Segal BM. Treatment of CNS sarcoidosis with infliximab and mycophenolate mofetil. *Neurology*. (2009) 72:337–40. doi: 10.1212/01.wnl.0000341278.26993.22

37. Pawate S, Moses H, Sriram S. Presentations and outcomes of neurosarcoidosis: a study of 54 cases. *QJM*. (2009) 102:449–60. doi: 10.1093/qjmed/hcp042

38. Androdias G, Maillet D, Marignier R, Pinède L, Confavreux C, Broussolle C, et al. Mycophenolate mofetil may be effective in CNS sarcoidosis but not in sarcoid myopathy. *Neurology*. (2011) 76:1168–72. doi: 10.1212/WNL.0b013e318212aafb

39. Langrand C, Bihan H, Raverot G, Varron L, Androdias G, Borson-Chazot F, et al. Hypothalamo-pituitary sarcoidosis: a multicenter study of 24 patients. *QJM*. (2012) 105:981–95. doi: 10.1093/qjmed/hcs121

40. Carlson ML, White JR Jr, Espahbodi M, Haynes DS, Driscoll CL, Aksamit AJ, et al. Cranial base manifestations of neurosarcoidosis: a review of 305 patients. *Otol Neurotol*. (2015) 36:156–66. doi: 10.1097/MAO.0000000000000501

41. Wang L, Li Y. Longitudinal ultra-extensive transverse myelitis as a manifestation of neurosarcoidosis. *J Neurol Sci*. (2015) 355:64–7. doi: 10.1016/j.jns.2015.05.017

42. Leonhard SE, Fritz D, Eftimov F, van der Kooi AJ, van de Beek D, Brouwer MC. Neurosarcoidosis in a tertiary referral center: a cross-sectional cohort study. *Medicine*. (2016) 95:e3277. doi: 10.1097/MD.0000000000003277

43. Bitoun S, Bouvry D, Borie R, Mahevas M, Sacre K, Haroche J, et al. Treatment of neurosarcoidosis: a comparative study of methotrexate and mycophenolate mofetil. *Neurology*. (2016) 87:2517–21. doi: 10.1212/WNL.0000000000003431

44. Tavee JO, Karwa K, Ahmed Z, Thompson N, Parambil J, Culver DA. Sarcoidosis-associated small fiber neuropathy in a large cohort: clinical aspects and response to IVIG and anti-TNF alpha treatment. *Respir Med*. (2017) 126:135–8. doi: 10.1016/j.rmed.2017.03.011

45. Cohen Aubart F, Bouvry D, Galanaud D, Dehais C, Mathey G, Psimaras D, et al. Long-term outcomes of refractory neurosarcoidosis treated with infliximab. *J Neurol*. (2017) 264:891–7. doi: 10.1007/s00415-017-8444-9

46. Riller Q, Cotteret C, Junot H, Benameur N, Haroche J, Mathian A, et al. Infliximab biosimilar for treating neurosarcoidosis: tolerance and efficacy in a retrospective study including switch from the originator and initiation of treatment. *J Neurol*. (2019) 266:1073–8. doi: 10.1007/s00415-019-09234-y

47. Lord J, Paz Soldan MM, Galli J, Salzman KL, Kresser J, Bacharach R, et al. Neurosarcoidosis: longitudinal experience in a single-center, academic healthcare system. *Neurol Neuroimmunol Neuroinflamm*. (2020) 7:e743. doi: 10.1212/NXI.0000000000000743

48. Kidd DP. Sarcoidosis of the central nervous system: safety and efficacy of treatment, and experience of biological therapies. *Clin Neurol Neurosurg*. (2020) 194:105811. doi: 10.1016/j.clineuro.2020.105811

49. Kidd DP. Sarcoidosis of the central nervous system: clinical features, imaging, and CSF results. *J Neurol*. (2018) 265:1906–15. doi: 10.1007/s00415-018-8928-2

50. Arun T, Pattison L, Palace J. Distinguishing neurosarcoidosis from multiple sclerosis based on CSF analysis: a retrospective study. *Neurology*. (2020) 94:e2545–54. doi: 10.1212/WNL.0000000000009491

51. Ten Dam L, van de Beek D, Brouwer MC. Clinical characteristics and outcome of hydrocephalus in neurosarcoidosis: a retrospective cohort study and review of the literature. *J Neurol*. (2022) 269:2727–33. doi: 10.1007/s00415-021-10882-2

52. Hutto SK, Kyle K, Cavanagh JJ, Reda H, Venna N. Adalimumab for CNS sarcoidosis: single-center experience and literature review. *J Neurol*. (2022) 269:2064–72. doi: 10.1007/s00415-021-10793-2

53. Nolte JYC, Ten Dam L, van de Beek D, Brouwer MC. Clinical characteristics and outcome of neurosarcoidosis-associated myelitis: a retrospective cohort study and review of the literature. *Eur J Neurol*. (2022) 29:1763–70. doi: 10.1111/ene.15295

54. Belperio JA, Shaikh F, Abtin F, Fishbein MC, Saggar R, Tsui E, et al. Extrapulmonary sarcoidosis with a focus on cardiac, nervous system, and ocular involvement. *EClinicalMedicine*. (2021) 37:100966. doi: 10.1016/j.eclinm.2021.100966

55. Eurelings LEM, Miedema JR, Dalm VASH, van Daele PLA, van Hagen PM, van Laar JAM, et al. Sensitivity and specificity of serum soluble interleukin-2 receptor for diagnosing sarcoidosis in a population of patients suspected of sarcoidosis. *PLoS ONE*. (2019) 14:e0223897. doi: 10.1371/journal.pone.0223897

56. Ramos-Casals M, Retamozo S, Sisó-Almirall A, Pérez-Alvarez R, Pallarés L, Brito-Zerón P. Clinically-useful serum biomarkers for diagnosis and prognosis of sarcoidosis. *Expert Rev Clin Immunol*. (2019) 15:391–405. doi: 10.1080/1744666X.2019.1568240

57. Durel CA, Marignier R, Maucort-Boulch D, Iwaz J, Berthoux E, Ruivard M, et al. Clinical features and prognostic factors of spinal cord sarcoidosis: a multicenter observational study of 20 BIOPSY-PROVEN patients. *J Neurol*. (2016) 263:981–90. doi: 10.1007/s00415-016-8092-5

58. Murphy OC, Salazar-Camelo A, Jimenez JA, Barreras P, Reyes MI, Garcia MA, et al. Clinical and MRI phenotypes of sarcoidosis-associated myelopathy. *Neurol Neuroimmunol Neuroinflamm*. (2020) 7:e722. doi: 10.1212/NXI.0000000000000722

59. Fritz D, van de Beek D, Brouwer MC, Booij J. Whole-body 18F-FDG PET-CT in the diagnosis of neurosarcoidosis. *Mayo Clin Proc*. (2020) 95:1082–4. doi: 10.1016/j.mayocp.2020.01.032

60. Zella S, Kneiphof J, Haghikia A, Gold R, Woitalla D, Thöne J. Successful therapy with rituximab in three patients with probable neurosarcoidosis. *Ther Adv Neurol Disord*. (2018) 11:1756286418805732. doi: 10.1177/1756286418805732

61. El Jammal T, Gerfaud-Valentin M, Sève P, Jamilloux Y. Les inhibiteurs de JAK: perspectives pour la médecine interne [JAK inhibitors: perspectives in internal medicine]. *Rev Med Interne*. (2019) 40:816–25. doi: 10.1016/j.revmed.2019.07.016

62. Talty R, Damsky W, King B. Treatment of cutaneous sarcoidosis with tofacitinib: a case report and review of evidence for Janus kinase inhibition in sarcoidosis. *JAAD Case Rep*. (2021) 16:62–4. doi: 10.1016/j.jdcr.2021.08.012

63. Scheinberg M, Maluf F, Wagner J. Steroid-resistant sarcoidosis treated with baricitinib. *Ann Rheum Dis*. (2020) 79:1259–60. doi: 10.1136/annrheumdis-2020-217271

64. Abrahamyan L, Feldman BM, Tomlinson G, Faughnan ME, Johnson SR, Diamond IR, et al. Alternative designs for clinical trials in rare diseases. *Am J Med Genet C Semin Med Genet*. (2016) 172:313–31. doi: 10.1002/ajmg.c.31533

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 31 October 2022
DOI 10.3389/fneur.2022.968322

OPEN ACCESS

EDITED BY
Davide Cossu,
University of Sassari, Italy

REVIEWED BY
Jiuliang Zhao,
Peking Union Medical College Hospital (CAMS), China
Vincent Van Pesch,
Université Catholique de Louvain, Belgium

*CORRESPONDENCE
Biman Kanti Ray
bimankantiray2019@gmail.com

†These authors have contributed equally to this work

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

RECEIVED 13 June 2022
ACCEPTED 11 October 2022
PUBLISHED 31 October 2022

CITATION
Das S, Ray BK, Chakraborty AP, Banerjee A, Pandit A, Das G and Dubey S (2022) Persistent "MRI-negative" lupus myelitis-disease presentation, immunological profile and outcome.
Front. Neurol. 13:968322.
doi: 10.3389/fneur.2022.968322

Persistent "MRI-negative" lupus myelitis-disease presentation, immunological profile and outcome

Shambaditya Das [1], Biman Kanti Ray [1]*, Arka Prava Chakraborty[1†], Abhirup Banerjee[2†], Alak Pandit[1], Gautam Das[1] and Souvik Dubey [1]

[1]Department of Neurology, Institute of Post Graduate Medical Education & Research, Bangur Institute of Neurosciences, Kolkata, India, [2]Department of General Medicine, Institute of Post Graduate Medical Education & Research, Kolkata, India

Introduction: Myelitis is the least common neuropsychiatric manifestation in systemic lupus erythematosus (SLE). Magnetic resonance imaging (MRI)-negative myelitis is even rarer. Here, we present the largest cohort of MRI-negative lupus myelitis cases to assess their clinical and immunological profiles and outcome.

Method: A single-center, observational study conducted over a period of 5 years (2017–2021) was undertaken to evaluate patients with MRI-negative lupus myelitis for the epidemiological, clinical, immunological, and radiological features at baseline and followed up at monthly intervals for a year, and the outcomes were documented. Among the 22 patients that presented with MRI-negative myelopathy (clinical features suggestive of myelopathy without signal changes on spinal-cord MRI [3Tesla], performed serially at the time of presentation and 7 days, 6 weeks, and 3 months after the onset of symptoms), 8 patients had SLE and were included as the study population.

Results: In 8 of 22 patients presenting with MRI-negative myelopathy, the etiology was SLE. MRI-negative lupus myelitis had a female preponderance (male: female ratio, 1:7). Mean age at onset of myelopathy was 30.0 ± 8.93 years, reaching nadir at 4.9 ± 4.39 weeks (Median, 3.0; range, 1.25–9.75). Clinically, cervical cord involvement was observed in 75% of patients, and 62.5% had selective tract involvement. The mean double stranded deoxyribonucleic acid, C3, and C4 titers at onset of myelopathy were 376.0 ± 342.88 IU/ml (median, 247.0), 46.1 ± 17.98 mg/dL (median, 47.5), and 7.3 ± 3.55 mg/dL (median, 9.0), respectively, with high SLE disease activity index 2,000 score of 20.6 ± 5.9. Anti-ribosomal P protein, anti-Smith antibody, and anti-ribonuclear protein positivity was observed in 87.5, 75, and 75% of the patients, respectively. On follow-up, improvement of myelopathic features with no or minimal deficit was observed in 5 of the 8 patients (62.5%). None of the patients had recurrence or new neurological deficit over 1-year follow-up.

Conclusion: Persistently "MRI-negative" lupus myelitis presents with white matter dysfunction, often with selective tract involvement, in light of high

disease activity, which follows a monophasic course with good responsiveness to immunosuppressive therapy. A meticulous clinical evaluation and a low index of suspicion can greatly aid in the diagnosis of this rare clinical condition in lupus.

KEYWORDS

myelitis in lupus, MRI-negative myelitis, MRI-negative lupus myelitis, systemic lupus erythematosus, neuropsychiatric systemic lupus erythematosus, selective tractopathy

Introduction

Systemic lupus erythematosus (SLE) affects multiple neurological systems (1). Neuropsychiatric SLE (NPSLE) encompasses a myriad of symptoms involving the central and/or peripheral nervous system during the disease progression of SLE. In 1999, the American College of Rheumatology (ACR) suggested 19 NPSLE syndromes involving the central or peripheral nervous system. Among these, "myelopathy" is used to specify injury of the spinal cord. It is termed as "myelitis" when spinal cord injury occurs due to inflammatory etiopathogenetic mechanisms (2, 3). It is characterized by neuronal damage resulting in paresis, sensory abnormalities, and autonomic dysfunction (4). Lupus myelitis is the least common presentation of NPSLE. However, its incidence is 1,000 times higher in patients with SLE than in the general population. Thus, this warrants keen attention during the evaluation of patients with SLE. In addition, it remains a serious complication of SLE, often portending a poor prognosis, and is difficult to diagnose and treat (4–6). Diagnostic challenges are compounded when clinically suspected lupus myelitis and the spinal imaging do not correlate (7, 8). The NPSLE case definition of lupus myelopathy does not consider the presence of abnormalities on spinal imaging as a mandatory criterion (3, 8). Magnetic resonance imaging (MRI)-negative lupus myelitis has rarely been reported, and literature pertaining to its clinical presentation, management, and outcome is sparse (1, 7, 8). This study was undertaken to assess the clinical characteristics, biochemical abnormalities, management, and outcome of MRI-negative lupus myelitis in the largest cohort of SLE patients to date (to our best knowledge).

Materials and methods

In a period of 5 years (2017–2021), 22 patients with MRI-negative myelopathy were either diagnosed or referred to the neuroinflammation clinic of our center (Bangur Institute of Neurosciences, IPGME&R, Kolkata). Among them 8 patients were diagnosed with lupus myelitis. The diagnosis of SLE was confirmed in accordance with the 2019 European Alliance of Associations for Rheumatology /ACR classification criteria. These 8 patients with SLE and MRI-negative myelopathy were included as the study population. A descriptive, observational study with prospective follow-up was conducted to decipher the clinical features, biochemical profile, management, and outcome of MRI-negative lupus myelitis. They were followed-up at monthly intervals with meticulous clinical (symptom analysis and neurological examination) and biochemical assessment for one year. Following a thorough etiological search (Supplementary material S1), out of the rest 14 patients with MRI-negative myelopathy, 10 were diagnosed with viral myelitis. Vitamin B12 deficiency, paraneoplastic disorder, spinal cord infarction, and Sjogren's syndrome were diagnosed in one patient each. Figure 1 shows the study design and enrolment of study population.

Each patient in the study cohort underwent MRI (Siemens 3Tesla MRI machine [Magnetom Verio DOT, 16 channels] using a standard quadrature head coil) imaging of the entire length of the spinal cord (Supplementary material S2) and brain at the time of presentation, followed by repeat spinal cord imaging 7 days later; furthermore, repeat spinal cord imaging was performed at 6 weeks and 3 months following the onset of myelopathic symptoms. An absence of signal change on spinal cord MRI on all four occasions, along with myelopathic evidence defined by the presence of acute/subacute clinical symptoms of motor and/or sensory changes, and/or sphincter dysfunction consistent with spinal cord lesion, corroborated at neurological examination, with exclusion of compressive cord lesion, were considered MRI-negative myelopathy.

They were evaluated under the following major headings: (a) epidemiological- sex, age at diagnosis of SLE, age at onset of myelopathic symptoms, and family history. (b) clinical features-time period between the onset of myelopathic symptoms to nadir, cross-sectional (tracts involved) and longitudinal (spinal cord level) localization, other concomitant central or peripheral nervous system involvement, previous episodes of neurological deficit, evidence of other organ involvement and its temporal relation to myelopathy, and SLE disease activity index-2000 (SLEDAI-2K) score. (c) immunological and

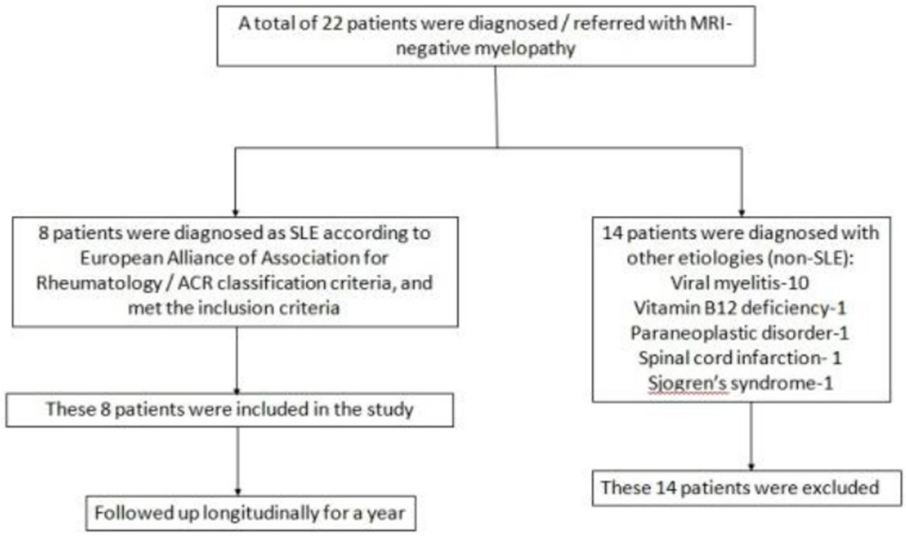


FIGURE 1
Flowchart showing the study design and enrolment of study population.

radiological features- double stranded DNA (dsDNA) titers (elevated if >100IU/ml), complement levels (decreased if C3 <90 mg/dL, C4 <10 mg/dL); anti-ribonucleoprotein (RNP), anti-ribosomal P protein (Rib-P), anti-Smith antibody (Sm), anti-Sjögren's-syndrome-related antigen A autoantibody (SS-A), anti-Sjögren's-syndrome-related antigen B autoantibody (SS-B) positivity; anti-phospholipid antibodies (lupus anticoagulant, β2-glycoprotein, and anti-cardiolipin) positivity; cerebrospinal fluid (CSF) pleocytosis (cell >5); protein levels (elevated if >45 mg/dL); anti-nuclear antibodies (ANA) positivity; and presence of brain imaging abnormalities. (d) management and outcome immunosuppressive therapy received- functional recovery (medical research council scale for muscle strength grading [MRC]), improvement in objective sensory symptoms, and bladder control (in terms of requirement of urinary catheter), all compared to the neurological status at the time of myelopathic presentation, SLEDAI-2K score at the latest follow-up, and any new-onset neurological deficit or recurrence of neurological symptoms.

The study was performed with the consent of the institutional ethical committee.

Statistical analysis

Data were summarized using routine descriptive statistics, namely mean and standard deviation for numerical variables that were normally distributed, median and interquartile range for skewed numerical variables, and counts and percentages for categorical variables. Numerical variables were compared between two groups by Student's independent samples *t*-test, if normally distributed, or by Mann-Whitney U test, if otherwise. Fischer's exact test or Pearson's Chi-square test were employed for intergroup comparisons of categorical variables. Analyses were two-tailed and statistical significance level was set at $p < 0.05$ for all comparisons.

Results

A female predominance was observed (male: female, 1:7) among the eight patients with MRI-negative lupus myelitis. The mean age at diagnosis of SLE and onset of myelopathy were 28.3 ± 8.24 years (median, 26.0; range, 21.75–33.5) and 30.0 ± 8.93 years (median, 28.0; range, 21.25– 37.0), respectively. The mean latency from diagnosis of SLE to onset of myelopathy was 24.5 ± 43.98 months (median, 11.5; range, 3.25– 19.5). The mean time period from onset of myelopathic symptoms to nadir was 4.9 ± 4.39 weeks (median, 3.0; range, 1.25– 9.75). The clinical characteristics suggestive of cervical cord involvement were found to be the most common (75%), followed by dorsal cord involvement, seen in 25% of patients. Selective tract involvement, affecting only the motor and autonomic tracts, was observed in 62.5% of our patients, while the rest had evidence of involvement of all three tracts. Concomitant involvement of other central or peripheral nervous system was observed in 75% of the patients, the most common being polyradiculoneuropathy (37.5%). Myelopathy occurred after other SLE-specific organ involvement in 67.5% of the patients. None of the patients had neurological manifestations prior to the onset of index myelopathic symptoms. Constitutional and mucocutaneous manifestations were seen in all patients (100%);

furthermore, there was involvement of musculoskeletal system in 87.5%, hematological and renal involvement in 50% each, and one patient had serosal (pleural) involvement. Among the 4 out of 8 patients with lupus nephritis, two patients denied renal biopsy and the other two had diffuse lupus nephritis (class IV). Those with hematological involvement (4 out of 8), autoimmune hemolytic anemia was seen in one, and leukopenia was found in three and thrombocytopenia was documented in two patients. The mean SLEDAI-2K score at the time of presentation was 20.6 ± 5.9, while it was 0.7 ± 0.95 at the time of most recent follow-up at 1 year. The mean dsDNA, C3, and C4 titers were 376.0 ± 342.88 IU/ml (median, 247.0; range, 177.75–501.5), 46.1 ± 17.98 mg/dL (median, 47.5; range, 22.0– 62.75), and 7.3 ± 3.55 mg/dL (median, 9.0; range, 3.38–10.00), respectively. CSF pleocytosis was seen in 50% patients, ranging from 10–40 cells; increased CSF protein levels were seen in all patients with a mean of 84.9 ± 41.23 mg/dL (median, 65.0; range, 57.25–128.75), and no patient showed CSF ANA positivity. IgG index was raised in 62.5% patients, none had OCB positivity. Among other autoantibodies, Rib-P positivity was observed in 87.5% of patients, and Sm and RNP positivity were observed in 75% of patients. None of the patients demonstrated antiphospholipid antibodies positivity. SS-A positivity was seen in 12.5% patient; while Scl-70, PM-Scl 100, Jo-1, centromere B, nucleosomes, histones, AMA-M2 and SS-B were negative in all patients. Brain imaging abnormalities were detected in 25% of patients. Intravenous pulse methyl prednisolone (IVMP) and cyclophosphamide were instituted in all patients except one, who died due to macrophage activation syndrome (MAS)-related complications prior to completion of IVMP or administration of cyclophosphamide. Two of the eight (25%) patients had an unsatisfactory response to the initial therapy. They were further subjected to rituximab therapy, with one patient receiving plasmapheresis. Among the seven surviving patients, five showed significant improvement with no or minimal neurological deficits. Two patients who received additional rituximab therapy had moderate residual neurological deficits. None of the patients had recurrence or the appearance of new neurological symptoms during the one year follow-up period. The results are summarized in Tables 1, 2.

MRI-negative myelopathy due to etiologies other than lupus had a significantly shorter time to nadir of myelopathic symptoms [6.8 ± 12.74 (median, 1.0; range, 0.5–5.75); p 0.010], lesser concomitant involvement of other central or peripheral nervous system (21.4%; p 0.026) and lesser magnitude of CSF protein elevation [54.6 ± 10.56 mg/dL (median, 54.5; range, 48.75–61.0); p 0.016] as compared to MRI-negative lupus myelitis (Table 3).

Illustrative case: A 45-year-old female had complaints of quadriparesis for last 7 days. It started as an acute retention of urine and paraparesis, followed by bilateral upper limbs weakness from the next day, without any sensory and cranial nerve symptoms. She also had history of oral ulcers, malar rash, alopecia, photosensitivity and symmetrical small joint pain and swelling for last 2.5 months along with low grade fever and pedal edema for last 1 month.

Neurological examination revealed diminished muscle power (MRC, Upper limbs: proximally and distally 4-/5; lower limbs: proximally 2/5, distally 3/5), spasticity of all 4 limbs except for hypotonia near both ankle joints, pan-hyper-reflexia except for absent ankle jerk, and bilateral extensor plantar response.

MRI spine didn't reveal any cord signal change on repeated imaging (Figure 2). Nerve conduction study showed acquired motor axonal polyradiculoneuropathy. CSF analysis had pleocytosis with mildly elevated protein. Biochemical investigations revealed ANA, anti-dsDNA, Rib-P, Sm, and RNP positivity with hypocomplementemia. There was presence of urinary RBC cast with macro-albuminuria on further searching for organ involvement. Patient denied permission for renal biopsy.

She was given 3 days of pulse IVMP therapy (1000 mg/day for 3 days) along with injection Cyclophosphamide (1 gm/month for 6 cycles). Oral Prednisolone was started at 1 mg/kg/day dosing and gradually tapered to 5mg/day by 6 months. Oral Mycophenolate mofetil (2 gm/day) was started following completion of Cyclophosphamide. She had substantial functional recovery in terms motor power (MRC, Upper limbs: proximally and distally 5/5; lower limbs: proximally 4+/5, distally 4/5) and bladder control at the end of 1 year.

Discussion

Myelitis, which is considered a serious complication of SLE, is one of its least common neuropsychiatric manifestations, occurring in 1–2% of patients with SLE. This may be due to the inherent diagnostic and therapeutic challenges and the increased risk of morbidity and mortality (4, 6). The diagnosis of lupus myelitis is even more obscure in the absence of correlation with imaging (9). Previous observations have suggested that lupus myelitis is the presenting manifestation in nearly half of the patients (6, 10). However, for unknown reasons, our observation suggested that MRI-negative lupus myelitis often occurred after (62.5%) the evidence of other SLE-specific organ involvement. The mean age for lupus myelitis varied from 25–42 years in previous studies (6, 11, 12). A similar predilection toward young adults was also observed in our cohort. Patients with MRI-negative myelitis were predominantly females (87.5%), in line with previous observations and a female predilection for SLE in general (13).

Birnbaum et al. classified lupus myelitis into gray matter and white matter myelitis. Gray matter myelitis is hyperacute and rapidly deteriorates to clinical nadir within 6 hours. It has a severe clinical presentation, with flaccidity and hyporeflexia, and is often monophasic. It more frequently presents with LETM and significant CSF abnormalities. It often occurs in

TABLE 1 Clinical features of our cohort of MRI-negative myelitis in SLE.

Case	Gender	Age at diagnosis of SLE(years)	Myelopathy						Previous episodes of neurological manifestation	Other system involved	SLEDAI-2K at presentation
			Age at onset (years)	Time to nadir (weeks)	Cross-sectional localization	Longitudinal localization	Other central or peripheral nervous system involved	Temporal relation to other organ involvement			
1.	Female	21	20	3	Motor, sensory, autonomic	Cervical	Cerebral cortex, Radiculoneuropathy	Preceding	Nil	Constitutional, Mucocutaneous, Musculoskeletal, Hematological	23
2.	Female	26	37	11	Motor, sensory, autonomic	Cervical	Neuropathy	succeeding	Nil	Constitutional, Mucocutaneous, Musculoskeletal, Lupus Nephritis	13
3.	Female	26	27	1	Motor, autonomic	Cervical	Cerebral cortex	succeeding	Nil	Constitutional, Mucocutaneous, Musculoskeletal, Hematological	28
4.	Female	35	37	12	Motor, autonomic	Dorsal	Nil	succeeding	Nil	Constitutional, Mucocutaneous, Musculoskeletal	15
5.	Female	20	20	3	Motor, autonomic	Cervical	Radiculoneuropathy	simultaneous	Nil	Constitutional, Mucocutaneous, Hematological, Lupus Nephritis, Serosal	27
6.	Female	24	25	6	Motor, sensory, autonomic	Dorsal	Neuropathy	succeeding	Nil	Constitutional, Mucocutaneous, Musculoskeletal	15
7.	Male	29	29	2	Motor, autonomic	Cervical	Nil	succeeding	Nil	Constitutional, Mucocutaneous, Musculoskeletal, Hematological, Lupus Nephritis	19
8.	Female	45	45	1	Motor, autonomic	Cervical	Radiculoneuropathy	simultaneous	Nil	Constitutional, Mucocutaneous, Musculoskeletal, Lupus Nephritis	25

10.3389/fneur.2022.968322

TABLE 2 Investigational details, mangement and follow-up of our cohort of MRI-negative myelitis in SLE.

Case	dsDNA titer	Complement levels	Other antibodies detected positive	Anti-phospholipid antibodies	CSF Analysis			Brain imaging	Therapy	Functional recovery	SLEDAI−2K at last follow-up	New-onset neurological defict or recurrence
					Pleocytosis	Protein	ANA					
1.	Elevated	Decreased	Rib-P	Negative	Present (Mononuclear)	Elevated	Negative	Unidentified bright objects	Steroid, cyclophosphamide, MMF, HCQS	(+)	0	(-)
2.	Elevated	Decreased	RNP, Sm, Rib-P	Negative	Absent	Elevated	Negative	Normal	Steroid, cyclophosphamide, Rituximab, HCQS	(+)	0	(-)
3.	Elevated	Decreased	RNP, Sm, SS-A, Rib-P	Negative	Absent	Elevated	Negative	Normal	Steroid, cyclophosphamide,MMF, HCQS	(+)	1	(-)
4.	Elevated	Decreased	RNP, Sm, Rib-P	Negative	Absent	Elevated	Negative	Normal	Steroid, plasmapheresis, cyclophosphamide, Rituximab, HCQS	(+)	2	(-)
5.	Elevated	Decreased	RNP, Sm, Rib-P	Negative	Present (Mononuclear)	Elevated	Negative	Normal	Steroid, HCQS	-	-	-
6.	Elevated	Decreased	RNP, Sm	Negative	Present (Mononuclear)	Elevated	Negative	Normal	Steroid, cyclophosphamide,MMF, HCQS	(+)	0	(-)
7.	Elevated	Decreased	Rib-P	Negative	Absent	Elevated	Negative	Normal	Steroid, cyclophosphamide,MMF, HCQS	(+)	0	(-)
8.	Elevated	Decreased	RNP, Sm, Rib-P	Negative	Present (Mononuclear)	Elevated	Negative	Unidentified bright objects	Steroid, cyclophosphamide,MMF, HCQS	(+)	2	(-)

ANA, Antinuclear antibody; Anti-dsDNA, Anti-double stranded DNA antibody; HCQS, Hydroxychloroquine; MMF, Mycophenolate Mofetil; RNP, Anti Ribonucleoprotein; Rib-P, Anti-Ribosomal P protein; Sm, Anti Smith antibody; SS-A, Anti–Sjögren's-syndrome-related antigen A autoantibody; SS-B, Anti–Sjögren's-syndrome-related antigen B autoantibody; SLEDAI-2K, Systemic Lupus Erythematosus Disease Activity Index 2000; UBO, Unidentified bright objects.

10.3389/fneur.2022.968322

TABLE 3 Comparison of baseline clinical and biochemical features between MRI-negative lupus myelitis and MRI-negative myelopathy due to other etiologies.

Parameters		MRI-negative lupus myelitis ($n = 8$)	MRI-negative myelopathy due to other etiologies ($n = 14$)	*P*-value
1. Gender (Male: Female)		1:7	8:6	
2. Mean age at onset of myelopathy (years)		30.0 ± 8.93 (median, 28.0; range, 21.25– 37.0)	30.0 ± 12.05 (median, 28.0; range, 19.75–36.0)	1.000
3. Time to nadir (days)		34.3 ± 32.42 (median, 20.5; range, 8.75–67.75)	6.8 ± 12.74 (median, 1.0; range, 0.5– 5.75)	0.010
4. Selective tract involvement		62.5%	28.6%	0.187
5. Other central or peripheral nervous system involved		75%	21.4%	0.026
6. Previous episodes of neurological manifestation		Nil	Nil	
7. CSF analysis	(a) Pleocytosis	50%	57.1%	1.000
	(b) Elevated protein	84.9 ± 41.23 mg/dL (median, 65.0; range, 57.25–128.75)	54.6 ± 10.56 mg/dL (median, 54.5; range, 48.75–61.0)	0.016

the background of severe systemic inflammation, with a high SLEDAI-2K score, dsDNA titers, and β2-glycoprotein positivity. It is poorly responsive to immunosuppressive therapy and often results in incomplete or poor recovery. White matter myelitis, on the other hand, is characterized by spasticity and hyperreflexia, and the clinical nadir is not reached until 72 hours. It has lower dsDNA positivity. It is more responsive to immunosuppressive therapy and usually has a good prognosis. It is more likely to meet the neuromyelitis optica spectrum disorder criteria and has a higher recurrence rate and lupus anticoagulant positivity (2, 4, 11).

Similar to previous observations, our patients also presented with symptoms related to the involvement of the bilateral motor, sensory, and/or autonomic tracts of variable severity and symmetricity (2, 4). However, it is interesting to note that the majority of our patients (62.5%) had selective tract involvement along the centro-anterior cord, affecting the motor and autonomic tracts, suggesting a predominantly white matter myelitis, according to Birnbaum classification. White matter myelopathy with selective tractopathy has classically been described in few conditions such as multiple sclerosis, paraneoplastic myelopathy, and vitamin B12 deficiency (1, 14). Thus, lupus myelitis, especially in those with lack of correlation with imaging, might be an important consideration in tract-specific white matter myelitis (1).

The proposed pathogenesis for lupus myelitis includes: (i) mechanisms related to anti-phospholipid antibodies, especially β2-glycoprotein, that may lead to thromboembolic effect on microcirculation of spine, or it may interact with certain spinal cord antigens leading to "co-operation between antibodies" and aquaporin-4 (AQP-4) synthesis induction, or may have direct cytotoxic effects. (ii) small vessel vasculitis leading to cord ischemia and necrosis (better explains longitudinal extensive transverse myelitis [LETM] in SLE). (iii) change in blood-brain barrier (BBB) due to complex interplay of overlapping autoantibodies; and (iv) co-clustering of various intertwining pathophysiological mechanisms (cord inflammation, venous hypertension, and cord ischemia) resulting in hemodynamic compromise (1, 2, 4, 6, 10–12, 15).

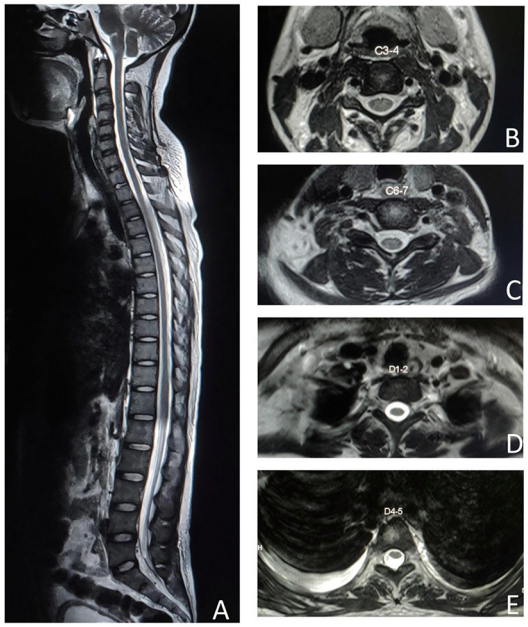


FIGURE 2
MRI spine T2 weighted image shows no spinal cord signal changes in the sagittal section **(A)**, cervical axial sections at C3–4 **(B)**, C6–7 **(C)**, dorsal axial sections at D1–2 **(D)**, and D4–5 **(E)**.

Lupus myelitis has been seen to develop even during the stages of low disease activity in 1/3rd of the patients (2, 4, 11). However, all patients with MRI-negative lupus myelitis in our cohort showed high disease activity at the onset of myelopathy. CSF analysis for lupus myelitis can vary. It can range from normal (20–33%) to marked pleocytosis, increased protein levels, and hypoglycorrhachia, mimicking bacterial meningitis (2, 4, 11, 12). All our patients had increased CSF protein, and half of them showed pleocytosis (all mononuclear predominant); however, none had more than 50 cells. Rib-P is considered the best biomarker for the diagnosis of NPSLE, and it strongly correlates with NPSLE. Both Rib-P and Sm have been implicated in BBB dysfunction and subsequent aberrant immune downsignaling (16). In our study, we observed high frequencies of antibodies against Rib-P (87.5%), Sm (75%), and RNP (75%). This is a much higher frequency than that of SLE, raising speculations regarding their possible pathogenetic association and causative implications in MRI-negative lupus myelitis. Thus, it may be conjectured that this variant of lupus myelitis is probably an accomplishment of systemic inflammation associated with lupus. The absence of anti-phospholipid antibodies in all patients undermines their role in etiopathogenesis.

Classically, lupus myelitis is acute in onset and progresses to its maximum clinical severity within hours to months (4, 17). Evolution of MRI-negative lupus myelitis in our patients was almost always subacute. The most noteworthy observation was the absence of hyperacute to acute presentation in our cohort, contrary to previous literature, where it has been frequently observed (2, 4, 11, 17). Clinically, the cervical region was the most commonly affected site in our cohort, differing from previous notions of frequent thoracic segment involvement in lupus myelitis. It has been argued that inherent vascular anatomy could be responsible for this thoracic cord predilection (4, 6, 10, 11, 13). The absence of propensity for thoracic cord involvement as well as the absence of hyperacute presentation in our cohort further strengthens our assumption of a lower likelihood of vascular insult-induced myelopathy in this subtype.

Lupus myelitis was commonly associated with concomitant involvement of other neurological systems in our cohort (75%). This value was much higher than that reported in previous studies (13). Associated central and peripheral nervous system manifestations were observed, with the latter being more common. Axonal polyradiculoneuropathy was the most commonly associated condition in our cohort, in line with its previously noted common occurrence in SLE (18).

MRI of the spinal cord with gadolinium contrast administration is considered to be the most sensitive test for the assessment of myelopathy (7). Negative spinal cord imaging is not an unusual phenomenon during the evaluation of clinically suspected acute-to-subacute myelopathy (7–9). As many as 1/5th of patients with myelopathy may not be supported by an obvious lesion on cord imaging (7). This is more commonly seen in idiopathic transverse myelitis (5%), paraneoplastic myelopathy (35%), myelin oligodendrocyte glycoprotein antibody-associated disease (MOGAD), and GFAP-IgG, glycine, and glutamic acid decarboxylase-65 receptor-associated myelopathy. Spinal cord infarction can have an initial negative MRI in 24% of patients, although a hyperacute clinical presentation and absent CSF pleocytosis usually aid in its distinction (7, 9). Sechi et al. postulated that imaging timing (transient lesion being missed on late imaging, or an early imaging failing to detect an evolving lesion) and less sensitivity of MRI (to detect the subtle signal changes related to inflammation of the cord or its surrounding meninges) are the probable reasons for the negative MRI results in MOGAD myelitis (9). An extrapolation to MRI-negative lupus myelopathy may not be far-fetched. However, our cohort underwent repeated imaging with standard sensitivity and negative results. Thus, the absence of imaging abnormalities in MRI-negative lupus myelitis beyond the early stages might suggest a functional disruption in the white matter tracts of the cord without any discernible structural insult. Although two of our patients had evidence of few scattered, tiny white matter changes on brain MRI, primary central nervous system demyelination seemed less likely in light of non-fulfillment of their clinical and biochemical diagnostic criteria (19, 20).

Several novel biomarkers that correlate with neuronal damage have emerged lately. Neurofilament protein levels in blood and CSF have shown promise in assessing the disease onset and progression of nervous system injury, including in Multiple sclerosis (21). Recent evidence has suggested the use of Glial fibrillary acidic protein (GFAP) in detecting subtle injury to CNS (22). The use of these potential biomarkers may contribute to the diagnostic accuracy in MRI-negative lupus myelitis, wherein conventional structural imaging fails to detect the evidence of pathology. Future research in this direction is warranted.

The combination of intravenous glucocorticoids and cyclophosphamide has been the mainstay treatment for lupus myelitis (2, 4, 23). In our cohort, 5 of the 8 patients showed significant improvement with intravenous glucocorticoids and cyclophosphamide therapy. One patient succumbed to MAS in the immediate acute phase just following the initiation of intravenous glucocorticoids. Remaining 2 out of the 8 patients failed to show any significant improvement following initial glucocorticoid and cyclophosphamide administration. Owing to its proposed role in refractory cases (2, 4, 23), plasmapheresis was instituted in one of the two non-responder patients (other patients did not consent to it). Mild improvement in myelopathic features was observed following plasmapheresis. Both patients were further administered rituximab. Clinical improvement was documented in both patients at the subsequent follow-up; although, residual disability persisted. Historically, nearly more than 1/3rd of patients with lupus myelitis have a good prognosis with full recovery or minimal

sequelae with appropriate therapy, while about 2/3rd patients suffer from moderate-to-severe disability. LETM has worse prognosis compared to acute transverse myelitis. The previously described poor prognostic factors include clinically severe deficits at onset, need for urinary catheterization, increased number and extension of spinal cord lesions ($\geq$ 4 segments), CSF abnormalities, failure to add cyclophosphamide in a timely manner, and absence of hydroxychloroquine therapy (2, 4, 6, 10, 11, 17). An assessment of the prognostic factors of MRI-negative lupus myelitis in our cohort can be biased due to the small number of patients. However, note must be taken of the fact that both of our patients with residual disabilities on follow-up had a more subacute to chronic evolution of myelopathy in comparison to patients with better functional recovery. The risk of recurrence has been reported to be 18–50%, with at least one episode recurring within a year, despite optimal therapy. None of our patients experienced relapse during the 1-year follow-up period. Positivity to AQP-4 and SS-A/Ro, which are known to increase the risk of recurrence, was absent in our cohort (2, 4, 6, 10).

All our patients with MRI-negative lupus myelitis had a more indolent course, less severe presentation with upper motor neuron (UMN) spasticity and hyperreflexia, milder CSF abnormalities, and relatively good responsiveness to immunosuppressive therapy, which was comparable to the manifestations of white matter myelitis. However, it shared some features with gray matter myelitis. It was monophasic, with a fever prodrome, occurring in light of high disease activity with increased dsDNA titers. Hence, we propose a new subtype of white-matter myelitis in lupus, "MRI-negative myelitis with selective tract involvement," that occurs in light of high disease activity, often with Rib-P protein positivity, and follows a similar indolent, monophasic course with good responsiveness to immunosuppressive therapy. Although the proposed new phenotype of lupus myelitis shares major similarities with white matter myelitis, the absence of all clinical and biochemical characteristics of gray matter myelitis must not be considered as a rule.

It may be emphasized that often the milder symptoms of myelopathy can be misinterpreted. Mild paresis may be attributed to generalized weakness from the burden of systemic illness. UMN-related bladder symptoms following a cord injury share great similarity with symptoms of urinary tract infection, and sensory symptoms are often vague and non-specific (14, 24, 25). The diagnostic dilemma of myelitis becomes compounded in the absence of correlating MRI findings (8, 26). Thus, diagnosis of MRI-negative lupus myelitis is often difficult, and only meticulous history taking and clinical examination with a low threshold of suspicion can help identify this entity.

Although there is a scope for selection bias due to prior diagnosis of SLE in 5 out of 8 patients in our cohort, the strength of the study lies in the sizeable number of this relatively rare condition of MRI-negative lupus myelitis patients included in the study population.

Conclusion

MRI-negative lupus myelitis may be an under-reported entity owing to the absence of correlating radiological findings. A high resolution MRI spinal cord imaging with appropriate sequences is essential before its attribution as MRI-negative myelitis. The clinical features of lupus myelitis can mimic other commonly encountered complications of SLE and pose a diagnostic dilemma. High clinical suspicion and meticulous clinical evaluation are mandated for diagnosis. It is mostly associated with high disease activity and a monophasic course. It should be emphasized that timely identification of this complication is of paramount significance, as most cases respond well to appropriately chosen immunosuppressive therapy.

Data availability statement

The original contributions presented in the study are included in the article/Supplementary material, further inquiries can be directed to the corresponding author.

Ethics statement

The studies involving human participants were reviewed and approved by Institute of Post Graduate Medical Education & Research, Kolkata, India. The patients/participants provided their written informed consent to participate in this study. Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

Author contributions

SDa: conceptualization-lead, data curation-lead, formal analysis-equal, investigation-equal, methodology-equal, resources-equal, supervision-equal, visualization-equal, statistical analysis-equal, writing—original draft-lead, and writing—review and editing-lead. BR: conceptualization-equal, formal analysis-equal, investigation-equal, methodology-equal, resources-equal, supervision-equal, visualization-equal, and writing—review and editing-equal. AC and AB: data curation-equal and writing—review and editing-equal. AP: conceptualization-equal, supervision-equal, visualization-equal, and writing—review and editing-equal. GD: supervision-equal, visualization-equal, and writing—review and editing-equal. SDu: conceptualization-equal, formal analysis-equal, investigation-equal, methodology-equal, resources-equal, supervision-lead, visualization-equal, and writing—review and editing-lead. All authors agreed upon the final form of the manuscript before submission.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2022.968322/full#supplementary-material

References

1. Das S, Ray BK, Pandit A, Dubey S. MRI negative myeloradiculoneuropathy unmasking systemic lupus erythematosus. *Rheumatology.* (2021) 60:e318–20. doi: 10.1093/rheumatology/keab216

2. Li XY, Xiao HB, Pai P. Myelitis in systemic lupus erythematosus. *J Clin Neurosci.* (2017) 44:18–22. doi: 10.1016/j.jocn.2017.06.003

3. Liang MH, Corzillius M, Bae SC, Lew RA, Fortin PR, Gordon C, et al. The American College of Rheumatology nomenclature and case definitions for neuropsychiatric lupus syndromes. *Arthritis Rheum.* (1999) 42:599–608. doi: 10.1002/1529-0131(199904)42:4<599::AID-ANR2>3.0.CO;2-F

4. Chiganer EH, Hryb JP, Carnero Contentti E. Myelitis and lupus: clinical manifestations, diagnosis and treatment. *Review Reumatol Clínica.* (2017) 13:344–8. doi: 10.1016/j.reumae.2016.06.005

5. Williams JN, Speyer CB, Kreps DJ, Kimbrough DJ, Costenbader K, Bhattacharyya S. Spinal cord syndromes in patients with systemic lupus erythematosus: differentiating lupus myelitis, neuromyelitis optica, and multiple sclerosis. *Lupus.* (2019) 28:1656–62. doi: 10.1177/0961203319886103

6. Kovacs B, Lafferty TL, Brent LH, DeHoratius RJ. Transverse myelopathy in systemic lupus erythematosus: an analysis of 14 cases and review of the literature. *Ann Rheum Dis.* (2000) 59:120–4. doi: 10.1136/ard.59.2.120

7. Wong SH, Boggild M, Enevoldson TP, Fletcher NA. Myelopathy but normal MRI: where next? *Pract Neurol.* (2008) 8:90–102. doi: 10.1136/jnnp.2008.144121

8. Monahan RC, Beaart HJL, Fronczek R, Terwindt GM, Beaart-Van de. Voorde LJJ, de Bresser J, et al. Suspected transverse myelitis with normal MRI and CSF findings in a patient with lupus: What to do? a case series and systematic review. *Neuropsychiatr Dis Treat.* (2020) 16:3173–86. doi: 10.2147/NDT.S267000

9. Sechi E, Krecke KN, Pittock SJ, Dubey D, Lopez-Chiriboga AS, Kunchok A, et al. Frequency and characteristics of MRI-negative myelitis associated with MOG autoantibodies. *Mult Scler J.* (2021) 27:303–8. doi: 10.1177/1352458520907900

10. Saison J, Costedoat-Chalumeau N, Maucort-Boulch D, Iwaz J, Marignier R, Cacoub P, et al. Systemic lupus erythematosus-associated acute transverse myelitis: manifestations, treatments, outcomes, and prognostic factors in 20 patients. *Lupus.* (2015) 24:74–81. doi: 10.1177/0961203314547795

11. Birnbaum J, Petri M, Thompson R, Izbudak I, Kerr D. Distinct subtypes of myelitis in systemic lupus erythematosus. *Arthritis Rheum.* (2009) 60:3378–87. doi: 10.1002/art.24937

12. Hryb JP, Chiganer E, Contentti EC, Pace JL Di, Lessa C, Perassolo MB. Myelitis in systemic lupus erythematosus: clinical features, immunological profile and magnetic resonance imaging of five cases. *Spinal Cord Ser Cases.* (2016) 2:16005. doi: 10.1038/scsandc.2016.5

13. Costallat BL, Ferreira DM, Costallat LTL, Appenzeller S. Myelopathy in systemic lupus erythematosus: clinical, laboratory, radiological and progression findings in a cohort of 1,193 patients. *Rev Bras Reumatol (English Ed.* (2016) 56:240–51. doi: 10.1016/j.rbre.2016.03.006

14. S D, S D, A P, BK R. Myeloradiculoneuropathy due to vitamin B 12 deficiency: an unusual clinical and radiological presentation. *BMJ Case Rep.* (2021) 14:239415. doi: 10.1136/bcr-2020-239415

15. Oiwa H, Kuriyama A, Matsubara T, Sugiyama E. Clinical value of autoantibodies for lupus myelitis and its subtypes: a systematic review. *Semin Arthritis Rheum.* (2018) 48:214–20. doi: 10.1016/j.semarthrit.2018.02.004

16. Manca E. Autoantibodies in Neuropsychiatric Systemic Lupus Erythematosus (NPSLE): can they be used as biomarkers for the differential diagnosis of this disease? *Clin Rev Allergy Immunol.* (2021) 2021 1:1–16. doi: 10.1007/s12016-021-08865-2

17. Schulz SW, Shenin M, Mehta A, Kebede A, Fluerant M, Derk CT. Initial presentation of acute transverse myelitis in systemic lupus erythematosus: demographics, diagnosis, management and comparison to idiopathic cases. *Rheumatol Int.* (2011) 32:2623–7. doi: 10.1007/s00296-011-2053-1

18. Bortoluzzi A, Silvagni E, Furini F, Piga M, Govoni M. Peripheral nervous system involvement in systemic lupus erythematosus: a review of the evidence. *Clin Exp Rheumatol.* (2019) 37:146–55.

19. Wingerchuk DM, Banwell B, Bennett JL, Cabre P, Carroll W, Chitnis T, et al. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. *Neurology.* (2015) 85:177. doi: 10.1212/WNL.0000000000001729

20. Thompson AJ, Banwell BL, Barkhof F, Carroll WM, Coetzee T, Comi G, et al. Diagnosis of multiple sclerosis: 2017 revisions of the McDonald criteria. *Lancet Neurol.* (2018) 17:162–73. doi: 10.1016/S1474-4422(17)30470-2

21. Yuan A, Nixon RA. Neurofilament proteins as biomarkers to monitor neurological diseases and the efficacy of therapies. *Front Neurosci.* (2021) 15:1242. doi: 10.3389/fnins.2021.689938

22. Abdelhak A, Foschi M, Abu-Rumeileh S, Yue JK, D'Anna L, Huss A, et al. Blood GFAP as an emerging biomarker in brain and spinal cord disorders. *Nat Rev Neurol.* (2022) 18:158–72. doi: 10.1038/s41582-021-00616-3

23. Bertsias GK, Ioannidis JPA, Aringer M, Bollen E, Bombardieri S, Bruce IN, et al. EULAR recommendations for the management of systemic lupus erythematosus with neuropsychiatric manifestations: report of a task force of the EULAR standing committee for clinical affairs. *Ann Rheum Dis.* (2010) 69:2074–82. doi: 10.1136/ard.2010.130476

24. Ahn GE, Ramsey-Goldman R. Fatigue in systemic lupus erythematosus. *Int J Clin Rheumtol.* (2012) 7:217. doi: 10.2217/ijr.12.4

25. Tan CW, Chlebicki MP. Urinary tract infections in adults. *Singapore Med J.* (2016) 57:485–90. doi: 10.11622/smedj.2016153

26. Ghosh R, Das S, De K, Dubey S. Benito-Leon J. Magnetic resonance imaging-negative myeloneuropathy and bilateral facial paresis unfurling systemic lupus erythematosus. *Qatar Med J.* (2022) 3:42. doi: 10.5339/qmj.2022.42

frontiers | Frontiers in Immunology

TYPE Case Report
PUBLISHED 08 December 2022
DOI 10.3389/fimmu.2022.1047992

OPEN ACCESS

EDITED BY
Ilka Kleffner,
University Hospital Bochum GmbH,
Germany

REVIEWED BY
Tobias Moser,
University Hospital Salzburg, Austria
Shuhei Nishiyama,
Harvard Medical School, United States

*CORRESPONDENCE
Weihe Zhang
z_weihe@hotmail.com
Dantao Peng
Peng13910908579@163.com

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Immunology

RECEIVED 19 September 2022
ACCEPTED 28 November 2022
PUBLISHED 08 December 2022

CITATION
Zhang W, Jiao Y, Jiao J, Jin M and Peng D (2022) Successful treatment of rituximab-unresponsive elderly-onset neuromyelitis optica spectrum disorder and hypogammaglobulinemia with ofatumumab plus intravenous immunoglobulin therapy in a patient with mutant *FCGR3A* genotype: A case report.
Front. Immunol. 13:1047992.
doi: 10.3389/fimmu.2022.1047992

Successful treatment of rituximab-unresponsive elderly-onset neuromyelitis optica spectrum disorder and hypogammaglobulinemia with ofatumumab plus intravenous immunoglobulin therapy in a patient with mutant *FCGR3A* genotype: A case report

Weihe Zhang[1]*, Yujuan Jiao[1], Jinsong Jiao[1], Ming Jin[2] and Dantao Peng[1]*

[1]Department of Neurology, China-Japan Friendship Hospital, Beijing, China, [2]Department of Ophthalmology, China-Japan Friendship Hospital, Beijing, China

Background: Elderly-onset neuromyelitis optica spectrum disorder (NMOSD) is a rare entity that poses a therapeutic challenge. We report a case of elderly-onset NMOSD with mutant *FCGR3A* genotype who was successfully treated with ofatumumab after multiple episodes of relapse.

Case Report: The patient was a 67-year-old woman who was diagnosed with NMOSD with high disease activity. She experienced six episodes of relapse over a period of 2 years despite immunosuppressant therapy with intravenous rituximab (RTX), oral steroids, mycophenolate mofetil, and tacrolimus. At the last relapse, she was unable to walk and developed immunosuppressant-induced hypogammaglobulinemia. Based on the insufficient B cell depletion and *FCGR3A-FF* genotype carrier, the patient was diagnosed as RTX non-responder. After subcutaneous ofatumumab plus intravenous immunoglobulin replacement therapy, she was able to walk independently, and experienced no further relapse. Ofatumumab was well-tolerated, and sufficiently depleted the circulating B cells.

Conclusion: Ofatumumab might be an effective alternative in RTX-unresponsive NMOSD, and seems to be safe in elderly patients.

KEYWORDS

neuromyelitis optica spectrum disorder, ofatumumab, rituximab, hypogammaglobulinemia, case report

Introduction

Neuromyelitis optica spectrum disorder (NMOSD) is an autoimmune disorder of the central nervous system that typically occurs in young women. Elderly-onset NMOSD is a rare entity, but is associated with the same relapse frequency and more severe attacks (1). The therapeutic decision-making is more challenging in elderly patients due to multiple comorbidities and high risk of drug-induced side effects. Ofatumumab is the first fully human anti-CD20 monoclonal antibody which has been approved for relapsing forms of multiple sclerosis. However, the efficacy of ofatumumab against NMOSD is unclear. Recently, ofatumumab was reported to be effective in a pediatric patient with NMOSD who failed to respond to rituximab (RTX) (2). Herein, we describe a case of RTX-unresponsive elderly-onset NMOSD with mutant *FCGR3A* genotype that also showed favorable response to ofatumumab. Moreover, the immunosuppressant-induced hypogammaglobulinemia posed a therapeutic challenge in this patient.

Case presentation

A 67-year-old woman was admitted to the China-Japan Friendship Hospital (Beijing, China) due to bilateral needle-like pain in upper back, lower extremity weakness, and sphincter dysfunction for one month. She had undergone thyroid cancer surgery four years ago, and was treated with oral levothyroxine sodium (Euthyrox®, 75 μg per day) to maintain normal thyroid function. She was also diagnosed with Sjogren Syndrome, but had not received any immunosuppressive treatment prior to admission. On admission, she exhibited malaise and was not able to walk; muscle strength in the right and left lower extremity was graded as 2/5 and 4/5 (Medical Research Council), respectively. There was bilateral tendon hyperreflexia and positive Babinski sign. In addition, she had hypoesthesia at the level of 6th thoracic segment. The Expanded Disability Status Scale (EDSS) score at nadir was 6.5. The diagnosis of NMOSD was established based on seropositivity for AQP4-IgG and longitudinally extensive transverse myelitis (LETM, continuous spinal cord lesions extending from cervical to thoracic segments) (Figure 1). No other abnormalities were detected on additional specific tests for autoimmune disorders (serum myelin oligodendrocyte glycoprotein antibodies and glial fibrillary acidic protein antibodies), metagenomic next-generation sequencing of cerebrospinal fluid for microbial infection, hematological examination, serum angiotensin-converting enzyme, tumor markers, and immunoglobulin levels. After 5 cycles of plasma exchange and 5 infusions of intravenous immunoglobulin (IVIg), the patient showed marked alleviation of symptoms and regained the ability to walk (EDSS=3.5). RTX standardized induction protocol (375 mg/m^2 infused once weekly for 4 weeks) was used to prevent relapse. Unfortunately, two months after RTX induction therapy, she suffered another severe attack with left optic neuritis (visual acuity=finger count) with insufficient depletion of CD19$^+$B cells (Figure 2). Based on the *FCGR3A-FF* genotype carrier, we believed that the patient was RTX non-responder. Sanger sequencing to determine *FCGR3A-V158F (rs396991)* gene polymorphism was performed by an independent medical agency. Therefore, we decided to switch the therapeutic strategy. However, she experienced five additional episodes of relapse during the last 1.5 years, despite sufficient immunosuppression with oral steroids (prednisone, 10–60 mg per day), mycophenolate mofetil (1000 mg, twice daily), and tacrolimus (1.5 mg, twice daily) (Figure 2). After the last relapse, she was severely disabled (bilateral optic atrophy, EDSS=7.5) and developed immunosuppressant-induced

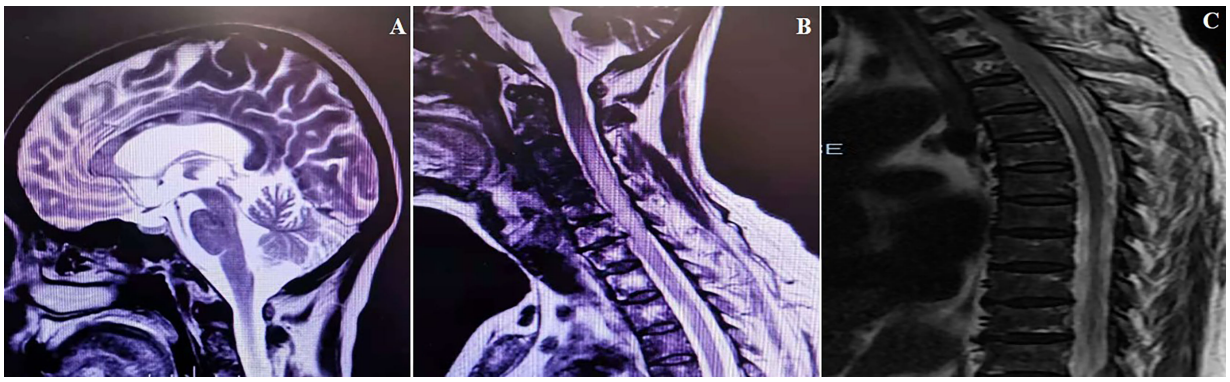


FIGURE 1
MRI scans obtained at the time of the first attack. **(A)** Cerebral MRI (sagittal T2 image) exhibiting disseminated lesions in corpus callosum. **(B, C)** Spinal MRI (sagittal T2 image) exhibiting longitudinally extensive lesion from C3 to T5.

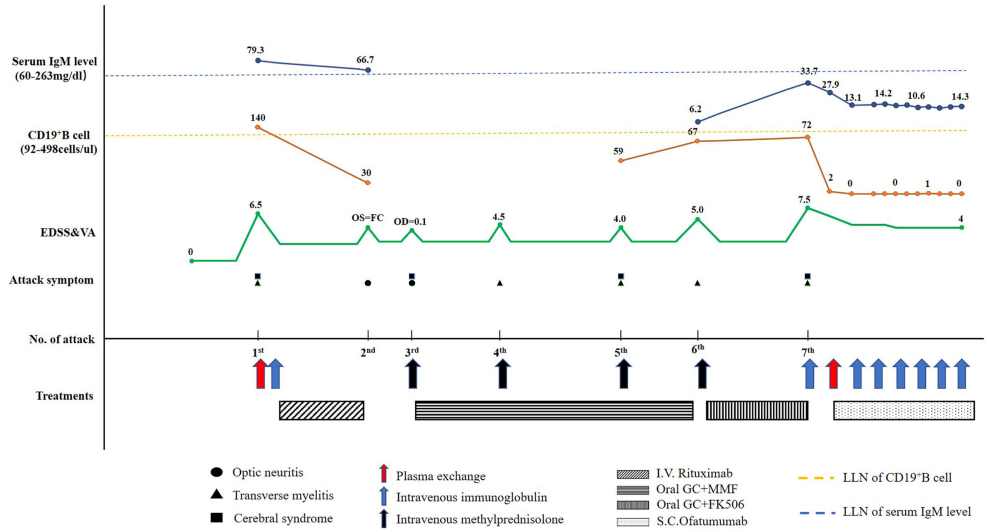


FIGURE 2
Schematic illustration of the disease course showing the temporal sequence of symptoms, disability, treatment details, $CD19^+$B cell count, and serum IgM level. EDSS, Expanded Disability Status Scale; FC, finger count; FK506, tacrolimus; GC, glucocorticoids; I.V., intravenous; S.C., subcutaneous; MMF, mycophenolate mofetil; OD, right visual acuity; OS, left visual acuity; LLN, lower limits of normal; VA, visual acuity.

hypogammaglobulinemia (with greatest impact on IgM). In consideration of the first-line treatment resistance and secondary immunodeficiency, subcutaneous ofatumumab was prescribed: once weekly injection of 20 mg for 3 weeks and then one injection of 20 mg every 4 weeks, in combination with IVIg (2 g/kg each month for the first 3 months followed by 1.2 g/kg each month) replacement therapy. Six months later, she was able to walk independently (EDSS=4.0), and experienced no further relapse. The peripheral $CD19^+$B cell count decreased to 2 cells/μL (reference range, 92–498 cells/μL) after the first dose and was maintained at low-level during ofatumumab treatment. Serum IgM level remained stable (Figure 2).

Discussion

As the first-generation anti-CD20 antibody, RTX depletes B cells mainly *via* antibody-dependent cell-mediated cytotoxicity pathways due to the linking of fragment c gamma receptors (FcγR IIIA) on natural killer cells (3). Consequently, patients with *FCGR3A-V158F* genetic mutation may have greater probability of insufficient depletion of B cells, and relapse during RTX treatment (4). Ofatumumab exhibits a greater potency in recruiting complement than RTX, thus exerting a higher complement-dependent cytotoxic efficacy (5). Thus, the low FcγR IIIA pathway dependent property of ofatumumab may explain its better efficacy than RTX in the present case, even in the presence of homozygous mutation of *FCGR3A-V158F* genotype.

In addition, hypogammaglobulinemia may occur during prolonged treatment with anti-CD20-depleting therapies and may lead to serious infections. In randomized controlled trials, a small percentage of ofatumumab-treated patients developed decreased immunoglobulin levels (6, 7). Our patient developed drug-induced hypogammaglobulinemia, but still showed high disease activity. Therefore, subcutaneous ofatumumab plus IVIg replacement therapy were introduced. Fortunately, ofatumumab was well-tolerated by our patient. Besides, ofatumumab decreased the probability of relapse with sufficient depletion of B cells.

In summary, the current case suggests that ofatumumab might be an effective alternative in patients with incomplete B cell depletion after RTX genetic testing in NMOSD, and highlights the potential safety of ofatumumab in elderly patients.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding authors.

Ethics statement

The studies involving human participants were reviewed and approved by Institutional Review Board of China-Japan

Friendship Hospital. The patients/participants provided their written informed consent to participate in this study.

Author contributions

WHZ drafted the manuscript. WHZ, YJJ, JSJ, MJ, and DTP prepared the materials, collected, and analyzed the data. WHZ and DTP revised the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This work was sponsored by the National Natural Science Foundation of China (Grant No. 82174440).

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Nakahara K, Nakane S, Nagaishi A, Narita T, Matsuo H, Ando Y. Very late onset neuromyelitis optica spectrum disorders. *Eur J Neurol* (2021) 28:2574–81. doi: 10.1111/ene.14901

2. Maillart E, Renaldo F, Papeix C, Deiva K, Bonheur J, Kwonet T, et al. Dramatic efficacy of ofatumumab in refractory pediatric-onset AQP4-IgG neuromyelitis optica spectrum disorder. *Neurol Neuroimmunol Neuroinflamm* (2020) 7:e683. doi: 10.1212/NXI.0000000000000683

3. Cornec D, Tempescul A, Querellou S, Hutin P, Pers JO, Jamin C, et al. Identification of patients with indolent b cell lymphoma sensitive to rituximab monotherapy. *Ann Hematol* (2012) 91:715–21. doi: 10.1007/s00277-011-1369-y

4. Kim SH, Jeong IH, Hyun JW, Joung A, Jo HJ, Hwang SH, et al. Treatment outcomes with rituximab in 100 patients with neuromyelitis optica: Influence of FCGR3A polymorphisms on the therapeutic response to rituximab. *JAMA Neurol* (2015) 72:989–95. doi: 10.1001/jamaneurol.2015.1276

5. Teeling J, French R, Cragg M, van den Brakel J, Pluyter M, Huang H, et al. Characterization of new human CD20 monoclonal antibodies with potent cytolytic activity against non-Hodgkin lymphomas. *Blood* (2004) 104:1793–800. doi: 10.1182/blood-2004-01-0039

6. Bar-Or A, Grove RA, Austin DJ, Brakel J, Pluyter M, Huang H, et al. Subcutaneous ofatumumab in patients with relapsing remitting multiple sclerosis: the MIRROR study. *Neurology* (2018) 90:e1805–14. doi: 10.1212/WNL.0000000000005516

7. Hauser SL, Bar-Or A, Cohen JA, Comi G, Correale J, Coyle PK, et al. Ofatumumab versus teriflunomide in multiple sclerosis. *N Engl J Med* (2020) 383:546–57. doi: 10.1056/NEJMoa1917246

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 17 April 2023
DOI 10.3389/fneur.2023.1142160

OPEN ACCESS

EDITED BY
Mario U. Manto,
University of Mons, Belgium

REVIEWED BY
David R. Benavides,
University of Maryland, United States
Amanda Piquet,
University of Colorado Anschutz Medical Campus, United States

*CORRESPONDENCE
Saeed Alghamdi
✉ neurosaeed@yahoo.com

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Neurology

RECEIVED 11 January 2023
ACCEPTED 29 March 2023
PUBLISHED 17 April 2023

CITATION
Khojah O, Makkawi S and Alghamdi S (2023) Anti-mGluR1 encephalitis: Case illustration and systematic review.
Front. Neurol. 14:1142160.
doi: 10.3389/fneur.2023.1142160

Anti-mGluR1 encephalitis: Case illustration and systematic review

Osama Khojah[1,2,3], Seraj Makkawi[1,2,3] and Saeed Alghamdi[4]*

[1]College of Medicine, King Saud Bin Abdulaziz University for Health Sciences, Jeddah, Saudi Arabia, [2]King Abdullah International Medical Research Center, Jeddah, Saudi Arabia, [3]Department of Medicine, Ministry of the National Guard-Health Affairs, Jeddah, Saudi Arabia, [4]Neuroscience Department, King Faisal Specialist Hospital and Research Center, Jeddah, Saudi Arabia

Background: The literature for immune-mediated neurological disorders is evolving like no other field of neurological illnesses. Many new antibodies or disorders have been described in the last decade. The cerebellum is a brain structure susceptible to these immune-mediated pathologies, and anti-metabotropic glutamate receptor 1 (mGluR1) antibody has a predilection to the cerebellar tissue. Anti-mGluR1 encephalitis is a rare autoimmune disease affecting the central and peripheral nervous systems, triggering an acute or subacute cerebellar syndrome with varying degrees of severity. Anti-mGluR1 encephalitis is a rare autoimmune disease affecting the central nervous system. We aimed to systematically review reported cases of anti-mGluR1 encephalitis and summarize their clinical presentation, management, outcomes, and case reports.

Methods: A search of the PubMed and Google Scholar databases was conducted and included all cases of anti-mGluR1 encephalitis published in English before October 1, 2022. A comprehensive systematic review was conducted using "metabotropic glutamate receptor type 1," "mGluR1," autoantibodies," "autoantibodies," "autoimmunity," and "antibody" as keywords. The risk of bias assessment of the evidence was performed using appropriate tools. The qualitative variables were presented as frequency and percentage.

Results: Including our case, 36 cases of anti-mGluR1 encephalitis (19 males, median age 52.5years, 11.1% pediatric cases) have been reported. The most common clinical manifestations are ataxia, dysarthria, and nystagmus. Initial imaging was normal in 44.4% of patients; however, 75% of patients showed abnormality later in the disease course. The first-line therapy options include glucocorticoids, intravenous immunoglobulin, and plasma exchange. Rituximab is the most commonly used second-line treatment. Complete remission was achieved in only 22.2% of patients, and 61.8% were disabled by the end of their course.

Conclusion: Anti-mGluR1 encephalitis manifests as symptoms of cerebellar pathology. Although the natural history has not been completely elucidated, early diagnosis with prompt initiation of immunotherapy could be imperative. Any patient suspected to have autoimmune cerebellitis should be tested for the presence of anti-mGluR1 antibody in the serum and cerebrospinal fluid. Escalation to an aggressive therapy approach should be applied in cases that do not respond to first-line therapies, and extended follow-up durations are required in all cases.

KEYWORDS

autoimmune, cerebellar ataxia, metabotropic glutamate receptor 1, mGluR1, antibodies, metabotropic glutamate receptor 1 (mGluR1) antibodies

1. Introduction

Metabotropic glutamate receptors (mGluR) are pre- and postsynaptic receptors found in the central and peripheral nervous systems and extensively expressed in Purkinje cells. These receptors are involved in cerebellar development, synaptic transmission, modulation, plasticity, pain perception, memory, learning, and anxiety (1). In the cerebellum, these G-protein coupled receptors are mainly located postsynaptically. mGluR1 is not only expressed at the dendrites of the Purkinje cells but also in parallel fibers and climbing fiber inputs (2). These receptors are essential for cerebellar motor learning, as activating mGluR1 leads to long-term depression of Purkinje cell-parallel fiber synapses (3). Rarely, mGluR1 is targeted by autoantibodies that cause a subacute form of cerebellitis or encephalitis (2). This antineuronal autoimmune reaction was hypothesized to be paraneoplastic in nature as it was associated with malignancies like lymphomas. However, the majority of cases were not associated with any tumors (4). Detection of the antibodies in the cerebrospinal fluid (CSF) or serum and the presence of clinical symptoms are diagnostic of the disease. Stepwise escalation with immunotherapeutic agents, including high-dose intravenous glucocorticoids, intravenous immunoglobulins (IVIg), and/or plasma exchange (PLEX), is used as a first-line treatment for the disease (5). Early initiation of immunotherapy yields better results and prognosis. If the case is severe or not clinically improving, rituximab, cyclophosphamide, azathioprine, or mycophenolate mofetil is used as second-line therapy (6). In this case illustration and systematic literature review, we report the clinical features, 5-year treatment course, and outcomes of a patient with anti-mGluR1 encephalitis. We also describe the disease course, diagnostic test findings, patient outcomes, and treatment approaches for anti-mGluR1 encephalitis outlined in the literature.

2. Materials and methods

2.1. Literature review

2.1.1. Search methods

We performed a comprehensive systematic review by searching the PubMed and Google Scholar databases. We used "metabotropic glutamate receptor type 1," "mGluR1," autoantibodies," "autoantibody," "autoimmunity," and "antibody" keywords in combination with Boolean operators to ensure the inclusivity of all possible results. The search included all reports published until October 1st, 2022. The study followed the preferred reporting items for systematic reviews and meta-analyses (PRISMA) guidelines.

2.1.2. Inclusion and exclusion criteria

All published studies that reported at least one anti-mGluR1 encephalitis patient were included. Only studies published in or translated into English were included. The diagnosis of anti-mGluR1 encephalitis had to be based on clinical findings and the presence of mGluR1 antibodies in the serum or CSF. Patients who fulfilled the following criteria were excluded: (1) presence of anti-mGluR1 antibodies in the serum only; (2) low anti-mGluR1 serum titer; (3) positive antibody testing to another neurological autoimmune disease better explaining the patient symptoms. Patients excluded by these criteria included the one reported by Durovic et al. who described a patient diagnosed with anti-MOG encephalitis and was found to be anti-mGluR1 seropositive (titer of 1:40) (7).

2.1.3. Selection of studies

All authors independently assessed the eligibility of each article from the database search. The eligibility of the articles was determined by screening titles and abstracts and then reviewing the full-text versions of the articles. Titles and abstracts were screened by assessing the type of article and population targeted. For example, screened articles involving non-human subjects were excluded. Furthermore, titles and abstracts reporting at least one patient diagnosed with anti-mGluR1 encephalitis underwent further assessment by reviewing the full-text versions of the articles. All disagreements were resolved by consensus.

2.1.4. Data collection

The data collected from each eligible article included age, sex, presence of prodromal symptoms, associated malignancies, clinical manifestations on the first presentation and their duration, leukocyte count, presence of oligoclonal bands in the CSF, antibody titers or presence in the serum and CSF, brain magnetic resonance imaging (MRI) findings, management for acute presentation, maintenance therapy, remission, relapses, presence of antibodies post-therapy, duration of follow-up in months, and disability. Relapse was defined as the acute appearance of new neurological symptoms and/or the recurrence of old symptoms. The geographical origin of a case was determined based on the location of the center described in the methodology or the location of the authors' primary affiliation. Remission was subdivided into complete, partial, or no remission. Complete remission was defined as complete or near-complete resolution of all symptoms without associated disability, partial remission was defined as meaningful clinical improvement from the first presentation, and no remission was defined as no clinical improvement or worsening of the clinical status. Disability was defined as a limitation in the patient's physical ability which may or may not require using medical assistive devices such as a wheelchair on their last visit.

2.1.5. Risk of bias and quality assessment

All authors independently and critically appraised the methodological quality of the studies using a modified version of the framework by Murad et al., (8) which was developed to evaluate the risk of bias in case reports and case series. We adapted this tool to assess the reported patients with anti-mGluR1 encephalitis. Of the eight questions described in the assessment framework, five were deemed compatible with our design and adjusted to fit our population. Each question could be answered with a "Yes" or "No" after critically appraising each study. Studies were appraised based on the following: (1) whether the study was specifically conducted to assess patients with anti-mGluR1 encephalitis; (2) if the treatment for anti-mGluR1 encephalitis patients, such as IVIg, glucocorticoids, PLEX, rituximab, tacrolimus, azathioprine, mycophenolate mofetil, cyclophosphamide, and hydroxychloroquine, was adequately ascertained; (3) if the outcome of anti-mGluR1 encephalitis patients was adequately (clinically and radiologically) ascertained; (4) if anti-mGluR1 encephalitis patients were followed up for long enough to determine outcomes, such as relapses or disability, which was set at 12 months or longer; (5) if the study was described in sufficient detail for replication

by another investigator or to allow other investigators to make inferences.

A study was considered high quality if it scored "Yes" in more than three questions, moderate quality if it scored "Yes" in two or three questions, and low quality if it scored "Yes" in one or none of the questions. All disagreements were resolved by consensus (Supplementary Table 1).

2.2. Ethical considerations

This study was approved by the Institutional Review Board of King Abdullah International Medical Research Center. Written informed consent was obtained from the patient for the publication of any potentially identifiable images or data included in this article.

3. Results

3.1. Case illustration

A 56-year-old woman presented to a nearby community hospital with slurred speech, unsteady gait, and low-grade fever, which persisted for 2 days. She was admitted with a suspected central nervous system infection and started on antimicrobial therapy. Her medical history was remarkable for type 2 diabetes, hypertension, and osteoarthritis. Computed tomography (CT) findings of the brain were normal. Two days later, the patient was transferred to our hospital (King Abdulaziz Medical City, Jeddah, Saudi Arabia) because her condition did not improve. Upon further questioning, she complained of double vision, fatigue, and generalized body ache. Neurological examination revealed head tremors (titubation), skew deviation, saccadic pursuit with hypometric saccades in horizontal gaze in both directions, and gaze-evoked nystagmus. Hypotonia was noted in both upper and lower limbs. Sensory examination showed reduced sensation in the length-dependent lower extremity. Coordination was impaired with dysmetria in both the upper and lower limbs and severe truncal ataxia with an inability to walk without assistance. Cognitive examination results were normal. CSF analysis showed mild lymphocytic pleocytosis (6 leukocytes/μL) (Normal level: 0–5 leukocytes/μL), a slightly elevated red blood cell count (10 cells/μL) (Normal level: 0 cells /μL), high glucose (6.5 mmol/L) (Normal level: 2.5–4.4 mmol/L), and normal protein (0.42 mg/ml) (Normal level: 0.15–0.6 mg/ml) levels. We made a presumed diagnosis of post-infectious cerebellitis and started the patient on pulse intravenous methylprednisolone (IVMP) (1,000 mg/day) for 3 days, followed by IVIg (1,000 mg/kg) for 5 days. The patient reported mild improvement without functional recovery. CSF cytology and flow cytometry revealed no abnormalities. Serums and CSF samples were sent to Bioscientia International labs in Germany for extensive autoimmune, microbiological, and rheumatological markers analyses. It showed positive oligoclonal bands (OCB) in the CSF, which were absent in the serum, and normal angiotensin-converting enzyme levels in both the CSF and serum. Polymerase chain reaction detected no CSF herpes simplex virus (HSV) DNA types 1 and 2. Results of CSF autoantibody panel that included antibodies against Ca channel (P/Q type), Hu, Ri, Yo, collapsin response mediator protein 5 (CV5/CRMP5), AMPA-1 receptor, metabotropic glutamate receptor 5 (mGlluR5), and metabotropic glutamate receptor 1 (mGluR1) were all negative except for anti-mGluR1 antibodies. CSF and serum anti-mGluR1 antibodies were both detected through indirect immunofluorescence assays with titers of 1:32 and 1:1,000, respectively. Therefore, based on her clinical features and investigational findings, anti-mGluR1 encephalitis was diagnosed. The patient was readmitted for additional immune-modulating therapies and an expedited workup for occult malignancy. Brain MRI showed bilateral, almost symmetrical, subcortical high signal intensity, mostly in the occipital lobes, with no diffusion restriction and no cerebellar signal changes or atrophy. Chest, abdomen, and pelvis CT, mammography and whole-body positron emission tomography (PET)-CT did not show any lesions suspicious of malignancy. Another round of IVMP (1,000 mg/day) and IVIg (1,000 mg/kg) was administered, followed by rituximab (1,000 mg 2 weeks apart then then followed by maintenance of 1,000 mg every 6 months), daily azathioprine (100 mg) and oral steroids upon discharge. Three months later, she was able to walk short distances without assistance, and her slurred speech improved dramatically. After completing 3 doses of rituximab, she was able to function normally at baseline, with mild residual dysarthria and titubation. Two months later, the patient relapsed with a recurrence of disabling ataxia requiring the use of a wheelchair. She was admitted for PLEX and IVMP (1,000 mg/day). The results of repeated CSF analysis remained unchanged and brain MRI showed mild cerebellar atrophy (Figure 1B). After 3 months of biweekly PLEX, remission was achieved again, and the patient could walk unassisted. The patient was then kept on monthly IVIg (1,000 mg/kg). After almost 2 years of monthly IVIg treatment and slow deterioration of her condition, she could not walk without assistance and carry out her activities of daily living. The decision was made to stop IVIg and restart rituximab (1,000 mg) every 6 months. However, due to logistical issues created by the COVID-19 pandemic we were not confident that rituximab infusion will be provided on time, azathioprine (100 mg) was added for about 1 year; once these issues were resolved, we discontinued azathioprine. Six months later, the patient regained functional ability and was able to perform activities of daily living while relying on a walker. A follow-up brain MRI (Figure 1C) showed moderate cerebellar atrophy involving both cerebellar hemispheres and the vermis, with occipital T2 signal changes visualized on her first brain MRI completely resolved. Although repeated CSF analysis showed no signs of inflammation, nor was it positive for OCB, anti-mGluR1 antibodies continued to be present at the same titer. As of now—5 years after the initial presentation—she has mild dysarthria, bilateral dysmetria, and truncal ataxia; her mRS score is 1 and clinical assessment scale in autoimmune encephalitis (CASE) score is 3, repeated cancer screening is still negative, brain MRI is stable and she is on maintenance dose and rituximab (1,000 mg) every 6 months.

3.2. Systematic review

A PRISMA flow diagram describing the case selection process is shown in Figure 2. Fifteen articles (12 case reports and 3 case series) described 35 cases of anti-mGluR1 encephalitis in the literature. Overall, 36 patients were analyzed. The cases originated in the United States of America (9), Spain (10), France (4), Germany (4), Netherlands (3), Italy (2), Brazil (1), Japan (1), Saudi Arabia (1), and Singapore (1).

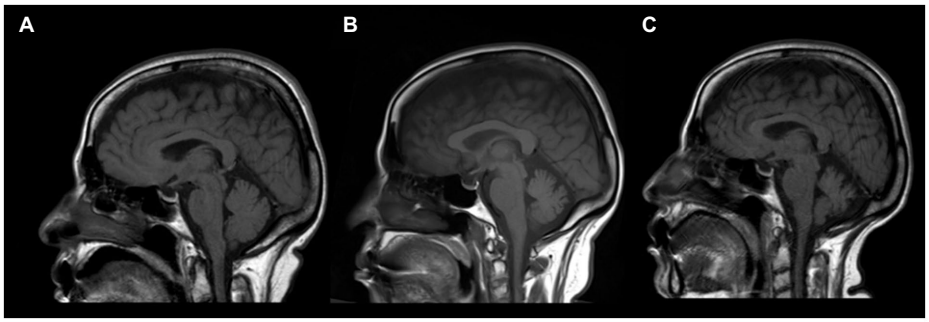


FIGURE 1
Sagittal brain MRI of an anti-mGluR1 encephalitis patient showing T1 sequence of **(A)** the cerebellar hemisphere upon initial presentation, which later showed progressive cerebellar atrophy at 10 months **(B)** and 20 months **(C)** of follow-up.

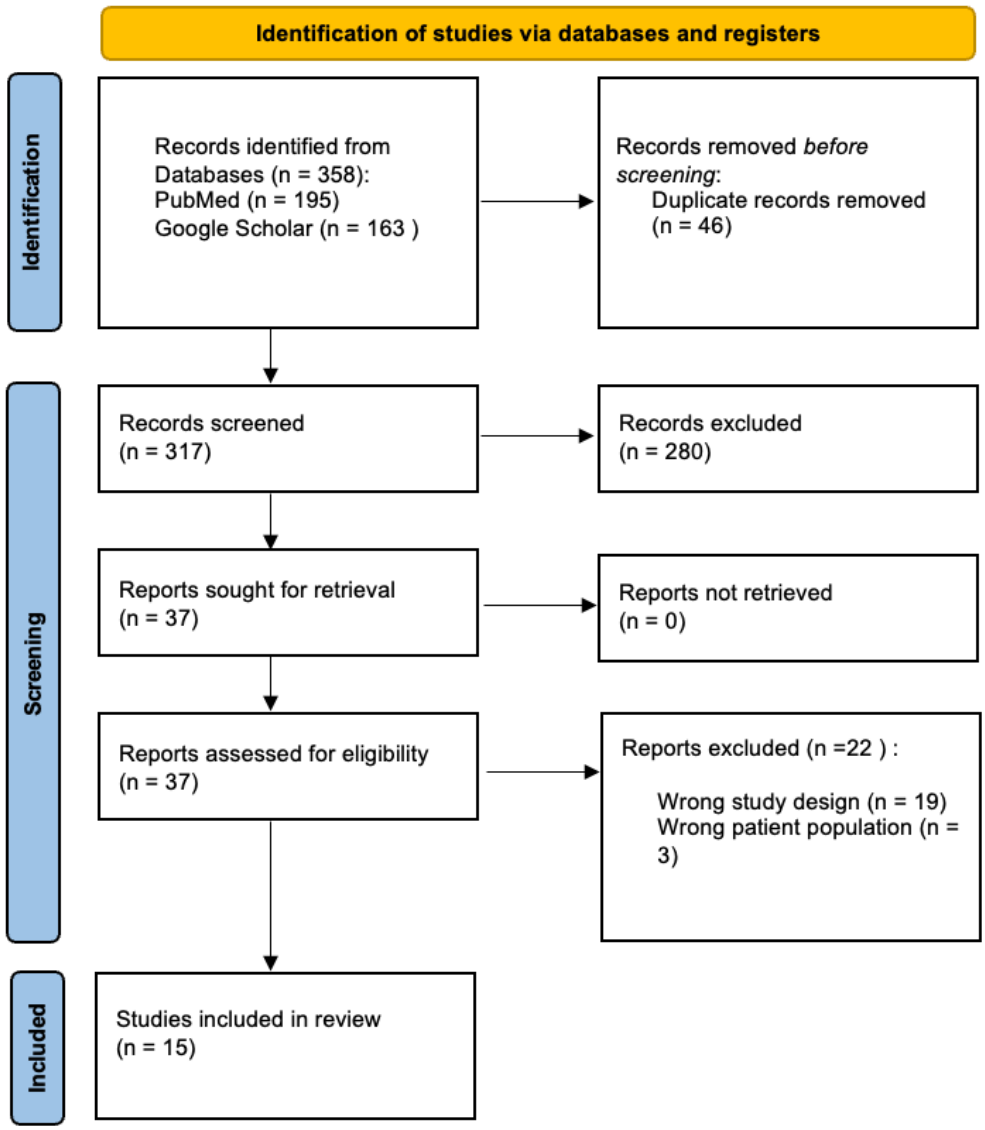


FIGURE 2
PRISMA 2020 flow diagram for systematic reviews which included searches of databases.

3.3. Literature review

3.3.1. Demographic data and clinical presentations

Overall, 35 cases of anti-mGluR1 encephalitis have been reported in the literature (2, 4, 9–21). Anti-mGluR1 encephalitis affected both sexes equally (1.12:1 M:F). The median age at presentation was 52.5 years (range: 3–81 years). Patients younger than 18 years of age represented 11.1% of all patients and were mostly male (3:1 M:F); 22.2% of all patients had an associated malignancy, six of whom had a lymphoma. Fifty percent of patients with malignancy had it within 5 years of the autoimmune cerebellitis or encephalitis onset; 16.7% (n = 6/36) of patients were diagnosed with an autoimmune disease other than that involving mGluR1 antibodies. These autoimmune diseases included multiple sclerosis, Hashimoto's thyroiditis, Sjögren's syndrome, and pernicious anemia. Twenty-five percent of patients had one or more prodromal symptoms, including fever, headache, fatigue, weight loss, nausea, vomiting, night sweats, and/or flu-like symptoms; 83.3% of patients had one or more cerebellar symptoms on the first presentation, and later in the disease course, almost all of the patients (94.4%) manifested one or more cerebellar symptoms. Table 1 summarizes the demographic data and clinical features of the previously reported cases. Table 2 describes the symptoms and their proportions during the disease course.

3.3.2. Investigations

Reported CSF analysis revealed elevated leukocyte counts in 51.7% and OCBs in 47.8% of patients. Anti-mGluR1 antibodies were detected in the serum of 97.1% and CSF of 96.3% of patients. These antibodies reportedly persisted in 77.8% of patients who were tested in either the CSF or the serum. The presence of anti-mGluR1 antibodies was found in the serum and CSF of 61.1% of patients. In one of patients, it was positive in the serum rather than in the CSF; and positive in two of patients in the CSF but not in the serum. Assessment of the presence of these antibodies in both the serum and CSF was not performed in 30.56% of patients. Initial imaging was normal in 44.4% of patients, but follow-up imaging showed one more finding in 75% of patients. Brain MRI findings included cerebral atrophy, enhancing and non-enhancing brain and spinal cord lesions, as well as cerebellar findings, including cerebellar hyperintensity, enhancement of cerebellar leptomeninges, atrophy, or edema. These cerebellar findings were observed in 52.7% of patients and tended to occur in the medial cerebellar hemispheres and the vermis. Imaging results on the first presentation revealed that 14.7% of patients had generalized or focal brain atrophy, and 41.1% of patients had brain atrophy on follow-up MRIs. Table 3 summarizes the investigative findings of patients reported in the literature.

3.3.3. Management and outcomes

First-line therapy options include glucocorticoids, IVIg, and PLEX. One or more of the aforementioned treatments were used in 83.3% of patients. Patients were administered glucocorticoids (66.7%), IVIg (38.9%), and PLEX (13.9%). Second-line therapy was used in 41.7% of patients as follows: rituximab (27.8%), azathioprine and cyclophosphamide (13.9%), mycophenolate mofetil (11.1%), and tacrolimus and hydroxychloroquine (2.8%); 93.3% of patients who received second-line therapy, failed to have complete remission. More than three treatment modalities were used in 36.1% of cases, yet only 15.4% of whom were able to achieve complete remission. Patients had complete, partial, and no remission in 22.2, 55.6, and 19.4% of cases, respectively. Eventually, 61.8% of patients ended up with some dependency or disability, of whom 57.1% required walking aid, and 9.5% required wheelchair support. Further, 22.2% of patients had one or more relapses—all of whom experienced a disability. Most relapsing patients experienced one relapse, though approximately three episodes have been reported. The median follow-up duration was 24 months. All treatment modalities of the reported cases are summarized in Table 4.

4. Discussion

Our illustrated case was an adult patient presented with subacute cerebellar syndrome, diagnosed with anti-mGluR1 encephalitis, and requiring multiple treatment modalities. Our systematic review demonstrated that most patients present with symptoms of cerebellar pathology. Hence, it is imperative to consider anti-mGluR1 encephalitis as a part of the differential diagnosis in any patient suspected to have autoimmune cerebellitis. Brain imaging might be normal in approximately half of the patients. Many patients require multiple treatment options and regimens. However, a minority of patients return to their baseline. In 2000, Smitt et al. published the first two cases of anti-mGluR1 encephalitis in which both patients developed cerebellar ataxia (11). Both patients had Hodgkin's lymphoma, which had been in remission for multiple years. A history of malignancy, in addition to normal brain MRI findings, prompted the authors to analyze serum and CSF samples for the presence of antineuronal antibodies. After injecting these samples into mice, they were able to elicit an immunohistochemical staining pattern in the brains of the mice that had a distribution pattern similar to that of mGluR1. Evidence for the pathogenicity of anti-mGluR1 antibodies was demonstrated when IgG was injected into mice. After less than an hour, the mice began to show symptoms of cerebellar pathology (11). Novel mutations in the GRM1 gene, which encodes for mGluR1, also reportedly caused progressive forms of cerebellar ataxia in five affected families in Italy (22). Tumor tissue samples were obtained from the patients in Smitt et al. reports; however, none of them expressed mGluR1 or a cross-reactive epitope (11). Contrastingly, the fifth reported case was a patient in remission from mycosis fungoides who presented with ataxia and dysarthria. Eighteen months later, the patient was diagnosed with prostate adenocarcinoma, which after further testing, showed rich expression of mGluR1 and reactivity with anti-mGluR antibodies (20). Of the patients reported in the literature, 77.8% did not have an associated malignancy. Hence, it is still unclear whether malignancies play a role in the development of anti-mGluR1 encephalitis, though continuous testing is still of utmost importance (2). First-line screening for malignancies is CT of the chest, abdomen, and pelvis, although negative results prompt further investigations. PET scans play a substantial role in ruling out occult malignancy. The European Federation of Neurological Societies recommends following up a negative CT with fluorodeoxyglucose-PET in cases with a high index of suspicion for paraneoplastic syndrome (23–25). The trigger of this autoimmune reaction in a non-paraneoplastic form is yet to be completely understood; however, it has been noticed that prodromal symptoms, which echo a viral infection, like in our patient, might trigger this reaction (1). Vague symptoms occurring before the

TABLE 1 Demographic features and clinical manifestations on first presentation of 36 patients diagnosed with anti-mGluR1 encephalitis.

Case number	References	Age/gender	Prodromal Symptoms	Associated malignancy	Clinical manifestations on first presentation
1	Sillevis Smitt et al. (11)	19/F	None	Hodgkin's lymphoma	Cerebellar syndrome
2	Sillevis Smitt et al. (11)	49/F	None	Hodgkin's lymphoma	Cerebellar syndrome and cognitive decline
3	Marignier et al. (10)	50/F	Yes	None	Cerebellar syndrome and headache
4	Lancaster et al. (9)	69/M	None	None	Cerebellar syndrome
5	Iorio et al. (20)	65/M	None	Mycosis fungoides and prostate adenocarcinoma	Cerebellar syndrome and cognitive decline
6	Lopez-Chiriboga et al. (4)	64/M	None	None	Cerebellar ataxia
7	Lopez-Chiriboga et al. (4)	54/M	None	None	Cerebellar ataxia
8	Lopez-Chiriboga et al. (4)	81/M	None	None	Cerebellar ataxia and cognitive impairment
9	Lopez-Chiriboga et al. (4)	77/M	None	None	Cerebellar ataxia
10	Lopez-Chiriboga et al. (4)	51/M	None	Testicular seminoma	Psychiatric symptoms and dysgeusia
11	Lopez-Chiriboga et al. (4)	60/F	None	None	Dysgeusia
12	Lopez-Chiriboga et al. (4)	58/F	None	None	Cerebellar syndrome and dysgeusia
13	Lopez-Chiriboga et al. (4)	67/M	None	Cutaneous T-cell lymphoma	Cerebellar ataxia
14	Lopez-Chiriboga et al. (4)	67/F	None	None	Paresthesia, vertigo, and dysgeusia
15	Lopez-Chiriboga et al. (4)	33/F	None	Acute lymphocytic leukemia	Cerebellar syndrome and cognitive impairment
16	Lopez-Chiriboga et al. (4)	77/F	None	Mantle cell non-Hodgkin's lymphoma	Ataxia, spastic paresis, and cognitive impairment
17	Yoshikura et al. (2)	61/F	None	None	Cerebellar syndrome and dysphagia
18	Pedroso et al. (21)	39/F	None	None	Behavioral changes, catatonia, and cerebellar syndrome
19	Christ et al. (12)	45/M	None	None	Dysarthria
20	Gollion et al. (13)	64/M	None	None	Cerebellar ataxia and myoclonic jerks
21	Chaumont et al. (14)	22/F	Yes	None	Cough, headache, and cerebellar syndrome
22	Spatola et al. (15)	29/M	Yes	None	Sleeping difficulties
23	Spatola et al. (15)	22/F	Yes	None	Cerebellar syndrome, fever, hallucinations, and cognitive decline
24	Spatola et al. (15)	45/F	Yes	None	Slowness in writing, hypophonia, and cerebellar syndrome
25	Spatola et al. (15)	59/M	None	None	Cerebellar syndrome
26	Spatola et al. (15)	54/M	Yes	None	Visual loss, cerebellar syndrome, behavioral changes
27	Spatola et al. (15)	56/M	None	None	Behavioral changes and cognitive decline
28	Spatola et al. (15)	62/M	None	Sarcoma	Gait instability
29	Spatola et al. (15)	24/M	None	Hodgkin's lymphoma	Cerebellar syndrome
30	Spatola et al. (15)	49/F	None	None	Focal seizures and impaired level of consciousness
31	Spatola et al. (15)	6/M	Yes	None	Cerebellar syndrome, tremor and choreiform movements.
32	Bien et al. (16)	3/M	None	None	Unsteady gait
33	Chandler et al. (17)	5/F	Yes	None	Nausea, vomiting, abdominal pain, fever, headache and altered mental status and cerebellar syndrome
34	Vinke et al. (18)	50/F	None	None	Seizures and hallucinations
35	Goh et al. (19)	15/M	None	None	Cerebellar syndrome
36	Current case	56/F	Yes	None	Febrile illness followed by cerebellar syndrome

M, male; F, female.

TABLE 2 Summary of anti-mGluR1 encephalitis symptoms and their proportions.

Symptom	Number of patients	%
Cerebellar symptoms	34	94.44
Ataxia	31	86.11
Dysarthria	19	52.78
Nystagmus	10	27.78
Titubation	7	19.44
Dysmetria	7	19.44
Vertigo	6	16.67
Diplopia	4	11.11
Intention tremor	4	11.11
Oscillopsia	2	5.56
Extra-cerebellar neurological symptoms	16	44.4
Dysgeusia	4	11.11
Motor changes	3	8.33
Seizures	3	8.33
Myoclonic jerks	2	5.56
Sensory changes	2	5.56
Choreiform movement	1	2.78
Dysphagia	1	2.78
Dystonia	1	2.78
Hypophonia	1	2.78
Loss of vision	1	2.78
Slowness in writing	1	2.78
Behavioral symptoms	10	29.41
Apathy	6	16.67
Hallucinations	4	11.11
Catatonia	3	8.33
Personality changes	3	8.33
Irritability	2	5.56
Depression	1	2.78
Impulsivity	1	2.78
Loss of initiative	1	2.78
Paranoia	1	2.78
Cognitive symptoms	10	29.41
Cognitive impairment	7	19.44
Memory loss	6	16.67
General symptoms	9	25
Fever	4	11.11
Headache	4	11.11
Fatigue	2	5.56
Weight loss	2	5.56
Nigh sweat	1	2.78
Sleep difficulties	1	2.78

onset of neurological symptoms were reported in 25% of patients. The cause of these prodromal symptoms is unknown. However, one case was preceded by herpes zoster infection in the trigeminal nerve a month before the disease onset, another was preceded by streptococcal pharyngitis 2 months prior, and yet another case was found to have evidence of dengue virus infection. These findings suggest a post-infectious element in the occurrence of anti-mGluR1 encephalitis or that infection may trigger its onset (4, 14, 15). Unlike most autoimmune disorders which favor women, coincident autoimmunity in anti-mGluR1 encephalitis patients affected both genders equally (26). The diagnosis of anti-mGluR1 encephalitis has been based on the presence of neurological symptoms that tend to affect the cerebellum and anti-mGluR1 antibodies in the CSF or serum. However, a threshold for antibody titers to make a diagnosis is yet to be established. The presence of anti-mGluR1 antibodies was found in the serum and CSF in more than half of the patients. The presence of these antibodies in the serum alone was not sufficient to diagnose the patient with anti-mGluR1 encephalitis, as shown by Durovic et al. (7). Relying on the presence of anti-mGluR1 antibodies in the serum alone—of asymptomatic patients or patients diagnosed with other neurological autoimmune diseases—complicate the process of diagnosis (7). Durovic et al. reported a case of MOG encephalitis with anti-mGluR1 antibodies in the serum but not in the CSF (7). They deemed a titer of 1:40 to be too low to be clinically relevant, and the lack of typical cerebellar signs and symptoms made the diagnosis of anti-mGluR1 encephalitis unlikely (7). However, titers lower than those reported by Durovic et al. have been described in a case where the patient had a serum titer of 1:20, although that diagnosis was supported by a CSF titer of 1:8 (16). Lopez et al. described a patient who presented with cognitive and cerebellar symptoms and fulfilled the diagnostic criteria for multiple sclerosis, yet had anti-mGluR1 antibodies in the serum but not in the CSF (4). In both cases, it is difficult to be certain whether these pathologies were particularly due to anti-mGluR1 encephalitis or whether the detection of anti-mGluR1 antibodies was an incidental finding in the context of another autoimmune disease (4, 7). The appearance of anti-mGluR1 encephalitis associated with other autoimmune diseases was reported in 16.7% of cases. Smitt et al. found that the anti-mGluR1 titer per unit of IgG was significantly higher in the CSF than in the serum (11). Moreover, Vinke et al. reported a patient with antibodies detected in the CSF but not in the serum. Further, our patient showed positive OCBs in the CSF but not in the serum, providing evidence of intrathecal synthesis of these antibodies (18). Detection of antibodies in the CSF might be more sensitive than in the serum; this has been illustrated in other antibody-mediated autoimmune encephalitides, such as anti-N-methyl-D-aspartate receptor (NMDAR) encephalitis (27). Thus, studies aiming to assess the sensitivity and specificity of serum and CSF antibody testing in patients with anti-mGluR1 encephalitis are necessary. Lack of cerebellar signs and symptoms is rare but insufficient to exclude the diagnosis. Two cases have been described where the patients never developed any cerebellar signs or symptoms (15). mGluR1 is highly expressed in the cerebellum but is also expressed in the limbic system (hippocampus and olfactory bulb), basal ganglia (globus pallidus, ventral pallidum, and substantia nigra), thalamus, lateral septum, superior colliculus, and parts of the posterior region of the tongue (4, 28). Brain MRI findings and symptoms correlating with each of these structures, such as seizures and

TABLE 3 Summary of the investigative findings of 36 patients diagnosed with anti-mGluR1 encephalitis.

Case number	References	CSF features		Antibodies titer in serum	Antibodies titer in CSF	Brain MRI
		Leukocytes/μL	*Oligoclonal Bands*			
1	Sillevis Smitt et al. (11)	28	Negative	1:3200	1:512	Normal
2	Sillevis Smitt et al. (11)	NA	NA	1:3200	Positive	Normal
3	Marignier et al. (10)	190	Negative	1:20000	1:500	Diffuse cerebellar hyperintensity Follow-up showed moderate cerebellar atrophy
4	Lancaster et al. (9)	8	NA	Positive	Positive	Initially normal Follow-up showed cerebellar atrophy
5	Iorio et al. (20)	Normal	NA	Positive	Positive	Mild cerebellar atrophy
6	Lopez-Chiriboga et al. (4)	Normal	Negative	1:960	NA	Normal
7	Lopez-Chiriboga et al. (4)	NA	NA	1:1920	1:256	NA
8	Lopez-Chiriboga et al. (4)	Normal	Negative	1:1920	1:64	Mild global atrophy and hyperintensity in the central superior cerebellum
9	Lopez-Chiriboga et al. (4)	Normal	Negative	1:61440	NA	Cerebral atrophy
10	Lopez-Chiriboga et al. (4)	29	NA	1:7680	NA	Normal
11	Lopez-Chiriboga et al. (4)	Normal	Negative	1:3840	NA	Normal
12	Lopez-Chiriboga et al. (4)	NA	NA	1:480	NA	NA
13	Lopez-Chiriboga et al. (4)	NA	NA	1:1920	NA	NA
14	Lopez-Chiriboga et al. (4)	NA	NA	1:960	NA	Mild cerebral and cerebellar atrophy
15	Lopez-Chiriboga et al. (4)	NA	Positive	1:1000	Negative	Multiple enhancing brains and spinal cord T2 lesions
16	Lopez-Chiriboga et al. (4)	NA	Positive	1:3200	NA	Multiple non-enhancing brains and spinal cord T2 lesions
17	Yoshikura et al. (2)	5	NA	1:3200	Positive	Initially normal Follow-up showed cerebellar atrophy
18	Pedroso et al. (21)	2	NA	1:12	1:512	Initially normal Follow-up showed cerebellar vermal atrophy
19	Christ et al. (12)	7	Negative	1:100	1:32	Hyperintensity in the medial thalamus and pulvinar predominantly on the left and low cerebellar volume
20	Gollion et al. (13)	Normal	Positive	NA	Positive	Normal
21	Chaumont et al. (14)	214	Positive	Positive	Positive	Cerebellar leptomeningeal contrast enhancement
22	Spatola et al. (15)	17	NA	Positive	Positive	Initially normal Follow-up showed cerebellar atrophy
23	Spatola et al. (15)	214	Positive	Positive	Positive	Gd enhancement of cerebellar leptomeninges
24	Spatola et al. (15)	3	Negative	Positive	Positive	Initially normal Follow-up showed cerebellar vermal atrophy
25	Spatola et al. (15)	2	Positive	Positive	Positive	Cerebellar and brain atrophy
26	Spatola et al. (15)	<5	Negative	Positive	Positive	Unspecific Subcortical dot-like lesions; Follow-up showed cerebellar atrophy
27	Spatola et al. (15)	9	Negative	Positive	Positive	Unspecific Subcortical dot-like lesions; Follow-up showed cerebellar atrophy
28	Spatola et al. (15)	27	NA	Positive	NA	Old ischemic lesions
29	Spatola et al. (15)	4	NA	Positive	Positive	Normal

(Continued)

TABLE 3 (Continued)

Case number	References	CSF features		Antibodies titer in serum	Antibodies titer in CSF	Brain MRI
		Leukocytes/μL	*Oligoclonal Bands*			
30	Spatola et al. (15)	<5	Positive	Positive	Positive	Hyperintensities in cerebellar vermis and right frontal lobe
31	Spatola et al. (15)	125	Positive	Negative	Positive	Initially normal Follow-up showed bi-hemispheric cerebellar edema
32	Bien et al. (16)	28	Positive	1:20	1:8	Normal
33	Chandler et al. (17)	39	Positive	NA	1:64	Hyperintensity in cerebellar vermis and medial cerebellar hemispheres
34	Vinke et al. (18)	6–10	Positive	Negative	Positive	Vascular damage
35	Goh et al. (19)	Normal	Normal	Positive	Positive	Normal
36	Current case	6	Positive	1:1000	1:32	Initially normal; Follow-up showed cerebellar atrophy

CSF, cerebrospinal fluid; MRI, magnetic resonance imaging; NA, not applicable; Gd, gadolinium

psychiatric and cognitive impairment in association with the limbic system, have been reported (18). It is possible for patients who initially present without cerebellar findings to develop them later. Six cases have been reported to first have presented without any cerebellar signs or symptoms, only to develop them later. Additionally, patients presenting with cerebellar signs and symptoms tend to develop other non-cerebellar neurological symptoms later (4). Pediatric patients seem to have a family history of autoimmune diseases, acute symptoms, and symptoms akin to those of movement disorders and cerebellar pathologies (15, 17, 29).

Electroencephalography (EEG) was not utilized in most cases; however, Christ et al. recommended its use for diagnostic purposes, especially when imaging and CSF cell counts were normal (12). CSF cell counts and initial MRI findings were normal in approximately half of the patients. However, over time, brain MRI findings were positive in three-quarters of the patients. These changes in the MRI findings from normal to abnormal are due to Purkinje cell degeneration after continuous exposure to antibodies, which should emphasize the importance of early treatment (2). Treatment of anti-mGluR1 encephalitis relies on immunosuppression, similar to other autoimmune encephalitides. Among patients who received any form of treatment, all but one received one or a combination of glucocorticoids, IVIg, and PLEX. Failure of first-line therapy necessitates the utilization of one or more second-line therapies. Multiple treatment options were used due to ineffective therapy, utilizing another form of therapy during relapse, and/or intolerable side effects. Spatola et al. were unable to find any significant correlation between good outcomes (mRS $\leq$ 2) and immunotherapy (15). A multicenter study including 577 patients diagnosed with anti-NMDAR encephalitis showed that only 27% of patients required second-line therapy (rituximab and/or cyclophosphamide) (24). Contrastingly, almost all patients with anti-mGluR1 encephalitis who received second-line therapy failed to achieve complete remission. Relapses tended to occur shortly after discontinuation of therapy. However, this was not always the case; our patient relapsed 2 months after completing the third dose of rituximab. Fortunately, relapses responded well to the resumption of therapy. Similarly, Christ et al. found that their patient's dysarthria worsened while the patient was on IVIg (12). Both the case reported by Christ et al. and our patient were started on rituximab therapy after their functional status continued to deteriorate. This decision yielded a dramatic improvement in both patients. Persistence or resolution of these antibodies does not seem to affect outcomes or disability. Hence, the treatment response should follow clinical symptoms rather than antibody titers in the serum or CSF. Compared to other autoimmune encephalitides, such as anti-mGluR5 encephalitis, anti-NMDAR encephalitis, or anti-LGI1 encephalitis, poorer outcomes are observed in patients diagnosed with anti-mGluR1 encephalitis (15, 30, 31). Non-paraneoplastic cases of anti-mGluR1 encephalitis reportedly have poorer responses to immunotherapy and higher numbers of relapses (1).

4.1. Limitations

Considering the retrospective nature of the reports included in this review, data retrieval may be incomplete because of the lack of standardization of reporting and testing. Moreover, the generalizability is hindered by the small number of published reports. Language also represents a barrier that has impeded our ability to retrieve and assess publications that were not written in English. Additionally, asserting that certain ataxia was due to cerebellar pathology might not be entirely possible. For example, thalamic lesions can cause cerebellar-like ataxia.

5. Conclusion

Anti-mGluR1 encephalitis is an immune disorder that requires early diagnosis and timely initiation of therapy to achieve improved outcomes. Testing for anti-mGluR1 antibodies should be considered for any acute or subacute cerebellar ataxia, especially following a

TABLE 4 Summary of management of all reported cases of 36 patients with anti-mGluR1 encephalitis.

Case Number	References	Therapy									Remission	Relapses	Antibodies	Duration of follow-up (Month)	Disability
		Glucocorticoids	*IVIg*	*PLEX*	*Rituximab*	*Tacrolimus*	*Azathioprine*	*Mycophenolate mofetil*	*Cyclophosphamide*	*Hydroxychloroquine*					
1	Sillevis Smitt et al. (11)	Yes	Yes	Yes	-	-	-	-	-	-	Complete	None	Resolved	7	No
2	Sillevis Smitt et al. (11)	-	-	Yes	-	-	-	-	-	-	No	None	Persistent	24	Yes
3	Marignier et al. (10)	Yes	Yes	-	-	-	-	Yes	-	-	Partial	None	Persistent	40	Yes
4	Lancaster et al. (9)	Yes	-	-	-	-	-	-	-	-	No	None	NA	36	Yes
5	Iorio et al. (20)	Yes	Yes	-	-	-	-	-	-	-	Partial	None	NA	36	No
6	Lopez-Chiriboga et al. (4)	Yes	-	-	Yes	-	-	-	-	-	Partial	One	NA	17	Yes
7	Lopez-Chiriboga et al. (4)	Yes	Yes	-	-	-	-	-	-	-	No	None	NA	9	NA
8	Lopez-Chiriboga et al. (4)	-	Yes	-	-	-	-	-	-	-	Partial	One	NA	24	Yes
9	Lopez-Chiriboga et al. (4)	Yes	Yes	-	-	-	-	-	-	-	Partial	None	NA	27	Yes
10	Lopez-Chiriboga et al. (4)	Yes	Yes	Yes	-	-	-	-	-	-	Partial	None	NA	11	Yes
11	Lopez-Chiriboga et al. (4)	Yes	-	-	-	-	-	-	-	-	Partial	One	NA	168	Yes
12	Lopez-Chiriboga et al. (4)	-	-	-	-	-	-	-	-	-	Partial	None	NA	6	Yes
13	Lopez-Chiriboga et al. (4)	-	-	-	-	-	-	-	-	-	No	None	NA	4	Yes
14	Lopez-Chiriboga et al. (4)	-	-	-	-	-	-	-	-	-	No	None	NA	60	Yes
15	Lopez-Chiriboga et al. (4)	Yes	-	-	-	-	-	-	-	-	Partial	None	NA	6	No
16	Lopez-Chiriboga et al. (4)	-	-	-	Yes	-	-	-	-	-	No	None	NA	4	Yes
17	Yoshikura et al. (2)	Yes	Yes	Yes	Yes	-	Yes	-	-	-	Partial	Three	Persistent	67	Yes

(Continued)

10.3389/fneur.2023.1142160

TABLE 4 (Continued)

Case Number	References	Therapy									Remission	Relapses	Antibodies	Duration of follow-up (Month)	Disability
		Glucocorticoids	*IVIg*	*PLEX*	*Rituximab*	*Tacrolimus*	*Azathioprine*	*Mycophenolate mofetil*	*Cyclophosphamide*	*Hydroxychloroquine*					
18	Pedroso et al. (21)	Yes	Yes	-	Yes	-	-	-	Yes	-	NA	None	NA	46	NA
19	Christ et al. (12)	Yes	Yes	-	Yes	Yes	-	-	-	-	Partial	One	Persistent	24	Yes
20	Gollion et al. (13)	Yes	Yes	-	-	-	-	-	-	-	Complete	None	Persistent	10	No
21	Chaumont et al. (14)	-	Yes	-	Yes	-	-	-	Yes	-	Partial	None	NA	12	Yes
22	Spatola et al. (15)	Yes	-Yes	-	Yes	-	-	-	Yes	-	No	None	NA	55	Yes
23	Spatola et al. (15)	-	Yes	-	Yes	-	-	-	Yes	-	Partial	None	NA	12	No
24	Spatola et al. (15)	Yes	Yes	-	Yes	-	Yes	-	Yes	-	Partial	None	NA	20	Yes
25	Spatola et al. (15)	-	-	-	-	-	-	-	-	-	No	None	NA	168	Yes
26	Spatola et al. (15)	Yes	-	-	-	-	-	Yes	-	Yes	Complete	One	NA	120	No
27	Spatola et al. (15)	Yes	-	-	-	-	-	Yes	-	-	Partial	None	NA	90	Yes
28	Spatola et al. (15)	Yes	-	-	-	-	-	-	-	-	Complete	None	NA	66	No
29	Spatola et al. (15)	-	-	-	-	-	-	-	-	-	Partial	None	NA	6	Yes
30	Spatola et al. (15)	-	Yes	-	-	-	Yes	-	-	-	Partial	None	NA	84	No
31	Spatola et al. (15)	Yes	Yes	-	-	-	-	-	-	-	Complete	None	NA	2.5	No
32	Bien et al. (16)	Yes	-	-	-	-	-	-	-	-	Complete	None	Persistent	9.5	No
33	Chandler et al. (17)	Yes	Yes	-	-	-	-	-	-	-	Complete	None	NA	17	No
34	Vinke et al. (18)	-	Yes	-	-	-	Yes	Yes	-	-	Partial	Yes*	Persistent	67	No
35	Goh et al. (19)	Yes	-	-	-	-	-	-	-	-	Complete	None	NA	3	No
36	Current case	Yes	Yes	Yes	Yes	-	Yes	-	-	-	Partial	One	Persistent	61	No

IVIg, intravenous immunoglobulin; PLEX, plasma exchange; NA, not applicable*Number of relapses was not mentioned.

10.3389/fneur.2023.1142160

prodrome of febrile illness or associated with malignancy. Escalation to an aggressive therapy approach should be utilized in cases that do not respond to first-line therapies, and extended follow-up durations are required in all cases. More data are required to identify the most appropriate therapeutic plan to resolve clinical manifestations and prevent possible relapses.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving human participants were reviewed and approved by King Abdullah International Medical Research Center. The patients/participants provided their written informed consent to participate in this study.

Author contributions

OK: methodology, formal analysis, investigation, data curation, writing—original draft, and writing—review & editing. SM: conceptualization, methodology, investigation, writing—original draft, and writing—review & editing. SA: conceptualization, methodology, investigation, writing—original draft, writing—review & editing, and supervision. All authors contributed to the article and approved the submitted version.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2023.1142160/full#supplementary-material

References

1. Scotton WJ, Karim A, Jacob S. Glutamate receptor antibodies in autoimmune central nervous system disease: basic mechanisms, clinical features, and antibody detection In: *Glutamate Receptors. Methods in Molecular Biology*. (eds) C. Burger and M. Velardo, *Vol 1941*. New York, NY: Humana Press. (2019). 225–55.

2. Yoshikura N, Kimura A, Fukata M, Fukata Y, Yokoi N, Harada N, et al. Long-term clinical follow-up of a patient with non-paraneoplastic cerebellar ataxia associated with anti-mGluR1 autoantibodies. *J Neuroimmunol*. (2018) 319:63–7. doi: 10.1016/j.jneuroim.2018.04.001

3. Benarroch EE. Metabotropic glutamate receptors: synaptic modulators and therapeutic targets for neurologic disease. *Neurology*. (2008) 70:964–8. doi: 10.1212/01.wnl.0000306315.03021.2a

4. Lopez-Chiriboga AS, Komorowski L, Kümpfel T, Probst C, Hinson SR, Pittock SJ, et al. Metabotropic glutamate receptor type 1 autoimmunity. *Neurology*. (2016) 86:1009–13. doi: 10.1212/WNL.0000000000002476

5. Graus F, Titulaer MJ, Balu R, Benseler S, Bien CG, Cellucci T, et al. A clinical approach to diagnosis of autoimmune encephalitis. *Lancet Neurol*. (2016) 15:391–404. doi: 10.1016/S1474-4422(15)00401-9

6. Nosadini M, Mohammad SS, Ramanathan S, Brilot F, Dale RC. Immune therapy in autoimmune encephalitis: a systematic review. *Expert Rev Neurother*. (2015) 15:1391–419. doi: 10.1586/14737175.2015.1115720

7. Durovic E, Bien C, Bien CG, Isenmann S. MOG antibody-associated encephalitis secondary to Covid-19: case report. *BMC Neurol*. (2021) 21:414. doi: 10.1186/s12883-021-02449-5

8. Murad MH, Sultan S, Haffar S, Bazerbachi F. Methodological quality and synthesis of case series and case reports. *BMJ EBM*. (2018) 23:60–3. doi: 10.1136/bmjebm-2017-110853

9. Lancaster E, Martinez-Hernandez E, Titulaer MJ, Boulos M, Weaver S, Antoine JC, et al. Antibodies to metabotropic glutamate receptor 5 in the Ophelia syndrome. *Neurology*. (2011) 77:1698–701. doi: 10.1212/WNL.0b013e3182364a44

10. Marignier R, Chenevier F, Rogemond V, Smitt PS, Renoux C, Cavillon G, et al. Metabotropic glutamate receptor type 1 autoantibody-associated cerebellitis: a primary autoimmune disease? *Arch Neurol*. (2010) 67:627–30. doi: 10.1001/archneurol.2010.51

11. Sillevis Smitt P, Kinoshita A, De Leeuw B, Moll W, Coesmans M, Jaarsma D, et al. Paraneoplastic cerebellar ataxia due to autoantibodies against a glutamate receptor. *N Engl J Med*. (2000) 342:21–7. doi: 10.1056/NEJM200001063420104

12. Christ M, Müller T, Bien C, Hagen T, Naumann M, Bayas A. Autoimmune encephalitis associated with antibodies against the metabotropic glutamate receptor type 1: case report and review of the literature. *Ther Adv Neurol Disord*. (2019) 12:175628641984741. doi: 10.1177/1756286419847418

13. Gollion C, Dupouy J, Roberts M, Simonetta-Moreau M, Brefel Courbon C, Rascol O, et al. Reversible myoclonus-ataxia encephalitis related to anti-mGLUR1 autoantibodies. *Mov Disord*. (2019) 34:438–9. doi: 10.1002/mds.27634

14. Chaumont H, Petit A, Mameri T, Schollhammer R, Honnorat J, Lannuzel A. Successful management of anti-mGluR1 encephalitis with immunosuppressive treatment: dengue virus as a trigger? *Mov Disord Clin Pract*. (2019) 6:727–8. doi: 10.1002/mdc3.12841

15. Spatola M, Petit Pedrol M, Maudes E, Simabukuro M, Muñiz-Castrillo S, Pinto AL, et al. Clinical features, prognostic factors, and antibody effects in anti-mGluR1 encephalitis. *Neurology*. (2020) 95:e3012–25. doi: 10.1212/WNL.0000000000010854

16. Bien CG, Braig S, Bien CI. Antibodies against metabotropic glutamate receptor type 1 in a toddler with acute cerebellitis. *J Neuroimmunol*. (2020) 348:577366. doi: 10.1016/j.jneuroim.2020.577366

17. Chandler E, Arvantis N, Morgan B. A novel case of idiopathic MGluR1 encephalitis in a pediatric patient. *Child Neurol Open*. (2022) 9:2329048X2210956. doi: 10.1177/2329048X221095695

18. Vinke AM, Zong S, Janssen JH, Correia-Hoffmann C, Mané-Damas M, Damoiseaux JGMC, et al. Autoimmune encephalitis with mGluR1 antibodies presenting with epilepsy, but without cerebellar signs: a case report. *Neurol Neuroimmunol Neuroinflamm*. (2022) 9:e1171. doi: 10.1212/NXI.0000000000001171

19. Goh L, Wang FS, Han VX, Lin JB. Teaching video NeuroImage: subacute cerebellar ataxia in an adolescent with antibodies against metabotropic glutamate receptor type 1. *Neurology*. (2022) 99. doi: 10.1212/WNL.0000000000201268

20. Iorio R, Damato V, Mirabella M, Vita MG, Hulsenboom E, Plantone D, et al. Cerebellar degeneration associated with mGluR1 autoantibodies as a paraneoplastic manifestation of prostate adenocarcinoma. *J Neuroimmunol*. (2013) 263:155–8. doi: 10.1016/j.jneuroim.2013.07.015

21. Pedroso JL, Dutra LA, Espay AJ, Höftberger R, Barsottini OGP. Video NeuroImages: head titubation in anti-mGluR1 autoantibody-associated cerebellitis. *Neurology*. (2018) 90:746–7. doi: 10.1212/WNL.0000000000005338

22. Guergueltcheva V, Azmanov DN, Angelicheva D, Smith KR, Chamova T, Florez L, et al. Autosomal-recessive congenital cerebellar ataxia is caused by mutations in metabotropic glutamate receptor 1. *Am J Hum Genet*. (2012) 91:553–64. doi: 10.1016/j.ajhg.2012.07.019

23. Bresler R, Harry W, Chow D, Lim R. 18 F-fluorodeoxyglucose positron emission tomography/computed tomography in the diagnosis of suspected paraneoplastic syndromes: a retrospective analysis. *World J Nucl Med*. (2020) 19:124–30. doi: 10.4103/wjnm.wjnm_48_19

24. Titulaer MJ, Soffietti R, Dalmau J, Gilhus NE, Giometto B, Graus F, et al. Screening for tumours in paraneoplastic syndromes: report of an EFNS task force. *Eur J Neurol*. (2011) 18:19–e3. doi: 10.1111/j.1468-1331.2010.03220.x

25. Patel RR, Subramaniam RM, Mandrekar JN, Hammack JE, Lowe VJ, Jett JR. Occult malignancy in patients with suspected paraneoplastic neurologic syndromes: value of positron emission tomography in diagnosis. *Mayo Clin Proc*. (2008) 83:917–22. doi: 10.4065/83.8.917

26. Whitacre CC, Reingold SC, O'Looney PA, Blankenhorn E, Brinley F, Collier E, et al. A gender gap in autoimmunity: task force on gender. *Multiple Sclerosis Autoimmunity Sci*. (1999) 283:1277–8. doi: 10.1126/science.283.5406.1277

27. Gresa-Arribas N, Titulaer MJ, Torrents A, Aguilar E, McCracken L, Leypoldt F, et al. Antibody titres at diagnosis and during follow-up of anti-NMDA receptor encephalitis: a retrospective study. *Lancet Neurol*. (2014) 13:167–77. doi: 10.1016/S1474-4422(13)70282-5

28. Ferraguti F, Crepaldi L, Nicoletti F. Metabotropic glutamate 1 receptor: current concepts and perspectives. *Pharmacol Rev*. (2008) 60:536–81. doi: 10.1124/pr.108.000166

29. Ancona C, Masenello V, Tinnirello M, Toscano LM, Leo A, La Piana C, et al. Autoimmune encephalitis and other neurological syndromes with rare neuronal surface antibodies in children: a systematic literature review. *Front Pediatr*. (2022) 10:866074. doi: 10.3389/fped.2022.866074

30. Spatola M, Sabater L, Planagumà J, Martínez-Hernandez E, Armangué T, Prüss H, et al. Encephalitis with mGluR5 antibodies: symptoms and antibody effects. *Neurology*. (2018) 90:e1964–72. doi: 10.1212/WNL.0000000000005614

31. van Sonderen A, Thijs RD, Coenders EC, Jiskoot LC, Sanchez E, de Bruijn MAAM, et al. Anti-LGI1 encephalitis: clinical syndrome and long-term follow-up. *Neurology*. (2016) 87:1449–56. doi: 10.1212/WNL.0000000000003173

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 02 May 2023
DOI 10.3389/fimmu.2023.1106490

OPEN ACCESS

EDITED BY
Harald Hegen,
Innsbruck Medical University, Austria

REVIEWED BY
Benjamin Victor Ineichen,
University of Zurich, Switzerland
Souraya El Sankari,
Cliniques Universitaires Saint-Luc, Belgium

*CORRESPONDENCE
Limei Wang
lylimeiwang@126.com

SPECIALTY SECTION
This article was submitted to Multiple Sclerosis and Neuroimmunology, a section of the journal Frontiers in Immunology

RECEIVED 23 November 2022
ACCEPTED 11 April 2023
PUBLISHED 02 May 2023

CITATION
Zhu B, Sun M, Yang T, Yu H and Wang L (2023) Clinical, imaging features and outcomes of patients with anti-GFAP antibodies: a retrospective study. *Front. Immunol.* 14:1106490. doi: 10.3389/fimmu.2023.1106490

Clinical, imaging features and outcomes of patients with anti-GFAP antibodies: a retrospective study

Bingqing Zhu, Mengyang Sun, Ting Yang, Haizhen Yu and Limei Wang*

Department of Neurology, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, Henan, China

Objective: To evaluate and compare the clinical features, imaging, overlapping antibodies, and prognosis of pediatric and adult patients with anti-GFAP antibodies.

Methods: This study included 59 patients with anti-GFAP antibodies (28 females and 31 males) who were admitted between December 2019 and September 2022.

Results: Out of 59 patients, 18 were children (under 18 years old), and 31 were adults. The overall cohort's median age at onset was 32 years old, 7 for children, and 42 for adults. There were 23 (41.1%) patients with prodromic infection, 1 (1.7%) patient with a tumor, 29 (53.7%) patients with other non-neurological autoimmune diseases, and 17 (22.8%) patients with hyponatremia. Fourteen (23.7%) patients had multiple neural autoantibodies, with the AQP4 antibody being the most common. Encephalitis (30.5%) was the most common phenotypic syndrome. Common clinical symptoms included fever (59.3%), headache (47.5%), nausea and vomiting (35.6%), limb weakness (35.6%), and disturbance of consciousness (33.9%). Brain MRI lesions were primarily located in the cortex/subcortex (37.3%), brainstem (27.1%), thalamus (23.7%), and basal ganglia (22.0%). Spinal cord MRI lesions often involved the cervical and thoracic spinal cord. There was no statistically significant difference in the MRI lesion site between children and adults. Out of 58 patients, 47 (81.0%) had a monophasic course, and 4 died. The last follow-up showed that 41/58 (80.7%) patients had an improved functional outcome (mRS <3), and children were more likely than adults to have no residual disability symptoms ($p = 0.001$).

Conclusion: There was no statistically significant difference in clinical symptoms and imaging findings between children and adult patients with anti-GFAP antibodies; Patients with anti-GFAP antibodies may present with normal MRI findings or delayed MRI abnormalities, and patients with overlapping antibodies

were common. Most patients had monophasic courses, and those with overlapping antibodies were more likely to relapse. Children were more likely than adults to have no disability. Finally, we hypothesize that the presence of anti-GFAP antibodies is a non-specific witness of inflammation.

KEYWORDS

glial fibrillary astrocytic protein antibodies, clinical characteristics, imaging features, overlapping antibodies, prognosis

1 Introduction

Glial fibrillary acidic protein (GFAP) is an intermediate filament found primarily in astrocytes that serves as the skeleton of the cell and aids in cell communication and the formation of the blood-brain barrier. Abnormal regulation and expression of GFAP also play a key role in the onset and progression of various neurological diseases, including inflammation, traumatic brain injury, neurodegeneration, and so on (1–3). The Mayo Clinic (4) was the first to report a novel meningoencephalomyelitis with GFAP-IgG as a specific antibody that primarily affects the meninges, brain, spinal cord, and optic nerves in 2016. The condition was called autoimmune GFAP astrocytopathy (GFAP-A) (4). This neuroimmune disease has a distinct imaging feature known as paraventricular linear radial enhancement (4–7). The onset of this disease may be associated with a tumor or a viral infection, and it is frequently associated with overlapping antibodies (4, 5, 8–10). However, the French cohort questioned the existence of overlapping antibodies (11). Because the target antigen is intracellular, the pathogenicity of GFAP antibodies is debatable. The pathophysiological role of anti-GFAP antibodies in neuroimmunity is currently unknown. Despite various studies investigating the clinical characteristics and possible pathological features of patients with anti-GFAP antibodies, there is still no international consensus and guideline for diagnosis and treatment due to the disease's heterogeneity. More diagnostic clues are required to develop early consensus on GFAP autoimmune diseases. This study aimsto describe the clinical characteristics, imaging, overlapping antibodies, and prognosis of pediatric and adult patients with anti-GFAP antibodies, as well as to speculate on the potential pathogenic mechanism of GFAP antibodies.

2 Materials and methods

2.1 Patients

From December 2019 to September 2022, we reviewed the medical records of 59 patients who had anti-GFAP antibodies in their serum or cerebrospinal fluid (CSF) and were consecutively admitted to the First Affiliated Hospital of Zhengzhou University. Inclusion criteria included (1): CSF or serum GFAP antibody-positive patients with one or more clinical manifestations of meningitis, encephalitis, myelitis, or optic neuritis (2); available clinical data; and (3) reasonable exclusion of other disorders Exclusion criteria include (1): patients with positive serum GFAP antibodies after traumatic brain injury or spinal cord injury (2); patients with glioma. Demographics, clinical manifestations, imaging, laboratory results, immunotherapy, disease course, and prognosis were all described. The modified Rankin Scale (mRS) was used to assess disease severity, and residual disability was followed up by phone. mRS < 3 was considered to be a good functional outcome.

2.2 Laboratory and imaging examination

Lumbar puncture was performed at least once on all patients. CSF white cell count, protein content, and oligoclonal bands (OCBs) were recorded at the earliest available time. Cell-based assays (CBA) were used to detect anti-GFAP antibodies in patient serum or CSF. Demyelinating antibodies (AQP4, MOG), autoimmune encephalitis-associated antibodies (such as NMDAR, GAD65, GABABR, LGI1, Caspr2, IGLON5, mGluR1, mGluR5, Hu, Ri, Yo, etc.) and systemic autoimmunity antibodies (such as RA, ANA, ANCA, dsDNA, CCP, SSA, SSB, etc.) were also detected. CSF from all patients was tested for viral, bacterial, and tuberculous bacteria to rule out CNS (central nervous system) infections. All patients had magnetic resonance imaging (MRI) of the brain or spinal cord performed at the time of admission on the same 3T MAGNETOM Skyra scanner (Siemens Healthcare, Erlangen, Germany), and some of them also received intravenous gadolinium to assess potential contrast enhancement. SE T1WI (TR = 488 ms, TE = 15 ms) and TSE T2WI (TR = 4000 ms, TE = 103 ms) sequences were used in transverse view, and T2WI - FLAIR (TR = 9000 ms, TE =81 ms) sequences in coronal view. The scanning matrix was 384 × 384, the field of vision was 230 mm×230 mm, the layer thickness was 6 mm, the slice gap was 1.2 mm, and the number of scanning layers was 18 ~ 20 layers. Spinal cord MRI scans were recorded using sagittal and transverse TSE T1WI and fat suppression sequence T1WI (cervical TR = 480 ms, TE = 9.4 ms; thoracolumbar TR = 337 ms, TE = 9.4 ms), TSE T2WI and fat suppression T2WI (cervical TR = 2700 ms, TE = 82 ms; thoracolumbar TR = 3500 ms, TE =87 ms), in which the cervical field of vision was 240 mm × 240 mm, thoracolumbar visual field was 340 mm × 340 mm, scan matrix was 384 × 384, the layer

thickness was 3 mm, the slice gap was 0.3 mm, and the number of scanning layers was 15–18. The MRIs of the brain and spinal cord were reviewed by one neurologist and one neuroradiologist. A routine thyroid color ultrasound evaluation, abdominal color ultrasound, and chest CT examination were performed on all patients to rule out some common systemic tumors.

2.3 Standard protocol approvals

This study was ethically approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University (2022-KY-1205–002).

2.4 Statistics

Patients were divided into two groups based on their age of onset: pediatric (<18 years old) and adult (≥18 years old). Statistical analyses and data visualization were performed using SPSS 26.0 and OriginPro 2021 to compare the clinical features and prognosis of pediatric and adult patients with anti-GFAP antibodies. To describe normally distributed continuous variables, means (standard deviation) were used. In contrast, for non-normally distributed continuous variables, the median (interquartile range) was used, and for categorical variables, the frequency (percentage) was used. The Wilcoxon rank-sum test (continuous variables) and the chi-squared test or Fisher exact test were used to compare two groups (categorical variables). P-values of <0.05 (two-sided) were considered to be statistically significant. Due to the exploratory nature of this study, we did not correct for multiple comparisons.

3 Results

3.1 General Conditions

This study included 59 patients (28 females and 31 males) who had anti-GFAP antibodies in their CSF or serum. Four patients (6.8%) were only positive for serum antibody, while the remaining patients had anti-GFAP antibody positive CSF with or without positive serum antibody. Furthermore, serum antibody titers were higher in four patients than CSF antibody titers (6.8%). The median duration of follow-up was 9 months (Table 1). The overall cohort's median age at

TABLE 1 Demographics and clinical features of GFAP-IgG patients.

Characteristic	Total	Pediatric patients	Adult patients	P
Number of patients, n	59	18	41	
Female:Male (% female)	28:31 (47.5%)	8:10 (44.4%)	20:21 (48.8%)	0.759
Age at onset, years, median (IQR)	32 (34.5)	7 (7.5)	42 (19.5)	
Follow-up, months, median (IQR)	9 (15)	12 (16)	9 (14)	
Comorbidity, n/total (%)				
Coexisting autoimmune diseases	29/54 (53.7%)	8/15 (53.3%)	21/39 (53.8%)	0.973
Tumor	2/59 (3.4%)	0	2/41 (4.9%)	1
Hyponatremia	17/59 (22.8%)	4/18 (22.2%)	13/41 (31.7%)	0.459
Monophasic course, n/total (%)	47/58 (81.0%)	16/18 (88.9%)	31/40 (77.5%)	0.508
Symptoms at presentation, n/total (%)				
Fever	35/59 (59.3%)	13/18 (72.2%)	22/41 (53.7%)	0.181
Headaches	28/59 (47.5%)	9/18 (50%)	19/41 (46.3%)	0.796
Nausea and vomiting	21/59 (35.6%)	7/18 (38.9%)	14/41 (34.1%)	0.726
Disturbance of consciousness	20/59 (33.9%)	6/18 (33.3%)	14/41 (34.1%)	0.952
Dizzy	14/59 (23.7%)	4/18 (22.2%)	10/41 (24.4%)	1
Psychiatric symptoms	8/59 (13.6%)	2/18 (11.1%)	6/41 (14.6%)	1
Cognitive deficits	7/59 (11.9%)	1/18 (5.6%)	6/41 (14.6%)	0.578
Seizure	5/59 (8.5%)	1/18 (5.6%)	4/41 (9.8%)	0.979
Impaired vision	6/59 (10.2%)	3/18 (16.7%)	3/41 (7.3%)	0.531
Diplopia	5/59 (8.5%)	2/18 (11.1%)	3/41 (7.3%)	1
Ataxia	3/59 (5.1%)	0	3/41 (7.3%)	0.546

(Continued)

TABLE 1 Continued

Characteristic	Total	Pediatric patients	Adult patients	P
Involuntary movement	7/59 (11.9%)	2/18 (11.1%)	5/41 (12.2%)	1
Optic disc edema	4/59 (6.8%)	1/18 (5.6%)	3/41 (7.3%)	1
Cranial nerve palsy	5/59 (8.5%)	2/18 (11.1%)	3/41 (7.3%)	1
Speech disorder	4/59 (6.8%)	0	4/41 (9.8%)	0.418
Walking unstable	4/59 (6.8%)	0	4/41 (9.8%)	0.418
Area postrema syndrome	2/59 (3.4%)	1/18 (5.6%)	1/41 (2.4%)	0.521
Weakness	21/59 (35.6%)	7/18 (38.9%)	14/41 (34.1%)	0.726
Numbness	10/59 (16.9%)	0	10/41 (24.4%)	0.055
Autonomic dysfunction	11/59 (18.6%)	4/18 (22.2%)	7/41 (17.1%)	0.418
Paresthesias	4/59 (6.8%)	0	4/41 (9.8%)	0.917
ICU admission	19/59 (32.2%)	7/18 (38.9%)	12/41 (29.3%)	0.466
Tracheal intubation	13/59 (22.0%)	2/18 (11.1%)	11/41 (26.8%)	0.317
mRS at the peak of attack, median (IQR)	4 (3)	3 (3)	4 (3)	0.692
mRS at discharge, median (IQR)	2 (2)	1 (2)	2 (2.5)	0.035
mRS score at the last follow-up, median (IQR)	1 (3)	0 (1.25)	1.5 (3)	0.003
mRS>2 at the last follow-up	17/58 (29.3%)	3/18 (16.7%)	14/40 (35%)	0.156
Sequelae, n/total (%)				
No disability	23/58 (39.7%)	13/18 (72.2%)	10/40 (25%)	0.001
Motor	17/58 (29.3%)	4/18 (22.2%)	13/40 (32.5%)	0.426
Sensory	12/58 (20.7%)	2/18 (11.1%)	10/40 (25%)	0.391
Vision impairment	6/58 (10.3%)	2/18 (11.1%)	4/40 (10%)	1
Autonomic dysfunction	6/58 (10.3%)	2/18 (11.1%)	4/40 (10%)	1
Cognitive impairment	5/58 (8.6%)	0/18	5/40 (12.5%)	0.288
Involuntary movement	2/58 (3.4%)	1/18 (5.6%)	1/40 (2.5%)	0.528
Speech disorder	2/58 (3.4%)	0	2/40 (5%)	1
Dysphagia	1/58 (1.7%)	0	1/40 (2.5%)	1
Death	4/58 (6.9%)	0	4/40 (10%)	0.406

IQR, interquartile range; ICU, intensive care unit; mRS, modified Rankin Scale.

onset was 32, children aged 7 and adults aged 42. At the time of the first attack, 18 of the 59 GFAP-IgG-positive patients were under the age of 18. At the time of onset, only three patients (5.1%) were over 60 years old. The patient population in the other three age groups is comparable (20 patients in 0–20 years old, 15 patients in 21–40 years old, and 21 patients in 41–60 years old, respectively).

23/56 (41.1%) patients had prodromic infection or vaccination before or at the time of onset, including 11/17 (64.7%) children and 12/39 (30.8%) adults. One patient had been immunized against COVID-19 one week before the onset of the disease. One patient had been infected with the varicella-zoster virus one month before the onset of neurological symptoms, and another had been infected with the herpes simplex virus (HSV) 2 weeks before. One patient had viral encephalitis one month prior, and the other had staphylococcal meningitis 2 weeks before the GFAP-IgG was discovered. A next-generation sequencing (NGS) test detected CSF infection in ten patients, including nine Human herpesviruses (Epstein-Barr virus n = 6, HSV n = 1, Human herpesvirus 7 n = 2) cases and one Staphylococcus case. There were also 4 cases with hepatitis B virus cases, 1 with tuberculosis, 1 with influenza B virus case, and 2 with mycoplasma cases. In all patients, only one (1.7%) was found to have a tumor, who was hospitalized with neurological symptoms and later diagnosed with papillary thyroid cancer. In addition, hyponatremia was present in 17/59 (22.8%) patients. Furthermore, 29/54 (53.7%) patients had other non-nervous system autoimmune diseases, with antibodies for these diseases, including anti-thyroid, antinuclear, antineutrophil cytoplasmic, antiphospholipid, rheumatoid factors, anti-dsDNA antibody, and Sjogren's syndrome antibodies, among others. There was no statistically significant difference between

10.3389/fimmu.2023.1106490

children and adults in tumor, hyponatremia, or autoimmune antibodies (Table 1).

3.2 Clinical phenotype and clinical symptoms

The clinical course of 59 GFAP-IgG-positive children and adults is depicted in Figure 1 to visually describe the clinical phenotypes of an acute attack. Among the 59 patients in the cohort, encephalitis (18/59, 30.5%) was the most common clinical phenotypic syndrome, followed by encephalomyelitis (15/59, 25.4%), meningoencephalomyelitis (10/59, 16.9%), meningitis (7/59, 11.9%), myelitis (7/59, 11.9%), meningoencephalitis (3/59, 5.1%) and optic neuritis (2/59, 3.4%). Encephalomyelitis (9/18, 50%) and encephalitis (15/41, 36.6%) were the most common clinical phenotypic syndromes in children and adults, respectively. Fever (59.3%), headache (47.5%), nausea and vomiting (35.6%), limb weakness (35.6%), disturbance of consciousness (33.9%), dizziness (23.7%), autonomic dysfunction (18.6%), and limb numbness were the most common clinical manifestations in the entire cohort (16.9%). Moreover, other clinical manifestations were cognitive impairment, involuntary movement, visual impairment, seizures, cranial nerve palsy, diplopia, optic disc edema, speech disorders, walking instability, ataxia, area postrema syndrome (APS), and paresthesia, etc. (Table 1). Clinical manifestations did not differ significantly between children and adults.

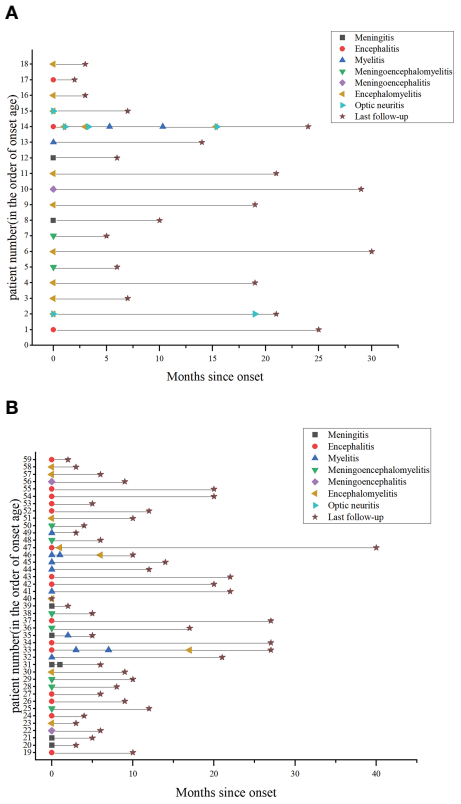


FIGURE 1
Clinical phenotype and disease course in patients with anti-GFAP antibodies. **(A)** Pediatric patients' clinical phenotype and disease course. **(B)** Adult patients' clinical phenotype and disease course.

3.3 Cerebrospinal fluid analysis

CSF test results were available in 58 patients at the time of their initial presentation. Pleocytosis (> 5 cells/mm^3) was found in 46 patients (79.3%), elevated protein level (> 0.5 g/L) in 35 patients (60.3%), and hypoglycorrhachia in 12 patients (20.7%). In the meantime, CSF-restricted OCBs (type 2) were found in 22 (40.7%) patients. CSF-elevated protein levels differed between children and adults (P = 0.005). (Table 2).

TABLE 2 Diagnostic testing and treatment of GFAP-IgG patients.

Characteristic	Value	Pediatric patients	Adult patients	P
CSF analysis at onset, n/total (%)				
Pleocytosis (> 5 cells/mm3)	46/58(79.3%)	15/18(83.3%)	31/40(77.5%)	0.875
Elevated protein level (> 0.5 g/L)	35/58 (60.3%)	6/18(33.3%)	29/40(72.5%)	0.005
Hypoglycorrhachia (<2.5 mmol/L)	12/58 (20.7%)	2/18(11.1%)	10/40(25%)	0.391
CSF Oligoclonal Bands (type 2)	22/54(40.7%)	6/14(42.9%)	16/40(40.0%)	0.851
Overlapping antibody n/total (%)				
AQP4-IgG	7/59(11.9%)	2/18(11.1%)	5/41(12.2%)	
MOG-IgG	5/59(8.5%)	3/18(16.7%)	2/41(4.9%)	
NMDAR-IgG	3/59(5.1%)	1/18(5.6%)	2/41(4.9%)	
Others*	3/59(5.1%)	0/18	3/41(7.3%)	
MRI, n/total (%)				

(Continued)

TABLE 2 Continued

Characteristic	Value	Pediatric patients	Adult patients	P
MRI (brain) abnormalities	43/59(72.9%)	16/18(88.9%)	27/41(65.9%)	0.130
Gadolinium enhanced lesion (brain)	23/40(57.5%)	5/7(71.4%)	18/33(54.5%)	0.689
Leptomeninges enhancement	11/40(27.5%)	2/7(28.6%)	9/33(27.3%)	1
Perivascular-radial enhancement	4/40(10.0%)	0	4/33(12.1%)	1
Lesion location				
Juxtacortical	22/59(37.3%)	8/18(44.4%)	14/41(34.1%)	0.451
Periventricular	9/59(15.3%)	2/18(11.1%)	7/41(17.1%)	0.847
Corpus callosum	9/59(15.3%)	4/18(22.2%)	5/41 (12.2%)	0.553
Basal ganglia	13/59(22.0%)	4/18(22.2%)	9/41 (22.0%)	1
Thalamus	14/59(23.7%)	6/18(33.3%)	8/41 (19.5%)	0.414
Brachium pontis	4/59(6.8%)	1/18(5.6%)	3/41(7.3%)	1
Brainstem tegmentum	16/59(27.1%)	3/18(33.3%)	10/41(24.4%)	0.751
Cerebellar hemispheres	5/59(8.5%)	2/18 (11.1%)	3/41(7.3%)	1
MRI (spinal cord) abnormalities	35/50(70.0%)	13/16(81.3%)	22/34(64.7%)	0.390
Gadolinium enhanced lesion (spinal cord)	13/21(61.9%)	3/3(100%)	10/18(55.6%)	0.409
LETM	17/50(34.0%)	8/16(50.0%)	9/34(26.5%)	0.101
Cervical cord	26/50 (52.0%)	10/16(62.5%)	16/34(47.1%)	0.308
Thoracic cord	24/50(48.%)	10/16(62.5%)	14/34(41.2%)	0.159
Medullary cone	2/50(4.0%)	2/16(12.5%)	0/34	0.098
Acute phase treatment n/total (%)				
IVMP alone	26/59(44.1%)	5/18(27.8%)	21/41(51.2%)	
IVMP+IVIG	22/59(37.3%)	11/18(61.1%)	11/41(26.8%)	
Others^	7/59(11.9%)	2/18(11.1%)	5/41(12.2%)	
No immunotherapy	4/59(6.8%)	0	4/41(9.8%)	
Maintenance therapy n/total (%)				
Glucocorticoids alone	40/59(67.8%)	15/18(83.3%)	25/41(61.0%)	
Mycophenolate mofetil with or without glucocorticoids	9/59(15.3%)	1/18(5.6%)	8/41(19.5%)	
Others #	2/59(3.4%)	1/18(5.6%)	1/41(2.4%)	

*: GAD65-IgG, Yo-IgG, GlyR-IgG
^:IVMP+RTX,n = 2;IVMP+PE+IVIG,n = 2;IVMP+IVIG+RTX,n = 1;IVMP+PE,n = 1;IVIG+EIA,n = 1.
#: tacrolimus in one adult patients, azathioprine in one pediatric patient
CSF, cerebrospinal fluid; AQP4, Aquaporin 4; MOG, Myelin oligodendrocyte glycoprotein; NMDAR, N-methyl-D-aspartate receptor; MRI, magnetic resonance imaging; LETM, longitudinally extensive transverse myelitis; IVIG, intravenous immunoglobulin; IVMP, intravenous methylprednisolone; RTX, rituximab; PE, plasma exchange; EIA, extracorporeal immunoadsorption.

3.4 Imaging manifestations

All patients underwent brain MRIs. During the acute phase, 43 patients (72.9%) had abnormal brain MRIs, displaying hyperintensities on T2-weighted and fluid-attenuated inversion recovery (FLAIR) sequences (Figure 2). Five patients' brain MRIs were normal at the start of their symptoms but gradually became abnormal. In four patients with clinical manifestations, the brain MRIs revealed no lesions. The most common lesions were in the cortical/subcortical (37.3%), brainstem (27.1%), thalamus (23.7%), basal ganglia (22.0%), periventricular (15.3%), and corpus callosum (15.3%). The cerebellar hemisphere (8.5%) and the pontine arm (6.8%) were also unusual sites (Table 2). One of the most common imaging features was lesions in the bilateral thalamus (20.3%) and bilateral basal ganglia (18.6%) (Figure 2). In 6/59 (10.2%) patients, reversible splenial lesion syndrome (RESLES) was discovered. Among the 40 patients who underwent brain gadolinium enhancement MRI, 23 had enhanced lesions, 11 had leptomeningeal enhancement, but only 4 patients had

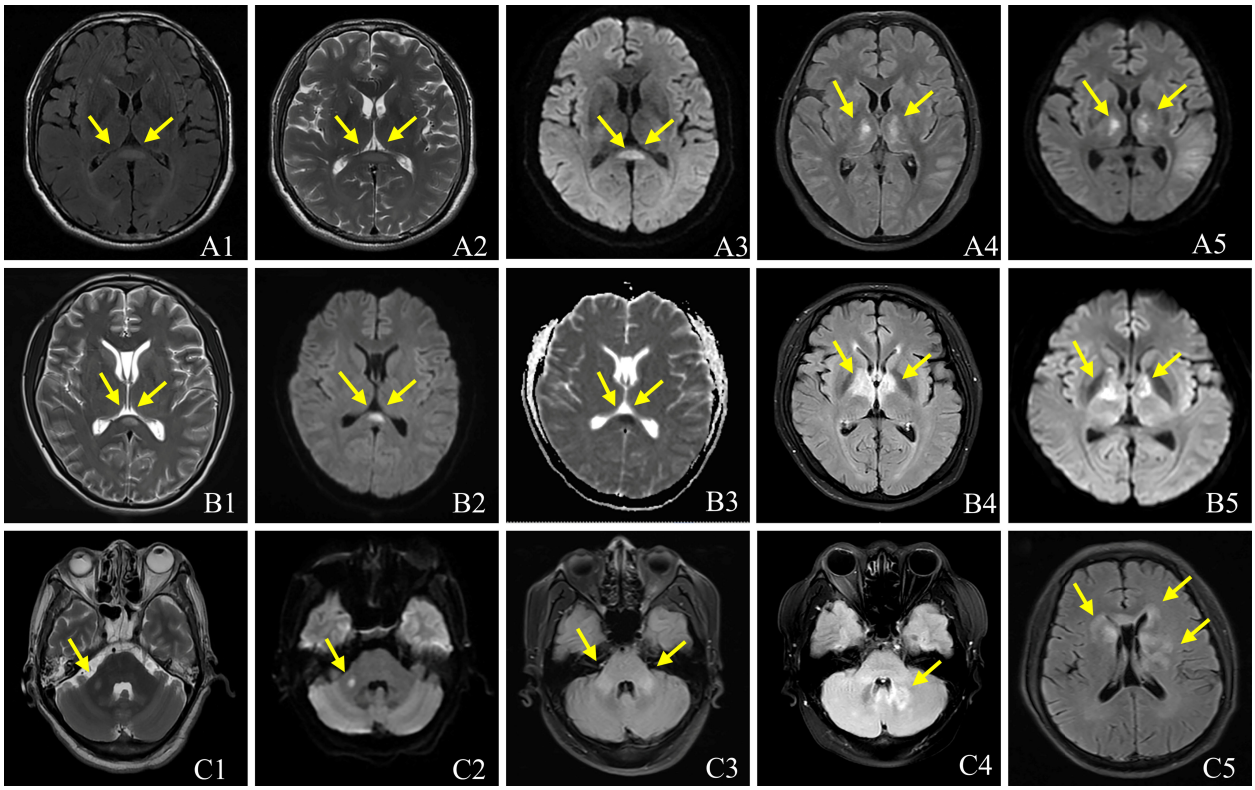


FIGURE 2
Brain MRI characteristics of patients with Anti-GFAP antibodies. The imaging looks like a reversible splenial lesion **(A1-A3, B1-B3)**. T2 FLAIR **(A4, B4)** and Diffusion Weighted Imaging **(A5, B5)** show lesions in bilateral thalamus, T2-weighted **(C1)** image and FLAIR **(C2, C3)** show lesions in brachium pontis; T2 FLAIR shows lesions in cerebellum **(C4)**, basal ganglia **(C5)** and paraventricular **(C5)**.

periventricular or cerebellar linear enhancement (Figure 3). There were 50 patients with spinal cord MRI, 35 (70.0%) of whom had abnormal signals (Figure 4), and 17 (34.0%) had a longitudinal extension to more than three adjacent vertebral segments (longitudinal extensive transverse myelitis, LETM). Cervical, thoracic, and spinal conus lesions were responsible for 26/50 (52.0%), 24/50 (48.0%), and 2/50 (4.0%) of the cases, respectively. The lumbar spinal cord was free of lesions. In 21 patients, enhanced MRI of the spinal cord was performed, and 13 cases were found to have enhanced lesions, including two cases of spinal membrane enhancement and one case of cauda equina nerve enhancement. Table 2 shows that there is no statistically significant difference in the MRI lesion site between children and adults.

3.5 Overlapping antibodies

Four males (28.6%) and ten females (71.4%) were among the 59 patients who coexisted with other neural autoantibodies (Table 2). Table 3 shows the information on patients who have overlapping antibodies. Four of fourteen patients (28.6%) with overlapping antibodies relapsed. Six of the 14 patients (14, 15, 32, 41, 46, and 52) were tested positive for AQP4 antibody in serum, GFAP and AQP4 antibody in CSF. Four patients with positive MOG antibodies were combined separately (patients 2, 6, 24, and 33, GFAP and MOG in serum, GFAP in CSF).

The first symptoms of Patient 14 were nausea and vomiting, which were quickly followed by fever, shaky walking, central facial paralysis, blurred vision, and limb weakness. Despite various immunotherapies, the condition recurred six times. At the last follow-up, Patient 46 still had weakness in both lower limbs due to recurrent myelitis-like symptoms. With dysarthria, asphyxia, blurred vision, and other symptoms, Patient 52 was discharged. Patient 2 presented with a fever and blurred vision in the right eye and was given methylprednisolone intravenously (IVMP). The symptoms were completely resolved at discharge and were treated with oral glucocorticoids and mycophenolate mofetil. The patient, however, lost vision in his left eye three months after glucocorticoids withdrawal and was discharged with visual impairment. Patient 6 was admitted to the hospital for two days with the chief complaint of headache, diplopia, and low spirits. He might have had viral encephalitis a month before. His symptoms completely resolved after IVMP combined with IVIG treatment, and he was discharged and diagnosed with acute disseminated encephalomyelitis (ADEM). The clinical manifestations of patient 30 were dizziness, unsteady walking, and limb weakness (anti-Yo antibodies in serum, anti-GFAP antibodies in CSF). Cerebellar and brainstem inflammation was diagnosed based on the clinical symptoms, but brain MRIs revealed no obvious abnormalities. The patient was later transferred to a nearby hospital and was still having difficulty walking at the time of the last check-up. In one

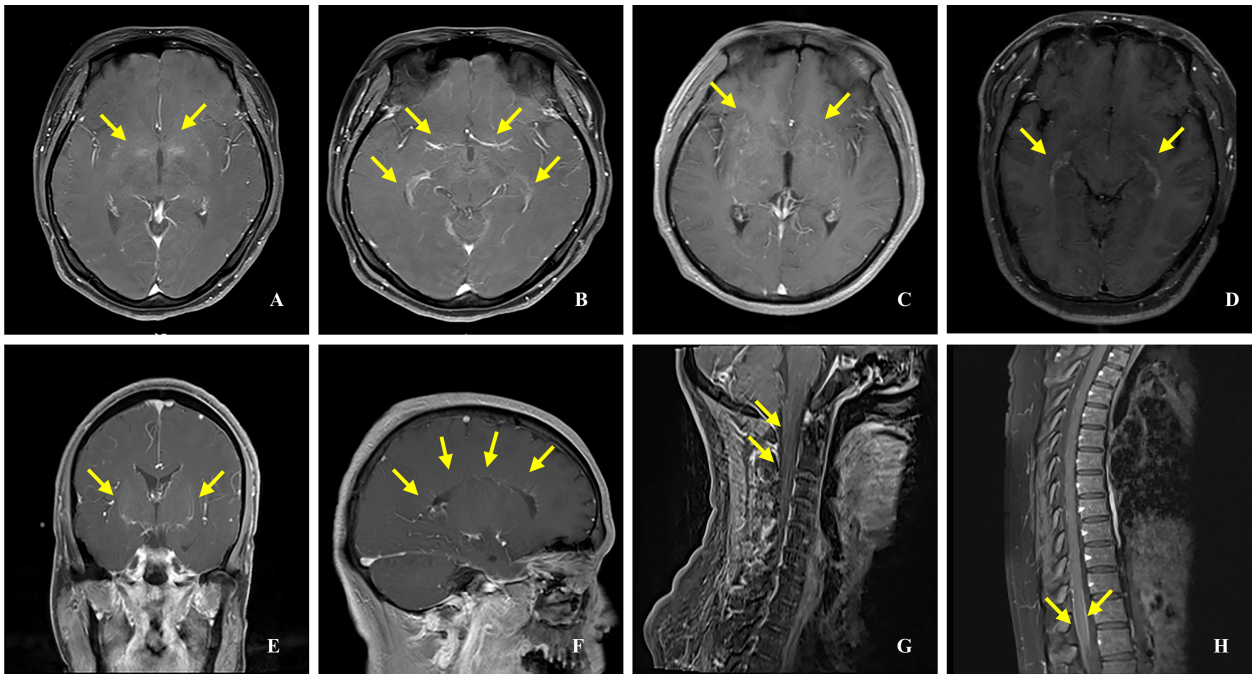


FIGURE 3
Gadolinium enhancement MRI in patients with anti-GFAP antibodies. **(A, C)** Punctate enhancement lesions. **(B, D)** Periependymal enhancement. **(E)** Linear enhancement of the cerebellum. **(F)** Linear enhancement perpendicular to the ventricle. **(G)** Linear enhancement of the spinal cord. **(H)** Enhancement of the spinal cord membranes.

patient (patient 36), GFAP and NMDAR antibodies were found in the CSF, as well as fever, lower limb weakness, and confusion. With an mRS score of 5, the patient was admitted to the ICU, and his family refused IVMP combined with IVIG treatment, requesting transfer to another hospital for treatment. The patient had fully recovered at the time of the last check-up.

3.6 Treatment, outcome, and follow-up

During the course of the disease, 19 (32.2%) and 13 (22.0%) patients were admitted to ICU and intubated, respectively. Two patients' families refused immunotherapy, and another two patients did not receive immunotherapy because they were diagnosed with clinically isolated syndrome (CIS) and cerebral infarction, respectively. Only 55 patients received acute-phase immunotherapy. 26/55 patients received IVMP alone, 22/55 received IVMP in combination with IVIG, and 7/55 received other combination immunotherapies.

Patients were contacted by phone in all cases, except one, who was missed due to a change in the phone number. During the follow-up period, 49 patients received oral glucocorticoids and were tapered, with 11 receiving additional immunosuppressive drugs (mycophenolate n = 9, azathioprine n = 1, tacrolimus n = 1). Figure 5 depicts the mRS distribution of the 59 patients at the peak of the attack, discharge, and the last follow-up. There were significant differences in mRS scores between children and adults at discharge ($p = 0.035$) and at the last follow-up ($p = 0.003$) 41/58 (80.7%) patients had good functional outcomes at the last follow-up (mRS<3). Recurrence occurred in 7/58 (12.1%) patients (2 children and 5 adults), with 4 patients recurring only once, 1 patient recurring twice (coexistence of AQP4-IgG), 1 patient recurring three times (coexistence of MOG-IgG), and 1 patient recurring six times (coexistence of AQP4-IgG). There were no residual symptoms in 23/58 patients (39.7%), including 13/18 (72.2%) children and 10/40 (25%) adults, which was statistically significant ($p = 0.001$). At a median of 9 months (range 0–40 months), 31/58 (53.4%) patients had residual symptoms. The most common type of disability was myelitis-like symptom. Blurred vision, cognitive dysfunction, involuntary movement, slurred speech, and dysphagia were among the other uncommon disabilities. Four patients (6.9%) died between the onset of the disease and the last follow-up.

4 Discussion

Our study included 59 patients with anti-GFAP antibodies in CSF or serum (patients with the meningoencephalomyelitis phenotype and excluding other diagnoses) to compare clinical characteristics, imaging, overlap antibodies, and prognosis in pediatric and adult patients, which has been rarely reported in previous studies.

In this study, the proportion of male and female patients was roughly equal older patients were less likely to be affected. Patients in our cohort frequently presented with symptoms of meningitis, encephalitis, myelitis, and optic neuritis. Non-specific symptoms such as fever, headache, nausea, and vomiting, as well as myelitis-

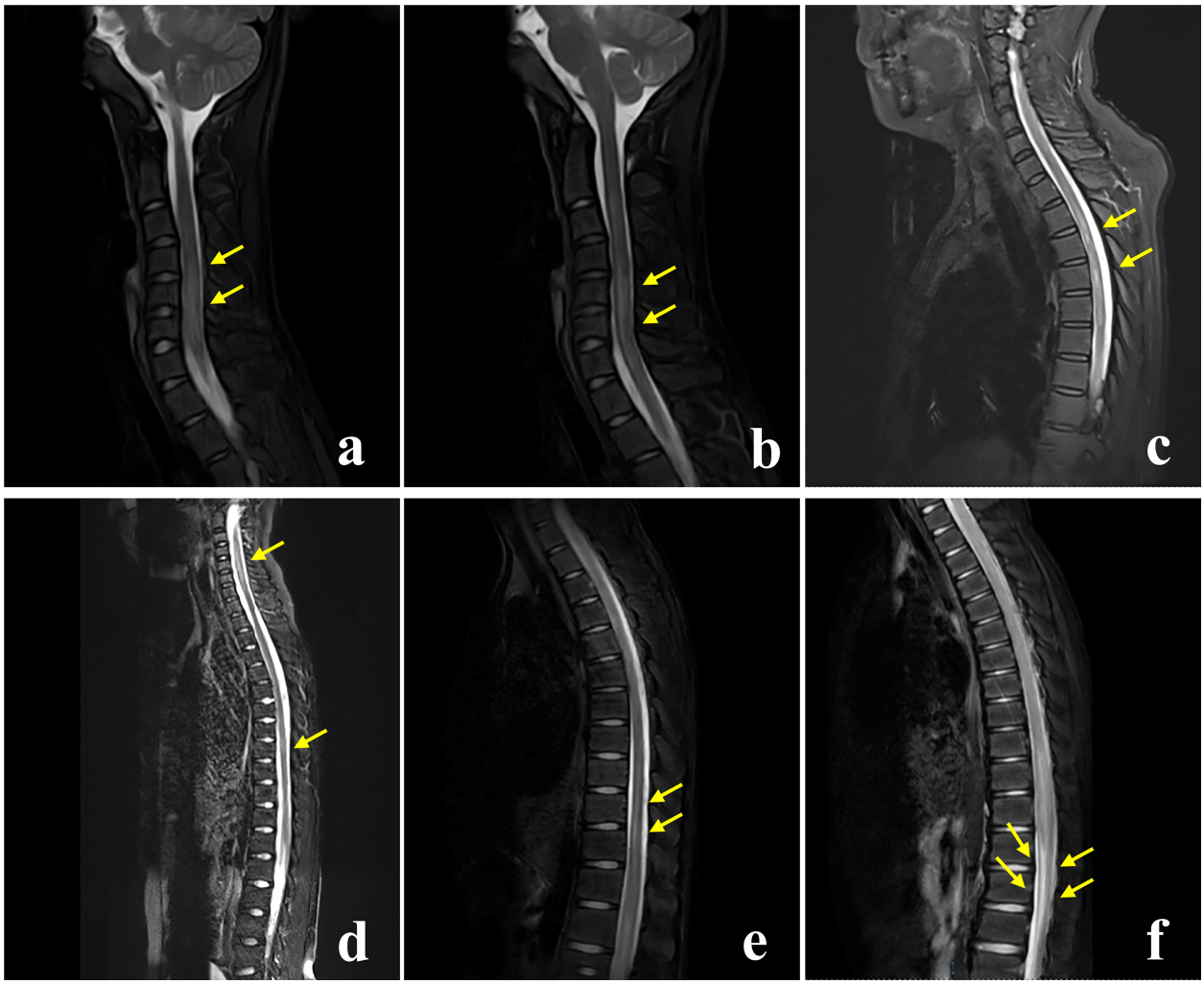


FIGURE 4
Spinal cord MRI characteristics of patients with Anti-GFAP antibodies. The MRI T2-weighted fat suppression sequence of the spinal cord in patients with anti-GFAP antibodies shows that the morphology of spinal cord lesions could be long-segment patchy lesions **(A, B)**, multiple short-segment lesions **(C)**, and long-segment linear lesions **(D, E)**. Abnormal signal of conus medullaris in 1 patient **(F)**.

like symptoms and consciousness disturbance, are common clinical manifestations. Monophasic course (81.0%) was common, whereas patients with overlapping antibodies were more likely to relapse, especially when combined with AQP4 and MOG antibodies. Four patients had symptoms of speech dysfunction, which had been rarely reported in earlier studies. Two patients developed APS, as previously reported (12), implying that APS should not only be considered as a diagnosis of neuromyelitis optica spectrum disorders (NMOSD) but should also be tested for anti-GFAP antibodies.

Some patients only with anti-GFAP antibodies in serum were included in this study because they presented with symptoms of autoimmune GFAP-A and ruled out other diagnoses. Antibody titers in serum were higher in some patients than in CSF, contradicting previous reports. A higher serum titer than CSF indicates that antibodies may have originated in the peripheral blood system. In contrast, a higher CSF than serum indicates that antibodies may have originated in the CNS via intrathecal synthesis. In our study, 40.7% of patients have CSF-restricted OCBs, which is an indicator of intrathecal synthesis. Therefore, it is reasonable to think that anti-GFAP antibodies may originate in different places. In light of these findings, we should look further into the pathogenic mechanisms and the source of anti-GFAP antibodies.

Some patients in this study had a history of herpes virus infection before the onset of neurological symptoms, whereas others had Epstein-Barr virus and herpes virus infection detected by CSF samples using NGS technology at the onset. Precursor infections are more common in children, possibly because the blood-brain barrier is not fully developed in some children. A previous study reported the first case of autoimmune GFAP-A following HSV encephalitis infection and proposed that HSV infection might activate the immune response to autoimmune GFAP-A (13). Infection appears to be associated with the pathogenesis of GFAP astrocytopathy but the neuroimmune mechanism that infection activates is unknown. One possible mechanism is that the infection damaged the astrocytes, exposing

10.3389/fimmu.2023.1106490

TABLE 3 Clinical, imaging, treatment, and prognosis of patients with GFAP overlapping antibodies.

Patient no. Sex/ age (y.)	Neural autoantibodies in serum	Neural autoantibodies in CSF	Symptoms	lesion location in MRI	Treatment	Disease course	mRS at last follow-up
14. F/12	AQP4	GFAP/AQP4	Fever, vomiting, stagger, central facial paralysis blurred vision, lower extremity weakness, involuntary movement, hearing loss, numbness and weakness of limbs	Pons, medulla oblongata, C1–5, C6–7	IVMP+IVIG +RTX	Relapse	1
15.F/14	AQP4	GFAP/AQP4	Fever, headache, blurred vision	Paraventricular third ventricle, periaqueductal gray, basal ganglia, C2–3, T4–5, T8	IVMP	Monophasic	0
32.F/35	AQP4	GFAP/AQP4	Numbness of limb, paroxysmal limb twitch	Medulla oblongata, pons, C1-C6,T2-T5	IVMP	Monophasic	1
41.F/44	AQP4	GFAP/AQP4	Paroxysmal limb twitch, paresthesias	C1-T1	IVMP+IVIG	Monophasic	1
46.F/48	AQP4	GFAP/AQP4	Fever, weakness and numbness in both lower extremities, dysuria	C2-T8 (multiple focal lesion)	IVMP+RTX	Relapse	4
52.F/55	AQP4	GFAP/AQP4	Nausea and vomiting, dysphagia, facial pain, numbness of limb	Dorsal medulla oblongata, hippocampus, basal ganglia, periventricular	IVMP	Monophasic	3
2.F/3	GFAP/MOG	GFAP/MOG	Fever, blurred vision	Bilateral frontal, parietal and temporal lobes, splenium of corpus callosum, pons, right cerebellar hemisphere, C4-T8	IVMP	Relapse	2
6.M/5	GFAP/MOG		Headache, double vision, dizziness, lethargy,	Bilateral thalamus and ganglia, bilateral parietal, temporal and occipital lobes, brainstem C5-T12	IVMP+IVIG	Monophasic	0
24.F/26	GFAP/MOG	GFAP	Fever, headache, nausea and vomiting, double vision, lower limb weakness, seizure	Bilateral cerebellar hemispheres, bilateral thalamus	IVMP	Monophasic	1
33.M/36	GFAP/MOG	GFAP	Headache, blurred vision numbness, weakness	Frontal cortex, subcortex, left thalamus, cerebral peduncle, around the fourth ventricle, bilateral cerebellar hemispheres, C1-T3	IVMP	Relapse	1
13.F/11	GFAP/NMDAR/ MOG	GFAP/NMDAR/ MOG	Lower extremity weakness, hypersomnia	C2–6, T9–12	IVMP	Monophasic	0
27.F/31	GFAP/AQP4	NMDAR/GAD65/ GlyR	Headache, dizziness, hypersomnia, disturbance of consciousness	Optic chiasma, bilateral thalamus, fornix column, and third ventricle area	IVMP+IVIG	Monophasic	1
30.M/32	Yo	GFAP	Dizziness, stagger, limb weakness	Normal	NA	Monophasic	2
36.M/41		GFAP/NMDAR	Fever, hypersomnia, delirium, lower extremity weakness, disturbance of consciousness	Leptomeninge enhancement in bilateral cerebral hemispheres and brain stem surface, T1–7	NA	Monophasic	0

F, female; M, male; C, cervical spinal cord; T, thoracic spinal cord; IVMP, intravenous methylprednisolone; IVIG, intravenous immune globulin; RTX, rituximab; mRS, modified Rankin Scale; NA, not available; MRI, magnetic resonance imaging.

10.3389/fimmu.2023.1106490

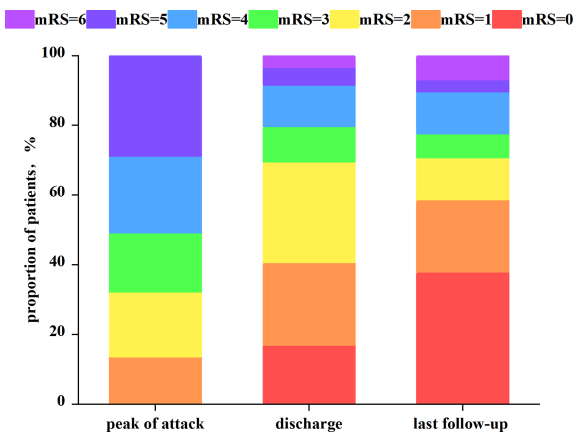


FIGURE 5
Distribution of the modified Rankin Scale score at the peak of the attack, discharge, and the last follow-up.

and releasing many GFAP antigenic determinants, leading to antibody production and secondary autoimmune responses. Another possibility is that some infectious pathogen components, such as amino acids, have a sufficiently similar structure or sequence to the host's GFAP antigen. The immune response to pathogen antigen may have an impact on the host's GFAP antigen. Therefore, we recommend testing for GFAP antibodies in patients with viral encephalitis who do not respond to antiviral therapy and checking for CSF infection status in patients with anti-GFAP antibodies. Furthermore, previous research in other countries has found that 12-38% has tumors (4, 5, 11, 14), and the occurrence of tumors may be associated with the production of GFAP antibodies. However, when compared to other studies, the incidence of tumors in our study is low. According to a Mayo Clinic study (5), 66% of tumors are detected within two years of the onset of symptoms, so the variability associated with tumors could be due to the study's small sample size and short follow-up time. On the other hand, a previous Chinese report (6) found no concomitant tumor, which is consistent with our findings. Therefore, we believe that tumor-related differences are more likely to be racial.

This study discovered that patients with anti-GFAP antibodies frequently had pathological findings in their CSF, including elevated cell counts and proteins. Hyponatremia occurs in some patients during the course of the disease, possibly due to thalamic lesion involvement. As a result, hypothalamic function is impaired, and normal mechanisms that regulate the secretion of antidiuretic hormones are disrupted. Furthermore, like those with NMOSD, these patients frequently have other systemic autoimmune diseases.

In previous studies, approximately half of GFAP antibody-positive patients had specific imaging findings of paraventricular linear radial enhancement. In contrast, in our study, only 4/40 (10%) patients had linear perivascular enhancement oriented to the ventricle, while leptomeningeal enhancement was more common (4, 5, 7, 15). Furthermore, lesions on brain MRI were mostly found in the cortex/subcortex, brainstem, thalamus, and basal ganglia. Lesions in the bilateral thalamus (20.3%) and basal ganglia (18.6%) were among the most frequent features and also matched with the findings from a Japanese study, including 14 participants (16). The cervical and thoracic spinal cords are frequently involved in spinal cord MRI lesions, but the lumbar spinal cord is rarely involved. Furthermore, LETM (34.0%) was more common in the cohort, which was consistent with previous research findings (5, 6). This study included five patients who initially presented with neurological symptoms without abnormalities on MRI but later developed radiographic lesions. Previous research have found that initial brain MRI reveals non-specific findings, but the brain MRI in reexamination and follow-up reveals characteristic autoimmune GFAP-A findings (17, 18). A case report also suggested that there was some light meningeal enhancement at first, followed by the gradual development of multiple intracranial lesions (19). These findings suggest that MRI abnormalities may delay the appearance of autoimmune GFAP-A and that MRI examinations may need to be repeated to properly diagnose this disease. Six patients with RESLES were identified on brain MRI in this study, which has previously been reported in autoimmune GFAP-A (11, 20, 21). RESLES is a rare clinical-radiographic disease with unknown pathogenesis. According to the French cohort, this unique MRI performance may support the hypothesis that GFAP autoimmunity is triggered by infection (11). In conjunction with this study, we consider RESLES to be a specific clinical imaging finding of GFAP-A, implying that patients with RESLES should also be considered for a diagnosis of GFAP autoimmune disease. Four patients in this study (three children and one adult) had clinical signs of neurological disease, but MRIs of the brain and spinal cord were normal. Previous studies have also reported on this occurrence (5, 14, 21). Previous research has suggested that normal MRI findings may be a common outcome in children, which is consistent with our findings (21). This phenomenon suggests that autoimmune GFAP-A should be considered in patients (particularly children) who have meningoencephalomyelitis-like clinical manifestations but no MRI abnormalities.

Several studies have shown that overlapping antibodies are common in autoimmune GFAP-A (5, 6, 8, 10, 22). 14/59 (23.7%) patients in our study had overlapping antibodies. NMDAR-IgG was the most common coexisting antibody in a Mayo Clinic study of 102 patients with autoimmune GFAP-A, followed by AQP4-IgG (5). Two Chinese studies discovered that AQP4-IgG and MOG-IgG were the most common coexisting antibodies (8, 22). The most common coexisting antibody in this study was AQP4-IgG, followed by MOG-IgG and NMDAR-IgG. Interestingly, our study is the first to show a specific multi-antibody overlaps: GFAP-IgG and AQP4-IgG in serum, NMDAR-IgG, GAD65-IgG, and GlyR-IgG in CSF. Although coexisting of MOG-IgG and AQP4-IgG were found in a French cohort study, simultaneous involvement of MOG-IgG in the peripheral nervous system was thought to be unusual, and AQP4-IgG was only found in CSF, casting doubt on the existence of an overlap syndrome in GFAP autoimmunity. The finding in the French cohort contradicts our findings, which is thought to be because some studies in China have found that AQP4-IgG is the most common coexisting antibody, and the detection rate of AQP4 antibodies in Asian populations is higher than in Caucasian populations. The incidence and prevalence of NMOSD vary

greatly by ethnicity and region, with Asians being particularly vulnerable. To summarize, the precise mechanism underlying the occurrence of overlapping antibodies is unknown, and how to correctly diagnose and classify patients with autoantibody overlapping syndrome is a problem that must be solved in the future. Two Mayo Clinic studies (5, 14) discovered that the presence of both GFAP-IgG and NMDAR-IgG at the same time was associated with an increased risk of tumors. However, no tumor was observed in this study when GFAP-IgG coexisted with NMDAR-IgG, which may be due to ethnic specificity, as tumors were rare in Chinese patients with anti-GFAP antibodies. At the moment, there is no clear pathogenesis for the co-occurrence of antibodies, and determining which antibodies are pathogenic is difficult. GFAP is an intracellular protein antigen, unlike AQP4, NMDAR, MOG, and other cell surface antigens, and its antibody cannot be directly contacted to produce humoral immunity. Furthermore, previous animal studies (23) have demonstrated that CD8 T cells targeting GFAP in the CNS can avoid tolerance mechanisms and cause gray and white matter lesions in the brain and spinal cord. How CNS-reactive CD8T cells are activated determines the clinical and histological characteristics of lesions. That is, spontaneously recruited GFAP-specific CD8T cells to infiltrate the CNS gray and white matter, resulting in relapse remission and chronic CNS autoimmunity. In contrast, virus-induced GFAP-specific CD8 T effector cells specifically target the meninges and vascular/perivascular spaces of gray matter and white matter, resulting in rapid, acute CNS disease. This pathogenic mechanism fits the disease course and clinical characteristics of autoimmune GFAP-A. Anti-GFAP antibodies may not be pathogenic, but they do serve as a marker of autoimmunity caused by cytotoxic T cells (4, 24).

According to a 2018 study, the immunopathological manifestations of GFAP astrocytic lesions were astrocyte and neuron loss (6). Another study, however, discovered that a patient with positive CSF GFAP antibody had no astrocyte involvement or demyelination in the autopsy and speculated that GFAP antibody was not the pathogenic antibody causing astrocyte inflammation but rather a bystander autoantibody of inflammation (25). The majority of patients in our study had other neuronal surface antibodies or viral infections, implying that GFAP antibodies might be a non-specific witness of inflammation. At the moment, the pathogenicity of GFAP autoantibodies is debatable, and more pathological evaluations are required to determine whether they are pathogenic.

The majority of patients in this study responded well to immunotherapy and were improved by the time they were discharged. Furthermore, the majority of patients had a good functional outcome, with 37.9% completely asymptomatic at the last follow-up (mRS = 0). Notably, 29.3% of patients still had poor functional outcomes (mRS > 2), including four patients (all adults) who died, indicating that immunotherapy did not work for all patients (26). Children were more likely than adults to have no residual disability at the last follow-up, implying that age may influence patient outcomes. Based on the foregoing, we can conclude that some patients have poor prognostic outcomes, and future research should look into the factors influencing poor prognosis. Furthermore, our patients were frequently diagnosed with viral encephalitis, tubercular meningitis, ADEM, and even CIS during the course of the disease, as previously reported (27–30), indicating that we should improve the relevant diagnostic criteria of GFAP autoimmune diseases and develop standardized treatment methods as soon as possible.

5 Conclusion

In conclusion, older patients were less likely to be affected, and male and female patients were roughly equally represented. Patients with anti-GFAP antibodies are often complicated with infection, autoimmunity, hyponatremia, and pathological CSF. In the meantime, patients with overlapping antibodies are common; however, the mechanism of overlapping antibodies and pathogenic antibodies is unknown. Tumors were discovered in a small number of patients. Patients frequently present with one or more of the following symptoms: encephalitis, meningitis, myelitis, and optic neuritis. Lesions on brain MRI are frequently found in the cortex/paracortex, brainstem, thalamus, and basal ganglia. One of the hallmark imaging findings of this disease may be bilateral thalamic and basal ganglia lesions. MRI lesions of the spinal cord are most commonly found in the cervical and thoracic medulla. In a small number of patients, MRI abnormalities may delay the appearance of autoimmune GFAP-A, or the MRI finding may be normal, or they may present with RESLES at the onset of autoimmune GFAP-A. Clinical manifestations and imaging findings did not differ significantly between children and adults with anti-GFAP antibodies. The majority of patients had a monophasic course, and those with overlapping antibodies were more likely to relapse. The majority of patients respond well to immunotherapy and have a good prognosis, but a few have a poor prognosis, such as death. Some patients may be misdiagnosed as having viral encephalitis, tuberculous encephalitis, ADEM, CIS, and other conditions. Children are more likely than adults to have no disability. Finally, we hypothesized that the presence of GFAP antibodies was a non-specific witness of inflammation.

6 Limitations

There are some limitations to this study. Firstly, the presented data were retrospectively obtained from an electronic medical record system. Secondly, the sample size was small, and the data were collected from a single center. Finally, selection bias might exist because the First Affiliated Hospital of Zhengzhou University is a tertiary referral center.

Patients are frequently misdiagnosed as having tuberculous meningitis, ADEM, or viral encephalitis due to a lack of knowledge about the disease, resulting in late and incorrect treatment. Therefore, we should develop early guidelines for diagnosing and treating autoimmune GFAP-A in collaboration with colleagues both at home and abroad. In addition, multicenter and large-sample clinical studies with long-term follow-up are suggested to identify factors associated with relapse

and poor prognosis in patients with anti-GPAP antibodies in the future.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The study was performed in adherence to ethical guidelines and was ethically approved by the Ethics Committee of The First Affiliated Hospital of Zhengzhou University (2022-KY-1205–002).

Author contributions

BZ and LW conceived and designed the study. BZ drafted the manuscript. MS and HY collected and analyzed the data. TY reviewed and edited the manuscript. LW revised the manuscript critically.

Acknowledgments

The authors are very grateful to the participants and their family members involved in this study.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Wang KK, Yang Z, Yue JK, Zhang Z, Winkler EA, Puccio AM, et al. Plasma anti-glial fibrillary acidic protein autoantibody levels during the acute and chronic phases of traumatic brain injury: a transforming research and clinical knowledge in traumatic brain injury pilot study. *J Neurotrauma* (2016) 33(13):1270–7. doi: 10.1089/neu.2015.3881

2. Olabarria M, Goldman JE. Disorders of astrocytes: Alexander disease as a model. *Annu Rev Pathol* (2017) 12:131–52. doi: 10.1146/annurev-pathol-052016–100218

3. Li D, Liu X, Liu T, Liu H, Tong L, Jia S, et al. Neurochemical regulation of the expression and function of glial fibrillary acidic protein in astrocytes. *Glia* (2020) 68 (5):878–97. doi: 10.1002/glia.23734

4. Fang B, McKeon A, Hinson SR, Kryzer TJ, Pittock SJ, Aksamit AJ, et al. Autoimmune glial fibrillary acidic protein astrocytopathy: a novel meningoencephalomyelitis. *JAMA Neurol* (2016) 73(11):1297–307. doi: 10.1001/jamaneurol.2016.2549

5. Flanagan EP, Hinson SR, Lennon VA, Fang B, Aksamit AJ, Morris PP, et al. Glial fibrillary acidic protein immunoglobulin G as biomarker of autoimmune astrocytopathy: analysis of 102 patients. *Ann Neurol* (2017) 81(2):298–309. doi: 10.1002/ana.24881

6. Long Y, Liang J, Xu H, Huang Q, Yang J, Gao C, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in Chinese patients: a retrospective study. *Eur J Neurol* (2018) 25(3):477–83. doi: 10.1111/ene.13531

7. Shan F, Long Y, Qiu W. Autoimmune glial fibrillary acidic protein astrocytopathy: a review of the literature. *Front Immunol* (2018) 9:2802. doi: 10.3389/fimmu.2018.02802

8. Yang X, Xu H, Ding M, Huang Q, Chen B, Yang H, et al. Overlapping autoimmune syndromes in patients with glial fibrillary acidic protein antibodies. *Front Neurol* (2018) 9:251. doi: 10.3389/fneur.2018.00251

9. Kunchok A, Zekeridou A, McKeon A. Autoimmune glial fibrillary acidic protein astrocytopathy. *Curr Opin Neurol* (2019) 32(3):452–8. doi: 10.1097/wco.0000000000000676

10. Iorio R, Damato V, Evoli A, Gessi M, Gaudino S, Di Lazzaro V, et al. Clinical and immunological characteristics of the spectrum of gfap autoimmunity: a case series of 22 patients. *J Neurol Neurosurg Psychiatry* (2018) 89(2):138–46. doi: 10.1136/jnnp-2017–316583

11. Gravier-Dumonceau A, Ameli R, Rogemond V, Ruiz A, Joubert B, Muñiz-Castrillo S, et al. Glial fibrillary acidic protein autoimmunity: a French cohort study. *Neurology* (2022) 98(6):e653–e68. doi: 10.1212/wnl.0000000000013087

12. Deng B, Wang J, Yu H, Jin L, Qiu Y, Liu X, et al. Area postrema syndrome in autoimmune glial fibrillary acidic protein astrocytopathy: a case series and literature review. *Neurol Neuroimmunol Neuroinflamm* (2022) 9(6):e200029. doi: 10.1212/nxi.0000000000200029

13. Li J, Xu Y, Ren H, Zhu Y, Peng B, Cui L. Autoimmune gfap astrocytopathy after viral encephalitis: a case report. *Mult Scler Relat Disord* (2018) 21:84–7. doi: 10.1016/j.msard.2018.02.020

14. Dubey D, Hinson SR, Jolliffe EA, Zekeridou A, Flanagan EP, Pittock SJ, et al. Autoimmune gfap astrocytopathy: prospective evaluation of 90 patients in 1 Year. *J Neuroimmunol* (2018) 321:157–63. doi: 10.1016/j.jneuroim.2018.04.016

15. Xiao J, Chen X, Shang K, Tang Y, Chen M, Deng G, et al. Clinical, neuroradiological, diagnostic and prognostic profile of autoimmune glial fibrillary acidic protein astrocytopathy: a pooled analysis of 324 cases from published data and a single-center retrospective study. *J Neuroimmunol* (2021) 360:577718. doi: 10.1016/j.jneuroim.2021.577718

16. Kimura A, Takekoshi A, Yoshikura N, Hayashi Y, Shimohata T. Clinical characteristics of autoimmune gfap astrocytopathy. *J Neuroimmunol* (2019) 332:91–8. doi: 10.1016/j.jneuroim.2019.04.004

17. Natori T, Fukao T, Watanabe T, Kurita T, Hata T, Kimura A, et al. Repeated brain magnetic resonance imaging provides clues for the diagnosis of autoimmune glial fibrillary acid protein astrocytopathy. *Intern Med* (2022) 61(19):2947–50. doi: 10.2169/internalmedicine.8964–21

18. Izumi M, Uzawa A, Aoki R, Suzuki M, Yoshizawa K, Suzuki Y, et al. Delayed appearance of brain magnetic resonance imaging abnormalities in a patient with glial fibrillary acidic protein astrocytopathy. *Intern Med* (2022) 62(3):465-8. doi: 10.2169/internalmedicine.9724–22

19. Imanaka S, Oka Y, Kimura A, Shimohata T, Matsumoto S. Autoimmune glial fibrillary acidic protein astrocytopathy with delayed abnormal magnetic resonance imaging findings. *eNeurologicalSci* (2022) 27:100403. doi: 10.1016/j.ensci.2022.100403

20. Héraud C, Capet N, Levraut M, Hattenberger R, Bourg V, Thomas P, et al. Glial fibrillary acidic protein (Gfap) astrocytopathy presenting as mild encephalopathy with reversible splenium lesion. *Neurol Ther* (2022) 11(1):499–505. doi: 10.1007/s40120–021–00302-y

21. Oger V, Bost C, Salah L, Yazbeck E, Maurey H, Bellesme C, et al. Mild Encephalitis/Encephalopathy with reversible splenial lesion syndrome: an unusual presentation of anti-gfap astrocytopathy. *Eur J Paediatr Neurol* (2020) 26:89–91. doi: 10.1016/j.ejpn.2020.03.002

22. Fang H, Hu W, Jiang Z, Yang H, Liao H, Yang L, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in children: a retrospective analysis of 35 cases. *Front Immunol* (2021) 12:761354. doi: 10.3389/fimmu.2021.761354

23. Sasaki K, Bean A, Shah S, Schutten E, Huseby PG, Peters B, et al. Relapsing-remitting central nervous system autoimmunity mediated by gfap-specific Cd8 T cells. *J Immunol* (2014) 192(7):3029–42. doi: 10.4049/jimmunol.1302911

24. Yuan Z, Li H, Huang L, Fu C, Chen Y, Zhi C, et al. Cd8(+) T-cell predominance in autoimmune glial fibrillary acidic protein astrocytopathy. *Eur J Neurol* (2021) 28 (6):2121–5. doi: 10.1111/ene.14778

25. Yamakawa M, Hogan KO, Leever J, Jassam YN. Autopsy case of meningoencephalomyelitis associated with glial fibrillary acidic protein antibody. *Neurol Neuroimmunol Neuroinflamm* (2021) 8(6):e1081. doi: 10.1212/nxi.0000000000001081

26. Yang X, Liang J, Huang Q, Xu H, Gao C, Long Y, et al. Treatment of autoimmune glial fibrillary acidic protein astrocytopathy: follow-up in 7 cases. *Neuroimmunomodulation* (2017) 24(2):113–9. doi: 10.1159/000479948

27. Yang X, Zhang C, Zhang J, Chen G, Zhao L, Yang P, et al. Autoimmune glial fibrillary acidic protein astrocytopathy mimics infectious meningitis: two case reports. *Mult Scler Relat Disord* (2020) 45:102350. doi: 10.1016/j.msard.2020.102350

28. Li J, Wang C, Cao Y, Shi J, Liu H, Zhou M, et al. Autoimmune glial fibrillary acidic protein astrocytopathy mimicking acute disseminated encephalomyelitis: a case report. *Med (Baltimore)* (2021) 100(25):e26448. doi: 10.1097/md.0000000000026448

29. Quek AM, Tang D, Chin A, Ng KW, Lin H, Seet RC. Autoimmune glial fibrillary acidic protein astrocytopathy masquerading as tuberculosis of the central nervous system: a case series. *Int J Infect Dis* (2022) 124:164–7. doi: 10.1016/j.ijid.2022.09.029

30. Sakashita Y, Nozaki I, Hamaguchi T, Kimura A, Shimohata T, Ono K. A case of autoimmune glial fibrillary acidic protein astrocytopathy presenting with magnetic resonance imaging mimics of multiple sclerosis. *Clin Neurol Neurosurg* (2022) 218:107272. doi: 10.1016/j.clineuro.2022.107272

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 07 June 2023
DOI 10.3389/fimmu.2023.1136955

OPEN ACCESS

EDITED BY
Oivind Torkildsen,
University of Bergen, Norway

REVIEWED BY
Maria Eleftheria Evangelopoulos,
National and Kapodistrian University of Athens, Greece
Nicolas Collongues,
Hôpitaux Universitaires de Strasbourg, France

*CORRESPONDENCE
Nanchang Xie
xienanchang2001@163.com

†These authors share first authorship

RECEIVED 03 January 2023
ACCEPTED 23 May 2023
PUBLISHED 07 June 2023

CITATION
Zhang W, Xie Y, Wang Y, Liu F, Wang L, Lian Y, Liu H, Wang C and Xie N (2023) Clinical characteristics and prognostic factors for short-term outcomes of autoimmune glial fibrillary acidic protein astrocytopathy: a retrospective analysis of 33 patients.
Front. Immunol. 14:1136955.
doi: 10.3389/fimmu.2023.1136955

Clinical characteristics and prognostic factors for short-term outcomes of autoimmune glial fibrillary acidic protein astrocytopathy: a retrospective analysis of 33 patients

Wanwan Zhang[1†], Yinyin Xie[1†], Yali Wang[2†], Fengxia Liu[1], Li Wang[2], Yajun Lian[1], Hongbo Liu[1], Cui Wang[3] and Nanchang Xie[1*]

[1]Department of Neurology, the First Affiliated Hospital of Zhengzhou University, Zhengzhou, China, [2]Department of Neurology, Henan Children's Hospital, Zhengzhou, China, [3]Department of Clinical Laboratory, the First Affiliated Hospital of Zhengzhou University, Zhengzhou, China

Background: Autoimmune glial fibrillary acidic protein astrocytopathy (GFAP-A) is a recently discovered inflammatory central nervous system (CNS) disease, whose clinical characteristics and prognostic factors for short-term outcomes have not been defined yet. We aimed to assess the symptoms, laboratory tests, imaging findings, treatment, and short-term prognosis of GFAP-A.

Methods: A double-center retrospective cohort study was performed between May 2018 and July 2022. The clinical characteristics and prognostic factors for short-term outcomes were determined.

Results: We enrolled 33 patients with a median age of 28 years (range: 2–68 years), 15 of whom were children (<18 years). The clinical spectrum is dominated by meningoencephalomyelitis. Besides, we also found nausea, vomiting, poor appetite, and neuropathic pain in some GFAP-A patients, which were not mentioned in previous reports. And adults were more prone to limb numbness than children. Magnetic resonance imaging revealed lesions involving the brain parenchyma, meninges, and spinal cord, exhibiting patchy, linear, punctate, and strip T2 hyperintensities. First-line immunotherapy, including corticosteroid and gamma globulin, was effective in most patients in the acute phase (P = 0.02). However, patients with overlapping AQP4 antibodies did not respond well to first-line immunotherapy and coexisting neural autoantibodies were more common in women. Additionally, the short-term prognosis was significantly better in children than in adults (P = 0.04). Positive non-neural autoantibodies and proven viral infection were independent factors associated with poor outcomes (P = 0.03, 0.02, respectively).

Conclusion: We expanded the spectrum of clinical symptoms of autoimmune GFAP-A. The clinical symptoms and short-term prognosis differed between children and adults. Positive non-neural autoantibodies and proven viral infection at admission suggest a poor short-term prognosis.

KEYWORDS

glial fibrillary acidic protein, autoimmune, inflammatory CNS disease, Short-term prognosis, clinical characteristics

1 Introduction

Glial fibrillary acidic protein (GFAP) is an intermediate filament protein that is mainly found in the astrocytic cytoplasm, and is involved in numerous astrocyte functions. The size of GFAP lies between that of microfilaments and microtubules (1). Tissue-based assays and cell-based assays (CBA) can be used to identify the immunoglobulin G (IgG) reactive with GFAP in the cerebrospinal fluid (CSF) or serum of patients with autoimmune GFAP astrocytopathy (GFAP-A), which is a novel inflammatory central nervous system (CNS) disease reported in 2016 (2, 3). Patients usually present with meningitis (headache and neck stiffness), encephalitis (psychiatric symptoms, seizures, tremor, or delirium), myelitis (weakness and sensory symptoms), optic neuritis (blurred vision), or a combination of the above (4). The characteristic imaging feature is perivascular radial enhancement perpendicular to the ventricles, which resolves with immunotherapy (5). Coexisting neural autoantibodies are common in autoimmune GFAP-A, which makes diagnosis difficult (6). Most patients respond well to first-line immunotherapy, including corticosteroids, intravenous immunoglobulin, and plasma exchange, alone or in combination, but some are prone to relapse or death (7).

Since autoimmune GFAP-A is a recent discovery, the complete range of clinical and imaging phenotypes is still unknown. Although several GFAP-A case series have been reported, to our knowledge, no study has identified the prognostic factors for short-term outcomes in GFAP-A. Therefore, we included 33 GFAP-A patients from two hospitals in China, and retrospectively analyzed the clinical manifestations, magnetic resonance imaging (MRI) findings, laboratory examination results, treatment, and short-term prognosis. This study aims to provide new insights and improve the clinicians' understanding of autoimmune GFAP-A.

Abbreviations: GFAP-A, glial fibrillary acidic protein astrocytopathy; CBA, cell-based assay; CNS, central nervous system; MRI, Magnetic resonance imaging; GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; CSF, cerebrospinal fluid; IVMP, intravenous methylprednisolone; IVIG, intravenous immunoglobulin; PE, plasma exchange; RIT, rituximab; TAC, tacrolimus; MMF, mycophenolate mofetil; mRS, Modified Rankin Scale; NMDAR, N-methyl-D-aspartate receptor; AQP-4, aquaporin-4; MOG, myelin oligodendrocyte glycoprotein; Yo, Purkinje cell type 1.

2 Methods

2.1 Study design and participants

In this double-center retrospective observational cohort study, we enrolled patients who presented with meningitis, encephalitis, and myelitis, and tested positive for GFAP antibodies in the CSF between May 1, 2018 and April 1, 2022 at the First Affiliated Hospital of Zhengzhou University and Henan Children's Hospital. This study was approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University (number 2022-KY-0053) and all patients provided their informed consent.

Demographic data, clinical manifestations, CSF examination, serological tests, imaging findings, intensive care unit admission, mechanical ventilation, treatment, and outcomes were recorded. CSF examination included white cell counts, protein level, glucose level, virus antibodies detection, and oligoclonal antibodies (IgG), which were assessed in all patients with GFAP-A. Neutrophilic granulocyte, monocyte, lymphocyte, blood sodium, non-neural autoantibodies, tumor markers, and virus antibodies detection were comprised in the serological tests. Tumor markers comprised ferritin, neuron-specific enolase, alpha-fetoprotein, carcino-embryonic antigen, tumor associated antigen 125, 19-9, 15-3, and 72-4, and non-small cell lung cancer antigen 21-1. Non-neural autoantibodies contained antinuclear, anti-endothelial cell, anti-cardiolipin, anti-neutrophil cytoplasmic, anti-double-stranded DNA, anti-RA33, rheumatoid factor, anti-PM-Scl antibody, anti-SSA, and anti-Ro52 antibodies. Virus antibodies detection in CSF and serum included Epstein-Barr virus, cytomegalovirus, coxsackie virus, measles virus, herpes simplex virus I and II, human parvovirus B-19, influenza b virus, parainfluenza virus, adenovirus, rubella virus, herpes zoster virus, and echovirus. All patients underwent the above serological tests except for one patient who did not undergo virus screenings in serum. Patients exhibiting symptoms such as fever, cough, fatigue, nausea, or vomiting that cannot be explained by other causes and who were positive for the virus antibodies (immunoglobulin M) in serum, were defined as proven viral infection. If they only showed symptoms associated with viral infection without laboratory evidence, patients were defined as suspected viral infection. First-line immunotherapy included intravenous methylprednisolone (IVMP), intravenous immunoglobulin (IVIG), or plasma exchange (PE). Second-line immunotherapy included rituximab (RIT), tacrolimus (TAC), and

mycophenolate mofetil (MMF). The Modified Rankin Scale (mRS) was used to evaluate the neurological status at admission, 4 weeks after the initiation of immunotherapy, and last follow-up. A favorable outcome was defined as an mRS score of <3, while a poor outcome was defined as an mRS score of ≥3. If the patient died, the mRS score was recorded as 6. All patients were followed up by telephone or in the outpatient clinic, and the last date for follow-up was July 1, 2022. We defined patients younger than 18 years as children. Relapse was defined as hospital readmission for meningoencephalomyelitis.

2.2 Antibody assay

The CSF and serum samples of the patients were simultaneously obtained before treatment and sent to the Neurology Laboratory of the First Affiliated Hospital of Zhengzhou University or Zhengzhou Jinyu Clinical Laboratory Center. Both institutes used fixed CBA for confirmation, with 100% agreement among positive results.

The CBA method used human embryonic kidney 293 cells transfected with plasmids (pc DNA3.1) encoding GFAP homo sapiens transcript variant (NM_002055) (Shanghai Genechem Co.,Ltd) using Lipofectamine 2000. 36 hours after transfection, cells were fixed with 4% paraformaldehyde for 20 min and permeabilized with PBS containing 0.25% Triton-100 for 30 min at room temperature. Cells were incubated for 30 min at room temperature (serum diluted at 1:10, and CSF 1:1). The fluid in the wells was removed and washed 3 times with PBS afterward. AlexaFluor 546 anti-human IgG (1:500; Thermo Scientific) was used as the secondary antibody to label autoantibodies for 1 h at room temperature. Images were obtained using a Zeiss Axiovert A1 fluorescence microscope (Figure 1).

In addition, autoantibodies to aquaporin-4 (AQP-4), myelin oligodendrocyte glycoprotein (MOG), myelin basic protein, N-methyl-D-aspartate receptor (NMDAR), glycine receptor, glutamic acid decarboxylase 65, γ–amino butyric acid type A receptor and B receptor, α-amino-3-hydroxy-5-methyl-4-isoxazol-propionic acid receptors 1 and 2, contactin-associated protein 2, leucine-rich glioma-inactivated protein 1, Purkinje cell type 1 (Yo), antineuronal nuclear antibodies type 1 and 2, were detected with fixed CBA to examine possible coexisting autoimmunity.

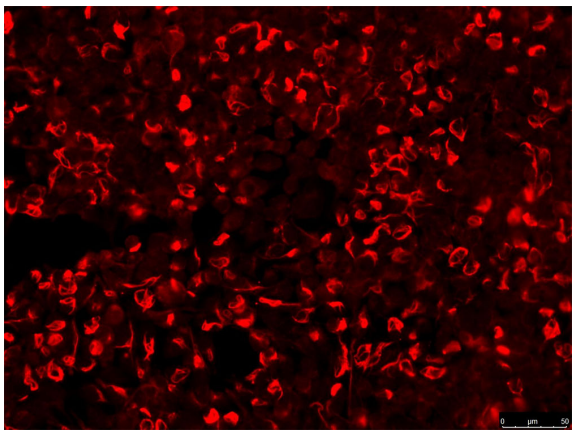


FIGURE 1
IgG in serum of patient (no.5, see Table 1). Those stained in red alone are GFAP antibodies. Images were obtained using a Zeiss Axiovert A1 fluorescence microscope.

2.3 Statistical analysis

Statistical analysis was performed using SPSS IBM 25.0 (SPSS Inc., Chicago, IL, USA). We used the fisher exact test to analyze the difference of short-term prognosis and abnormal spine cord MRIs between children and adults. The rank sum test was used for the effectiveness of first-line immunotherapy. The correlation analysis of the disease severity on admission or short-term prognosis with GFAP antibody titers and the number of symptoms was examined by Spearman's rank correlation. Univariable binary logistic regression models were used to assess the factors affecting the outcome, and factors associated with a poor outcome ($P < 0.1$) were included in the multivariate binary logistic regression model. All statistical tests were two-sided, and $P < 0.05$ was considered statistically significant in the multivariate binary logistic regression analysis.

3 Results

Overall, we identified 33 patients with positive GFAP antibodies in the CSF from May 2018 to April 2022. Of these, 25 patients were from the First Affiliated Hospital of Zhengzhou University, and 8 pediatric patients were from Henan Children's Hospital. The demographics and clinical characteristics of the patients with autoimmune GFAP-A are summarized in Table 1.

3.1 Demographic data and clinical manifestations

The median age at disease onset was 28 years (range: 2–68 years), and 15 patients were children with a median age of 8 years (range: 2–14 years). There were 17 males and 16 females in the cohort. Of these, 16 cases were positive for GFAP-IgG in both the CSF and serum, while 17 cases were positive for GFAP-IgG only in the CSF. GFAP antibody titers in the serum and CSF were not significantly associated with the disease severity on admission or short-term prognosis ($P = 0.17$, $r_s = 0.26$; $P = 0.60$, $r_s = -0.10$; $P = 0.20$, $r_s = -0.23$; $P = 0.15$, $r_s = -0.26$; respectively).

A total of 29 patients had three or more symptoms at admission. However, there was no statistical correlation between the number of symptoms and disease severity on admission or short-term prognosis ($P = 0.05$, $r_s = 0.34$; $P = 0.45$, $r_s = 0.14$ respectively). Seventeen of the thirty-three patients had prodromal symptoms, including fever (n = 15), vomiting (n = 2). The main clinical symptoms were fever (21 cases); limb weakness (13 cases); vomiting (13 cases); headache (12 cases); nausea (11 cases); disturbance of consciousness (10 cases); dysuria and constipation (9 cases); poor appetite (9 cases); involuntary movements, including

10.3389/fimmu.2023.1136955

TABLE 1 Clinical features, auxiliary examinations, treatment strategies, and short-term prognosis in patients positive for GFAP-IgG.

Patient no. sex/age of onset	Summary clinical symptoms	MRI findings	Antibody titer		CSF White Blood Cell Count,/L; Protein, g/L; Glucose, mmol/L	Therapy	mRS at admission/4 weeks after the initiation of immunotherapy	ICU admission
			Serum antibody	CSF antibody				
1.M/2	Fever, nausea, constipation	**Brain:** T2-hyperintense lesions in bilateral frontal and left parietal **Spine:** NA	GFAP-IgG (1:100)	GFAP-IgG (1:32)	55; 273.7; 3	IVMP, acyclovir	1/0	Yes
2.F/3	blurred vision	**Brain:** T2-hyperintense lesions in bilateral frontal, parietal and temporal, corpus callosum, pons, right cerebellum, optic nerve **Spine:** lesions in C4-T8	GFAP-IgG (1:32) MOG-IgG (1:100)	GFAP-IgG (1:32) MOG-IgG (1:10)	22; 407; 2.79	IVMP, penciclovir	3/0	No
3.M/3	Fever, nausea, vomiting, lethargy, poor appetite, limb weakness	**Brain:** T2-hyperintense lesions in bilateral cerebral hemisphere, mesencephalon and cerebellum, corpus callosum, right basal ganglia, cerebellar **Spine:** normal	GFAP-IgG (1:100)	GFAP-IgG (1:32)	1;211; 3.43	IVMP, IVIG, penciclovir	5/4	Yes
4.M/7	Fever, dizziness, vomiting, abdominal pain, abdominal distension, epilepsy, consciousness disturbance, limb weakness, dysuria, constipation	**Brain:** T2-hyperintense lesions in cerebellum, bilateral basal ganglia and thalamus, left frontal and parietal **Spine:** lesions in C2-T11	GFAP-IgG (1:32)	GFAP-IgG (1:32)	138; 751.1; 3.36	IVMP, IVIG, acyclovir	5/3	No
5.M/7	vomiting, poor appetite, limb pain, fever, abdominal pain, lethargy	**Brain:** T2-hyperintense lesions in bilateral frontal, left basal ganglia and parietal **Spine:** normal	GFAP-IgG (1:100)	GFAP-IgG (1:32)	31; 381.6; 3.83	IVMP, IVIG, acyclovir	2/4	Yes
6.M/7	Dizziness, headache, fever, abdominal pain, nausea, vomiting, psychosis	**Brain:** T2-hyperintense lesions in left frontal, bilateral lateral ventricles **Spine:** lesions in T10-S2	Antibody (-)	GFAP-IgG (1:3.2)	98; 1074.0; 2.66	IVMP, IVIG, penciclovir	3/1	No
7.M/8	Fever, headache, vomiting, poor appetite	**Brain:** T2-hyperintense lesions and enhancement in meninges **Spine:** lesions and enhancement in intermittent spinal cord segments below C5	Antibody (-)	GFAP-IgG (1:100)	165; 583.4; 2.26	IVMP	2/0	No

(Continued)

10.3389/fimmu.2023.1136955

TABLE 1 Continued

Patient no. sex/age of onset	Summary clinical symptoms	MRI findings	Antibody titer		CSF White Blood Cell Count,/L; Protein, g/L; Glucose, mmol/L	Therapy	mRS at admission/4 weeks after the initiation of immunotherapy	ICU admission
			Serum antibody	CSF antibody				
8.F/8	Headache, nausea, limb weakness	**Brain:** T2-hyperintense lesions in bilateral frontal and lateral ventricle, left thalamus **Spine:** normal	GFAP-IgG (1:32)	GFAP-IgG (1:32)	94; 844.0; 3.29	IVMP, acyclovir	3/0	No
9.F/10	Lethargy, epilepsy, fever, limb weakness, coughing when drinking water, blurred vision	**Brain:** T2-hyperintense lesions in bilateral cerebral peduncle, parietal, and temporal, pons, medulla oblongata, left frontal, occipital, basal ganglia and thalamus. **Spine:** lesions in C3-T12	GFAP-IgG (1:32)	GFAP-IgG (1:32)	4; 273.0; 3.00	IVMP, IVIG	4/0	No
10.F/11	Fever, cough	**Brain:** normal **Spine:** T2-hyperintense lesions in T8-L1	Antibody (-)	GFAP-IgG (1:100)	156; 664.4; 2.10	IVMP	1/0	No
11.F/11	Limb weakness, lethargy, poor appetite	**Brain:** normal **Spine:** T2-hyperintense lesions in C2-6 and T9-12	GFAP-IgG (1:32) NMDAR-IgG (1:10) MOG-IgG (1:100)	GFAP-IgG (1:100) NMDAR-IgG (1:10) MOG-IgG (1:100)	18; 262.0; 2.95	IVMP, IVIG, PE, acyclovir	5/0	Yes
12.M/12	Cough, fever, poor appetite	**Brain:** T2-hyperintense lesions and enhancement in meninges **Spine:** normal	GFAP-IgG (1:32)	GFAP-IgG (1:32)	128; 1097.6; 1.00	IVMP, IVIG, acyclovir, voriconazole, amphotericin B, ceftriaxone, rifamycin	3/0	No
13.F/12	Vomiting, intermittent blurred vision, walk unsteadiness	**Brain:** normal **Spine:** T2-hyperintense lesions in medulla oblongata and C1-C5	AQP4-IgG (1:320)	GFAP-IgG (1:32) AQP4-IgG (1:100)	31; 443.0; 3.48	IVMP, IVIG, penciclovir	3/3	No
14.M/14	Fever, cough, headache, dysuria, lethargy, vomiting, limb-shaking, limb weakness,	**Brain:** T2-hyperintense lesions in right lateral ventricle, corpus callosum **Spine:** lesions in C3-7, thoracic	Antibody (-)	GFAP-IgG (1:32)	304; 4433.0; 2.31	IVMP, IVIG, penciclovir	5/0	Yes
15.F/14	Fever, headache, vomiting, limb weakness, dysuria, constipation	**Brain:** T2-hyperintense lesions in meninges, bilateral basal ganglia and thalamus **Spine:** lesions in C4-T1 and T7-L1 Enhancement in meninge and C3-7	GFAP-IgG (1:100)	GFAP-IgG (1:32)	107; 1243.9; 3.00	IVMP, acyclovir	5/0	Yes

(Continued)

10.3389/fimmu.2023.1136955

TABLE 1 Continued

Patient no. sex/age of onset	Summary clinical symptoms	MRI findings	Antibody titer		CSF White Blood Cell Count,/L; Protein, g/L; Glucose, mmol/L	Therapy	mRS at admission/4 weeks after the initiation of immunotherapy	ICU admission
			Serum antibody	CSF antibody				
16.F/26	Fever, headache, nausea, vomiting, dysuria, constipation, blurred vision, limb weakness, epilepsy	**Brain:** T2-hyperintense lesions in bilateral cerebellum and thalamus **Spine:** normal	GFAP-IgG (1:32) MOG-IgG (1:10)	GFAP-IgG (1:100)	38; 359.5; 2.04	IVMP, MMF, penciclovir	3/0	No
17.F/28	headache, nausea, vomiting, fever	**Brain:** T2-hyperintense lesions and enhancement in meninges **Spine:** lesions and enhancement in C2-T12	Antibody (-)	GFAP-IgG (1:32)	950; 1837.9; 1.14	IVMP, IVIG, penciclovir	3/3	No
18.F/31	Headache, dizziness, lethargy, psychosis	**Brain:** T2-hyperintense lesions in bilateral frontal and lateral ventricles Enhancement in the optic chiasm, bilateral anterior portion of the thalamus, fornix column and triventricular area **Spine:** normal	Antibody (-)	GFAP-IgG (1:1) NMDAR-IgG (1:3.2) glutamic acid decarboxylase 65-IgG (1:3.2) glycine receptor -IgG (1:1) AQP4-IgG (1:3.2)	30; 296.7; 3.55	IVMP, IVIG	4/5	No
19.M/32	Fever, headache, tics, psychosis, dysuria	**Brain:** T2-hyperintense lesions in bilateral frontal, basal ganglia, lateral ventricle, cerebellum and thalamus, left hippocamp, pons, medulla oblongata us **Spine:** normal	GFAP-IgG (1:100)	GFAP-IgG (1:3.2)	207; 2402.0; 2.51	IVMP, MMF, penciclovir	5/5	Yes
20.M/32	Dizziness, limb weakness, poor appetite	**Brain:** T2-hyperintense lesions in right frontal **PET-CT:** cervicothoracic spinal cord segmental metabolic activation	YO- IgG (1:10)	GFAP-IgG (1:32) YO- IgG (1:10)	32; 656.4; 3.53	IVMP, etoricoxib	3/4	No
21.F/35	Limb numbness, tics	**Brain:** NA **Spine:** T2-hyperintense lesions in medulla oblongata, C1-6 and T3-5	AQP4-IgG (1:100)	GFAP-IgG (1:32) AQP4-IgG (1:32)	2; 202.7; 2.45	IVMP, tacrolimus	3/3	No
22.M/41	Fever, cough, nausea, poor appetite	**Brain:** T2-hyperintense lesions and enhancement in meninges **Spine:** lesions in T10-S2	Antibody (-)	GFAP-IgG (1:100)	260; 664.1; 1.96	IVMP, ganciclovir	1/5	Yes

(Continued)

10.3389/fimmu.2023.1136955

TABLE 1 Continued

Patient no. sex/age of onset	Summary clinical symptoms	MRI findings	Antibody titer		CSF White Blood Cell Count,/L; Protein, g/L; Glucose, mmol/L	Therapy	mRS at admission/4 weeks after the initiation of immunotherapy	ICU admission
			Serum antibody	CSF antibody				
23.M/42	Limb weakness, consciousness disturbance	**Brain:** T2-hyperintense lesions and enhancement in bilateral basal ganglia, lateral ventricle and thalamus, corpus callosum, insular, temporal, occipital, and left hippocampus **Spine:** normal	NA	GFAP-IgG (1:32)	54; 755.4; 4.01	IVMP, ganciclovir, vidarabine	3/3	No
24.M/44	Fever, psychosis, headache, tics, limb weakness, consciousness disturbance, dysuria	**Brain:** T2-hyperintense lesions in bilateral basal ganglia, lateral ventricle, cerebellum, thalamus, hippocampus and cerebral peduncle, pons, mesencephalon, medulla oblongata Enhancement in pons and medulla oblongata **Spine:** lesions and enhancement in C1-C2	GFAP-IgG (1:10)	GFAP-IgG (1:100)	9; 273.0; 7.71	IVMP, IVIG, penciclovir	5/5	Yes
25.F/45	mouth numbness, forehead pain, intercostal pain	**Brain:** T2-hyperintense lesions in bilateral frontal, pons Enhancement in pons **Spine:** normal	NA	GFAP-IgG (1:32)	2; 290.4; 2.31	ganciclovir	1/1	No
26.F/45	blurred vision, limb numbness, facial pain, dysuria, constipation, fever	**Brain:** T2-hyperintense lesions in bilateral frontal, left parietal **Spine:** lesions and enhancement in C2-3 and T5	Antibody (-)	GFAP-IgG (1:32)	6; 401.9; 2.94	IVMP	3/2	Yes
27.M/47	Fever, dizziness, chest tightness, alalia, deviated mouth	**Brain:** normal **Spine:** NA	GFAP-IgG (1:32)	GFAP-IgG (1:100)	108; 954.5; 1.86	IVMP, IVIG, ganciclovir	3/6	Yes
28.M/48	Limb numbness, dysuria, constipation, limb pain	**Brain:** T2-hyperintense lesions in bilateral frontal **Spine:** lesions in C4-T1	Antibody (-)	GFAP-IgG (1:32)	2; 734.7; 4.04	IVMP, penciclovir	3/1	No
29.F/51	Fever, headache, nausea, vomiting, poor appetite, consciousness disturbance, limb-shaking, psychosis,	**Brain:** T2-hyperintense lesions in bilateral cerebral hemisphere, basal ganglia and thalamus, cerebellum, meninges Enhancement in	GFAP-IgG (1:32)	GFAP-IgG (1:32)	108; 572.0; 2.94	IVMP, IVIG, RIT, acyclovir	3/3	Yes

(Continued)

TABLE 1 Continued

Patient no. sex/age of onset	Summary clinical symptoms	MRI findings	Antibody titer		CSF White Blood Cell Count,/L; Protein, g/L; Glucose, mmol/L	Therapy	mRS at admission/4 weeks after the initiation of immunotherapy	ICU admission
			Serum antibody	CSF antibody				
	abdominal distension	meninges **Spine:** lesions in C4						
30.F/55	Nausea, vomiting, facial pain, alalia, dysphagia, coughing when drinking water, limb numbness	**Brain:** T2-hyperintense lesions in bilateral frontal, lateral ventricle and parietal **Spine:** normal	AQP4-IgG (1:10)	GFAP-IgG (1:32) AQP4-IgG (1:32)	24; 368.1; 3.05	IVMP, ganciclovir	3/3	No
31.M/57	Hiccup, nausea, vomiting, chest tightness, belching, facial pain, limb weakness, limb numbness	**Brain:** T2-hyperintense lesions and enhancement in right basal ganglia and lateral ventricle, medulla oblongata **Spine:** normal	Antibody (-)	GFAP-IgG (1:32)	70; 452.6; 3.38	IVMP, ganciclovir	3/1	No
32.M/59	Fever, headache, limb-shaking, poor appetite, nausea, psychosis	**Brain:** T2-hyperintense lesions in bilateral basal ganglia, lateral ventricle, cerebellum and thalamus, left frontal **Spine:** NA	GFAP-IgG (1:32)	GFAP-IgG (1:100)	30; 759.4; 2.67	IVIG, penciclovir	1/0	No
33.F/68	Fever, limb weakness, limb numbness, alalia, limb-shaking	**Brain:** T2-hyperintense lesions in bilateral basal ganglia, lateral ventricle, cerebellum, frontal and parietal, pons Enhancement in bilateral basal ganglia, lateral ventricle and cerebellum, pons **Spine:** normal	NA	GFAP-IgG (1:32)	2; 570.8; 3.84	IVMP, penciclovir	4/4	No

tics and limb-shaking (7 cases); psychosis (6 cases); limb numbness (6 cases); neuropathic pain involving the face, limbs, and intercostal region (6 cases). Thirteen patients had hyponatremia (<135 mmol/L); four patients presented with blurred vision; and three patients had focal epilepsy. Ten patients showed neck stiffness on physical examination. Patient #27 died of acute brainstem failure during hospitalization, and patient #29 died of severe pneumonia 3 months after discharge. Figure 2A compared the symptoms in children and adults. And only adult patients presented with limb numbness in our cohort.

3.2 Laboratory examination

Thirteen patients had abnormal tumor markers in the serum, mainly ferritin (5/13) and neuron-specific enolase (5/13). However, none of the patients were diagnosed with a tumor as of July 2022. Viral antibodies (immunoglobulin M) were detected in the serum of 19 patients, which primarily comprised antibodies to Epstein-Barr virus (12/19), cytomegalovirus (11/19), and coxsackie virus (7/19). Ten patients had non-neural autoantibodies in the serum, including anti-SSA antibody (6/10), antinuclear antibody (5/10), rheumatoid factor (1/10), and anti-PM-Scl antibody (1/10) (Table 2).

CSF abnormalities were found in 30 patients. Pleocytosis was found in 27 patients (mainly lymphocytes), with the highest number being 950 × 10^6/L (reference range, 0–5 × 10^6/L). There were 19 cases with elevated proteins up to 4433 mg/L (reference range, 150–450 mg/L). Furthermore, 10 patients showed hypoglycorrhachia, with the minimum value being 1 mmol/L (reference range, 2.5–4.5 mmol/L). Viruses were detected in the CSF of five patients, namely Epstein-Barr virus (3/5), enterovirus (1/5), and herpes simplex virus I (1/5). *Aspergillus fumigatus* was detected in the CSF of patient #12. Oligoclonal antibodies (IgG) were identified in the CSF of four patients (Table 3).

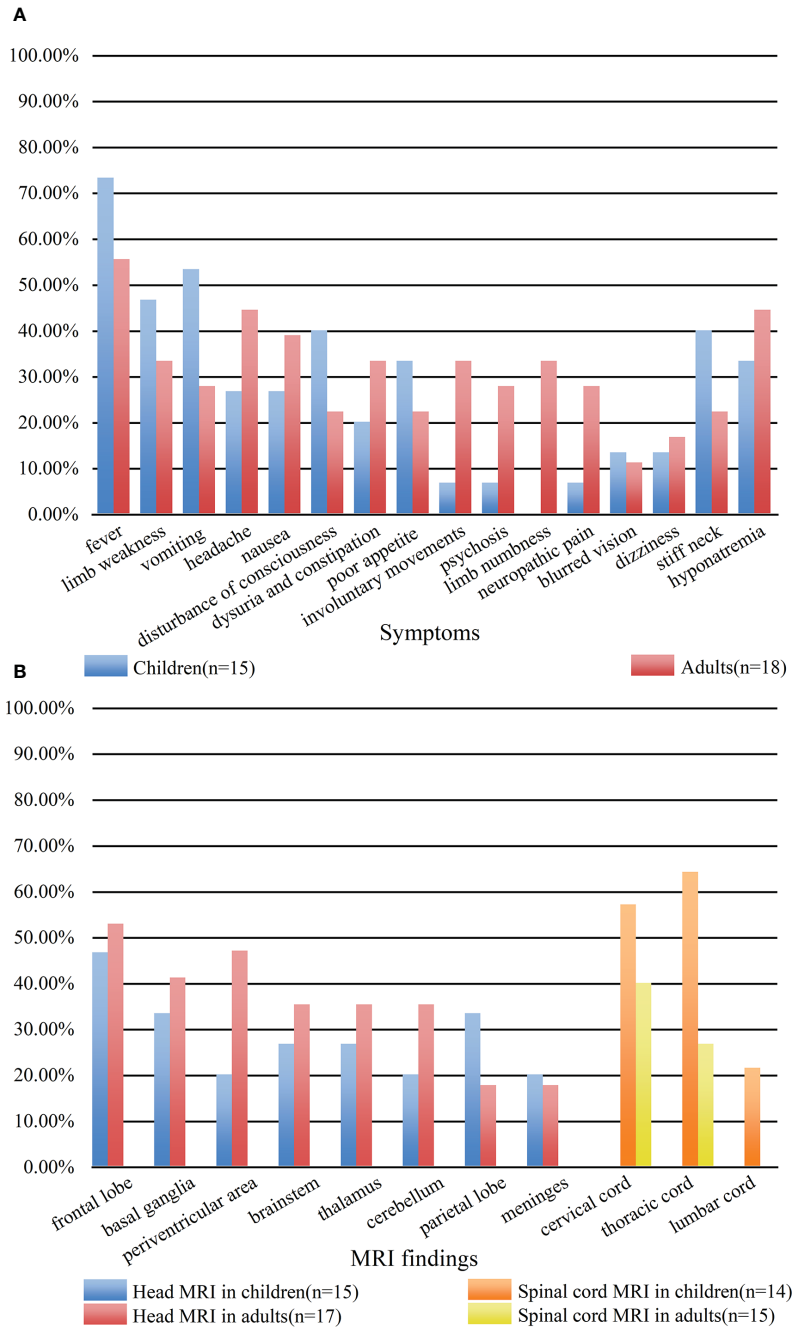


FIGURE 2
Comparison of clinical symptoms **(A)** and MRI findings **(B)** between children and adults. **(A)** The blue bar represented the clinical symptoms of children. The red bar represented the clinical symptoms of adults. **(B)** The blue bar represented the head MRI findings of children. The red bar represented the head MRI findings of adults. The orange bar represented the spinal MRI findings of children. The yellow bar represented the spinal MRI findings of adults.

Of the 33 patients, eight patients had one or more overlapping neural antibodies in the CSF or serum, including seven females and one male. AQP-4 was the most common overlapping antibody in the CSF of GFAP-A patients, followed by MOG and NMDAR. Furthermore, all patients with overlapping AQP4 antibodies responded poorly to first-line immunotherapy (IVMP, IVIG) in the acute phase, and two of them presented with longitudinally extensive transverse myelitis. Table 4 listed the clinical manifestation and MRI characteristics of patients with coexisting neural antibodies.

3.3 MRI findings

All 33 cases underwent MRI examinations, including 32 head MRIs and 29 spinal cord MRIs. MRI abnormalities were mainly T2

TABLE 2 Serological findings of 33 patients with autoimmune GFAP astrocytopathy.

Serological findings	Patients
abnormal tumor markers	**13/33**
Ferritin	5/13
neuron-specific enolase	5/13
tumor-associated antigen 72-4	3/13
carcinoembryonic antigen	2/13
tumor associated antigen 125	1/13
non-small cell lung cancer antigen 21-1	1/13
alpha-fetoprotein	1/13
tumor-associated antigen 19-9	1/13
viral antibodies (immunoglobulin M) in serum	**19/32**
Epstein-Barr virus	12/19
Cytomegalovirus	11/19
coxsackie virus	7/19
measles virus	6/19
herpes simplex virus I	3/19
human parvovirus B-19	2/19
influenza b virus	1/19
parainfluenza virus	1/19
Adenovirus	1/19
non-neural autoantibodies in the serum	**10/33**
anti-SSA antibody	6/10
antinuclear antibody	5/10
rheumatoid factor	1/10
anti-PM-Scl antibody	1/10

sequence high signal lesions, which appeared as patchy, linear, punctate, and stripe patterns. Results from head MRIs were varied: four patients had normal imaging, 28 patients demonstrated abnormal T2 hyperintensities, and 12 of 20 patients showed abnormal contrast enhancement. The lesions were mainly located in the frontal lobe (16/32), basal ganglia (12/32), periventricular area (11/32), brainstem (10/32), cerebellum (9/32), thalamus (9/32), parietal lobe (8/32), and meninges (6/32); enhancement patterns were observed in the meninges (n = 6), periventricular area (n = 3), and basal ganglia (n = 3) (Figures 3A, B). Regarding spinal cord MRI, T2 hyperintensities were observed in the cervical cord (14 cases), followed by the thoracic cord (13 cases). Five of nine patients who underwent contrast-enhanced spinal MRI showed abnormal enhancement. The lesion of patient #17 involved up to 18 spinal cord segments (Figures 3C, D). Additionally, there was no statistical difference in abnormal spine cord MRIs between children and adults (P = 0.264). Figure 2B compared the MRI characteristics between adults and children. Besides, the MRI results revealed that the lesions in GFAP-A patients generally reduced in size or disappeared after immunotherapy. Patient #3 presented with multiple lesions in the brain parenchyma on admission MRI (Figure 3E). Re-examination of MRI at 1 and 2 months after treatment with IVMP and IVIG showed that the lesions gradually reduced (Figures 3F, G). Four months after the treatment, his MRI lesions were significantly reduced in size (Figure 3H). Moreover, his symptoms were fully resolved 6 months after the onset.

3.4 Treatment, outcome, and prognosis analysis

Treatment responses and short-term outcomes are summarized in Figure 4. Most patients experienced a significant improvement in their symptoms after first-line immunotherapy (P = 0.02). Among the 33 patients, 17 were treated with IVMP only; 14 underwent IVMP plus IVIG therapy; 1 patient had only IVIG treatment. And 1 patient received PE for poor outcome after receiving IVMP plus IVIG. Nineteen patients underwent oral tapering of steroids in the maintenance period. Three patients responded poorly to treatment and subsequently received immunosuppressive therapy (MMF, RIT, and TAC). Moreover, five patients were mechanically ventilated because of respiratory failure. 19 proven viral infection and 7 suspected viral infection were treated with antiviral drugs in the early stage of admission.

TABLE 3 CSF findings of 33 patients with autoimmune GFAP astrocytopathy.

CSF findings	Patients	Median (range)
CSF abnormalities	**30/33**	
pleocytosis (>5 × 10^6/L)	27/33	38 (1-950)
Elevated proteins (>450.0 mg/L)	19/33	572.0 (202.7-4433.0)
hypoglycorrhachia (<2.50 mmol/L)	10/33	2.95 (1.00-7.71)
viral antibodies (immunoglobulin M) in CSF	**5/33**	
Epstein-Barr virus	3/5	
enterovirus	1/5	
herpes simplex virus I	1/5	
Oligoclonal antibodies (IgG)	4/33	

TABLE 4 Clinical manifestation and MRI characteristics of patients with coexisting neural antibodies.

Types of coexisting antibodies		Clinical manifestation	MRI characteristics	
			Head	Spinal cord
AQP4	Patient #13	vomiting, blurred vision, walk unsteadiness **(LETM)**	normal	medulla oblongata and C1-C5
	Patient #21	limb numbness, tics **(LETM)**	NA	medulla oblongata, C1-6 and T3-5
	Patient #30	nausea, vomiting, facial pain, alalia, dysphagia, coughing when drinking water, limb numbness **(encephalitis)**	frontal, lateral ventricle and parietal	normal
MOG	Patient #2	blurred vision **(ON)**	frontal, parietal, temporal, corpus callosum, pons, cerebellum, optic nerve	C4-T8
	Patient #16	Fever, headache, nausea, vomiting, dysuria, constipation, blurred vision, limb weakness, epilepsy **(encephalitis, myelitis)**	cerebellum and thalamus	normal
MOG and NMDAR	Patient #11	limb weakness, lethargy, poor appetite **(encephalitis, LETM)**	normal	C2-6 and T9-12
AQP4, NMDAR, GAD651, and GLYR1	Patient #18	headache, dizziness, lethargy, psychosis **(encephalitis)**	frontal and lateral ventricles enhancement in the optic chiasm, bilateral anterior portion of the thalamus, fornix column and triventricular area	normal
YO	Patient #20	dizziness, limb weakness, poor appetite **(encephalitis)**	frontal	NA

LETM, longitudinally extensive transverse myelitis; ON, optic neuritis.

The median mRS score at admission was 3 and that at 4 weeks after the initiation of immunotherapy was 2. Short-term prognosis was significantly better in children than in adults (P = 0.04). In the univariate binary logistic model, the factors associated with poor outcomes included positive non-neural autoantibodies and proven viral infection (P = 0.03, 0.03, respectively). Multivariate binary logistic regression model identified positive non-neural autoantibodies and proven viral infection as the independent factors associated with a poor outcome (P = 0.03, 0.02, respectively) (Table 5).

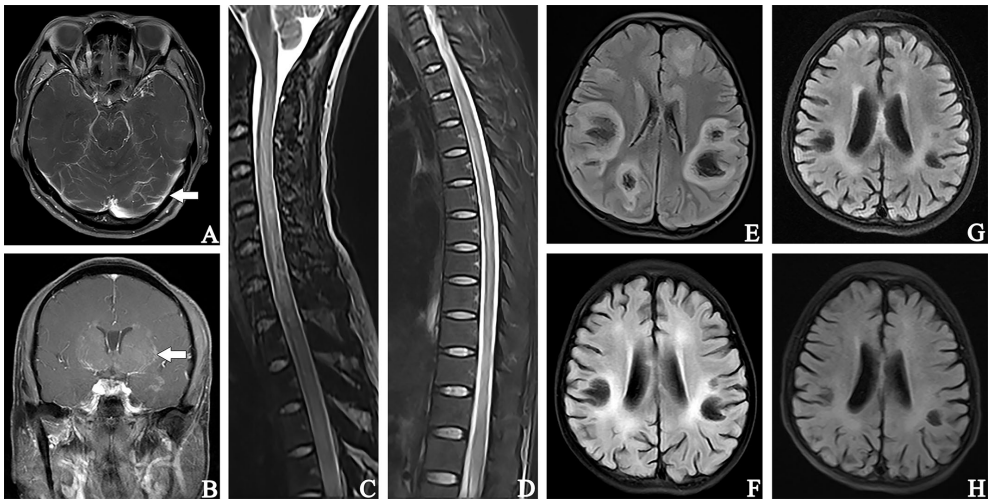


FIGURE 3
Contrast-enhanced T1-weighted magnetic resonance image **(A, B)**. **(A)** patient #18, showed soft meningeal linear enhancement (arrow). **(B)** patient #23, MR image suggested patchy enhancement around the lateral ventricles (arrow). **(C, D)**: patient #18, T2-hyperintense lesions in C2-T12. Fluid attenuated inversion recovery **(E–H)**. MR images of patient #3. **(E)** MR image showed multiple lesions of bilateral brain parenchyma at admission. **(F)** (1 month after treatment with IVMP and IVIG) and **(G)** (2 months after treatment) showed that the extent of lesions gradually decreased and brain atrophy began to appear. **(H)** 4 months after treatment, a significant reduction in the lesion range and high-signal lesions in the bilateral lateral periventricular. The widening and deepening of the cerebral sulcus and fissure were evident.

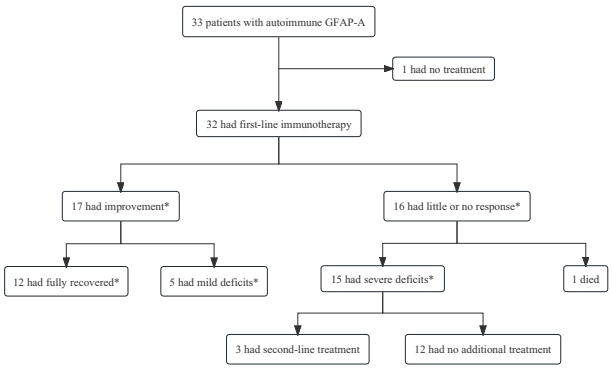


FIGURE 4
The treatment response and short-term outcomes of patients with autoimmune GFAP astrocytopathy. First-line immunotherapy included intravenous methylprednisolone, intravenous immunoglobulin, and plasma exchange, alone or in combination. Second-line immunotherapy included rituximab, tacrolimus, or mycophenolate mofetil. *The occurrence of improvement or absence of improvement was assessed at 4 weeks after the initiation of immunotherapy. Recovered was defined as mRS=0; mild deficits were defined as mRS= 1,2; and severe deficits were defined as mRS= 3,4.

At the last follow-up, seven patients had poor outcomes and 26 patients had good outcomes, with the median mRS score being 0. The average follow-up duration was 12 months, ranging from 3 to 47 months. 8 of 33 patients were hospitalized more than two times, including two patients who were hospitalized seven times (Patients #13 and #21). In addition, four patients relapsed during oral tapering of steroids: two of them had two relapses; one patient had three relapses; and one patient had five relapses. Three patients experienced worsening or recurrence of previous symptoms, and one patient had new symptoms which were significantly alleviated after immunotherapy.

4 Discussion

GFAP-A is a relatively rare autoimmune inflammatory CNS disorder. Despite several studies (2, 3, 7–13), there is limited understanding of its short-term prognosis, which motivated us to conduct this study. Our findings expanded the spectrum of symptoms, which comprise nausea, vomiting, poor appetite, and neuropathic pain. Importantly, patients with positive non-neural

TABLE 5 Factors associated with poor outcomes at 4 weeks after the initiation of immunotherapy (mRS ≥3).

	Odds ratio (95% CI)	P
Univariable analysis		
Demographic data		
age of onset	1.03 (0.99-1.07)	0.11
gender	0.69 (0.18-2.73)	0.60
hospital days	1.01 (0.97-1.05)	0.58
Serologic data		
MLR*	1.66 (0.22-12.54)	0.63
NLR*	0.98 (0.91-1.06)	0.66
tumor marker	0.86 (0.21-3.47)	0.83
proven viral infection in serum	5.71 (1.16-28.07)	**0.03**
positive for non-neural autoantibodies in serum	7.50 (1.28-44.09)	**0.03**
CSF examination		
white cells	1.002 (0.997-1.007)	0.45
protein level	1.000 (0.999-1.001)	0.66
viral antibodies in CSF	5.33 (0.53-54.03)	0.16
glucose level	1.89 (0.78-4.60)	0.16
Clinical symptoms		
fever	0.91 (0.22-3.76)	0.90
hyponatremia	1.43 (0.35-5.79)	0.62
headache	0.65 (0.16-2.72)	0.56
disturbance of consciousness	3.63 (0.74-17.81)	0.11
nausea	0.83 (0.20-3.56)	0.81

(Continued)

TABLE 5 Continued

	Odds ratio (95% CI)	P
vomiting	1.43 (0.35-5.79)	0.62
limb weakness	0.86 (0.21-3.47)	0.83
dizziness	5.33 (0.53-54.03)	0.16
stiff neck	1.09 (0.25-4.82)	0.91
limb numbness	1.08 (0.18-6.32)	0.94
dysuria and constipation	0.42 (0.09-2.01)	0.29
involuntary movements	3.41 (0.56-20.94)	0.19
neuropathic pain	1.08 (0.18-6.32)	0.94
psychosis	2.50 (0.39-16.05)	0.33
poor appetite	1.48 (0.32-6.90)	0.62
blurred vision	0.31 (0.03-3.35)	0.34
prodromal symptoms	0.89 (0.23-3.49)	0.87
ICU admission	1.87 (0.44-7.85)	0.39
mechanical ventilation	5.33 (0.53-54.03)	0.16
mRs at admission	1.44 (0.81-2.57)	0.21
Multivariable analysis		
positive for non-neural autoantibodies in serum	17.67 (1.60-195.11)	0.02
proven viral infection in serum	11.96 (1.26-113.92)	0.03

*MLR, monocyte-to-lymphocyte ratio; NLR, neutrophil-to-lymphocyte ratio.

autoantibodies and proven viral infections in the serum at admission were found to have a poor short-term prognosis, which was rarely reported before. Additionally, our study results differ from those of previous studies in that adult patients were more likely to have sensory symptoms such as limb numbness and poor short-term prognosis. Hence, the present study provides a relatively comprehensive description of the clinical characteristics and short-term prognosis of GFAP-A.

The median age of disease onset in GFAP-A patients was 28 years (range: 2–68 years) in our study, while it was 40–50 years (range: 11 months to 103 years) in previous studies (8, 10). We think that the difference may be because eight patients in our study were enrolled from children's hospitals. Our study confirmed that 29 patients with autoimmune GFAP-A had three or more clinical symptoms, which were diverse and non-specific. We also found other clinical presentations, including nausea and vomiting, which were rarely mentioned previously. The underlying mechanisms were heterogeneous, and included area postrema syndrome, hyponatremia, meningitis, and encephalitis. Poor appetite was not a rare symptom in our study, owing to GFAP expression by the enteric glial cells. It is an important component of the enteric nervous system, which regulates enteric neural reflexes and maintains intestinal homeostasis (14, 15). We also found that limb numbness was more likely to occur in adults, which suggests that adults are more prone to sensory disturbance in autoimmune GFAP-A. In contrast to the findings of Zhuang et al. (8), myelitis in children was also relatively common in our cohort, which was supported by no statistical difference in abnormal spinal cord MRIs between children and adults. As already noted by Flanagan et al., most patients had infectious prodromal symptoms, which indicates that autoimmune GFAP-A may be triggered by infection (2, 9). Additionally, hiccups were the main symptom of area postrema syndrome in autoimmune GFAP-A, supported by MRI findings of T2-hyperintense lesions in the dorsal medulla oblongata (16). Interestingly, frequent neuropathic pain was reported in six cases, and patient #13 presented with left-sided peripheral facial nerve palsy at the first relapse. It is worth noting that GFAP is also expressed by Schwann cells and satellite glial cells of peripheral nerves (17).

Thirteen patients with GFAP-A had serum tumor markers detected in our study. Therefore, clinicians should give high priority to tumor screening in autoimmune GFAP-A patients, especially within 2 years of the onset (4). Additionally, we found that viral antibodies are frequently detected in the serum of autoimmune GFAP-A patients. More importantly, proven viral infection in the serum on admission suggests a poor short-term prognosis. Of note, viral infections are related to CNS autoimmune disorders (18). Although a close relationship between GFAP-A and viral infection has been previously demonstrated, all previous studies detected the virus in the CSF (11, 19–21). We speculate

that serum viral antibodies enter the CNS through the blood-brain barrier and then participate in the pathophysiological process of autoimmune GFAP-A. The underlying mechanism still needs to be confirmed by further animal experiments. The other factor associated with poor short-term prognosis was overlapping non-neural autoantibodies, which were also encountered in 75% of the first Chinese cohort (11). Even though these antibodies are not specific per se, it suggests that clinicians should be alert for co-morbid autoimmune diseases other than CNS involvement in patients with GFAP-A, which may exacerbate the patient's condition. Iorio et al. (13) also reported that GFAP-A in combination with other autoimmune diseases was common. In our study, we noted that eight patients had coexisting neural autoantibodies, with the most common being AQP-4 antibodies, which tend to occur in women. Similar to the findings of Xiao et al. (10), patients with overlapping AQP-4 antibodies responded poorly to immunotherapy in the acute phase, but the exact mechanism is unclear.

Our results provide statistical evidence that first-line immunotherapy is effective in most patients with autoimmune GFAP-A in the acute phase, which is more convincing than the results of previous observational studies. As recently described (9), relapse may occur during oral tapering of steroids in most patients. The recurrence rate was 12.1% in our cohort, which was lower than that reported in previous studies (18%) (7, 12). Relapses usually involved the worsening of previous symptoms, although a few patients developed new symptoms. In contrast to the findings of Xiao et al. (10), we noticed that one patient treated with TAC still had frequent relapses during follow-up, questioning the effectiveness of TAC for preventing a relapse. Intriguingly, we found children had a better short-term prognosis, which was reported to be poor previously.

Our study findings provide novel insights into the clinical characteristics and short-term prognosis of GFAP-A patients. However, there are some limitations of our study. Firstly, this was not a randomized and prospective study, but a precursor to future trials to explore the prognostic factors. Secondly, we only evaluated 33 patients including 15 children, which is a small sample size and the presence of population heterogeneity. In the future, more studies are needed in a larger population. Lastly, patients with Alzheimer's disease and cancer have been shown to have serum GFAP antibodies (22). Therefore, the specificity of serum GFAP antibodies remains uncertain. We only included patients with positive CSF GFAP antibodies, which may have excluded some patients.

In conclusion, our study not only expands the known spectrum of clinical characteristics of GFAP-A, but also statistically confirms the effectiveness of first-line immunotherapy in the acute phase. Furthermore, we identified the prognostic factors associated with the short-term outcomes of GFAP-A as well as significant differences between children and adults with GFAP-A.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

Written informed consent was obtained from the individual(s), and minor(s)' legal guardian/next of kin, for the publication of any potentially identifiable images or data included in this article.

Author contributions

NX and WZ contributed to conception and design of the study. YX and YW organized the database. FL performed the statistical analysis. WZ wrote the first draft of the manuscript. LW, YL, HL, and CW wrote sections of the manuscript. All authors contributed to manuscript revision, read, and approved the submitted version.

Funding

This study was supported by the National Natural Science Foundation of China (No.81971214) and the Outstanding Young Talent Cultivation Project of Henan Science and Technology Innovation Talents (YXKC2022037).

Acknowledgments

The authors are grateful to the patients and their families for the support and cooperation.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Li D, Liu X, Liu T, Liu H, Tong L, Jia S, et al. Neurochemical regulation of the expression and function of glial fibrillary acidic protein in astrocytes. *Glia* (2020) 68 (5):878–97. doi: 10.1002/glia.23734

2. Flanagan EP, Hinson SR, Lennon VA, Fang B, Aksamit AJ, Morris PP, et al. Glial fibrillary acidic protein immunoglobulin G as biomarker of autoimmune astrocytopathy: analysis of 102 patients. *Ann Neurol* (2017) 81(2):298–309. doi: 10.1002/ana.24881

3. Fang B, McKeon A, Hinson SR, Kryzer TJ, Pittock SJ, Aksamit AJ, et al. Autoimmune glial fibrillary acidic protein astrocytopathy: a novel meningoencephalomyelitis. *JAMA Neurol* (2016) 73(11):1297–307. doi: 10.1001/jamaneurol.2016.2549

4. Kunchok A, Zekeridou A, McKeon A. Autoimmune glial fibrillary acidic protein astrocytopathy. *Curr Opin Neurol* (2019) 32(3):452–8. doi: 10.1097/WCO.0000000000000676

5. Yang X, Liang J, Huang Q, Xu H, Gao C, Long Y, et al. Treatment of autoimmune glial fibrillary acidic protein astrocytopathy: follow-up in 7 cases. *Neuroimmunomodulation* (2017) 24(2):113–9. doi: 10.1159/000479948

6. Yang X, Xu H, Ding M, Huang Q, Chen B, Yang H, et al. Overlapping autoimmune syndromes in patients with glial fibrillary acidic protein antibodies. *Front Neurol* (2018) 9:251. doi: 10.3389/fneur.2018.00251

7. Dubey D, Hinson SR, Jolliffe EA, Zekeridou A, Flanagan EP, Pittock SJ, et al. Autoimmune gfap astrocytopathy: prospective evaluation of 90 patients in 1 Year. *J Neuroimmunol* (2018) 321:157–63. doi: 10.1016/j.jneuroim.2018.04.016

8. Zhuang X, Jin K, Li X, Li J. Autoimmune glial fibrillary acidic protein astrocytopathy in children: a retrospective study. *Eur J Med Res* (2022) 27(1):11. doi: 10.1186/s40001-022-00641-y

9. Gravier-Dumonceau A, Ameli R, Rogemond V, Ruiz A, Joubert B, Muñiz-Castrillo S, et al. Glial fibrillary acidic protein autoimmunity: a French cohort study. *Neurology* (2022) 98(6):e653–e68. doi: 10.1212/WNL.0000000000013087

10. Xiao J, Chen X, Shang K, Tang Y, Chen M, Deng G, et al. Clinical, neuroradiological, diagnostic and prognostic profile of autoimmune glial fibrillary acidic protein astrocytopathy: a pooled analysis of 324 cases from published data and a single-center retrospective study. *J Neuroimmunol* (2021) 360:577718. doi: 10.1016/j.jneuroim.2021.577718

11. Long Y, Liang J, Xu H, Huang Q, Yang J, Gao C, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in Chinese patients: a retrospective study. *Eur J Neurol* (2018) 25(3):477–83. doi: 10.1111/ene.13531

12. Fang H, Hu W, Jiang Z, Yang H, Liao H, Yang L, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in children: a retrospective analysis of 35 cases. *Front Immunol* (2021) 12:761354. doi: 10.3389/fimmu.2021.761354

13. Iorio R, Damato V, Evoli A, Gessi M, Gaudino S, Di Lazzaro V, et al. Clinical and immunological characteristics of the spectrum of gfap autoimmunity: a case series of 22 patients. *J Neurol Neurosurg Psychiatry* (2018) 89(2):138–46. doi: 10.1136/jnnp-2017-316583

14. Jessen KR, Mirsky R. Glial cells in the enteric nervous system contain glial fibrillary acidic protein. *Nature* (1980) 286(5774):736–7. doi: 10.1038/286736a0

15. Seguella L, Gulbransen BD. Enteric glial biology, intercellular signalling and roles in gastrointestinal disease. *Nat Rev Gastroenterol Hepatol* (2021) 18(8):571–87. doi: 10.1038/s41575-021-00423-7

16. Deng B, Wang J, Yu H, Jin L, Qiu Y, Liu X, et al. Area postrema syndrome in autoimmune glial fibrillary acidic protein astrocytopathy: a case series and literature review. *Neurol Neuroimmunol Neuroinflamm* (2022) 9(6):e200029. doi: 10.1212/NXI.0000000000200029

17. Yang Z, Wang KKW. Glial fibrillary acidic protein: from intermediate filament assembly and gliosis to neurobiomarker. *Trends Neurosci* (2015) 38(6):364–74. doi: 10.1016/j.tins.2015.04.003

18. Getts DR, Chastain EML, Terry RL, Miller SD. Virus infection, antiviral immunity, and autoimmunity. *Immunol Rev* (2013) 255(1):197–209. doi: 10.1111/imr.12091

19. Handoko M, Hong W, Espineli E, Saxena K, Muscal E, Risen S. Autoimmune glial fibrillary acidic protein astrocytopathy following herpes simplex virus encephalitis in a pediatric patient. *Pediatr Neurol* (2019) 98:85–6. doi: 10.1016/j.pediatrneurol.2019.05.010

20. Issa N, Martin C, Dulau C, Camou F. Severe anti-gfap meningo-encephalomyelitis following viral infection. *Mult Scler Relat Disord* (2020) 45:102448. doi: 10.1016/j.msard.2020.102448

21. Li J, Xu Y, Ren H, Zhu Y, Peng B, Cui L. Autoimmune gfap astrocytopathy after viral encephalitis: a case report. *Mult Scler Relat Disord* (2018) 21:84–7. doi: 10.1016/j.msard.2018.02.020

22. Wang KKW, Yang Z, Yue JK, Zhang Z, Winkler EA, Puccio AM, et al. Plasma anti-glial fibrillary acidic protein autoantibody levels during the acute and chronic phases of traumatic brain injury: a transforming research and clinical knowledge in traumatic brain injury pilot study. *J Neurotrauma* (2016) 33(13):1270–7. doi: 10.1089/neu.2015.3881

frontiers | Frontiers in Immunology

TYPE Original Research
PUBLISHED 07 June 2023
DOI 10.3389/fimmu.2023.1065650

OPEN ACCESS

EDITED BY
Takao Koike,
Hokkaido Medical Center for Rheumatic Diseases, Japan

REVIEWED BY
Matteo Lucchini,
Catholic University of the Sacred Heart, Rome, Italy
Ikuma Nakagawa,
Hokkaido Medical Center for Rheumatic Diseases, Japan

*CORRESPONDENCE
Hongzhi Guan
pumchghz@126.com

†These authors have contributed equally to this work

RECEIVED 10 October 2022
ACCEPTED 19 May 2023
PUBLISHED 07 June 2023

CITATION
Fan S, Zhao J, Hou B, Liu M, Niu J, Zhou Y, Mao C, Ren H, Feng F, Li M, Zeng X, Zhu Y and Guan H (2023) Rheumatoid meningitis: a rare neurological complication of rheumatoid arthritis.
Front. Immunol. 14:1065650.
doi: 10.3389/fimmu.2023.1065650

Rheumatoid meningitis: a rare neurological complication of rheumatoid arthritis

Siyuan Fan[1†], Jiuliang Zhao[2,3,4,5†], Bo Hou[6], Mange Liu[1], Jingwen Niu[1], Yan Zhou[1], Chenhui Mao[1], Haitao Ren[1], Feng Feng[6], Mengtao Li[2,3,4,5], Xiaofeng Zeng[2,3,4,5], Yicheng Zhu[1] and Hongzhi Guan[1*]

[1]Department of Neurology, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China, [2]Department of Rheumatology and Clinical Immunology, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China, [3]National Clinical Research Center for Dermatologic and Immunologic Diseases, Ministry of Science and Technology, Beijing, China, [4]State Key Laboratory of Complex Severe and Rare Diseases, Peking Union Medical College Hospital, Beijing, China, [5]Key Laboratory of Rheumatology and Clinical Immunology, Ministry of Education, Beijing, China, [6]Department of Radiology, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

Objective: To describe the clinical and neuroimaging characteristics of rheumatoid meningitis (RM) in Chinese patients.

Methods: The patients admitted to our hospital with the diagnosis of RM in the past 8 years were retrospectively analyzed.

Results: Six patients with RM were identified among 933 patients admitted with rheumatoid arthritis (RA). The symptoms of meningitis occurred after onset of arthritis in five patients and before onset in one. Headache (n=6), hyperacute focal neurological deficits (n=4) and seizures (n=3) were the most prevalent symptoms. The nadir modified Rankin Scale score was ≥3 in five patients. Rheumatoid factor was elevated in all patients, and interleukin-6 levels in cerebrospinal fluid were dramatically elevated in three of four tested patients. Magnetic resonance imaging of the brain revealed that the meninges were affected in all patients and the cerebral parenchyma was affected in one patient. The lesions were generally located in the frontoparietal region and showed restricted diffusion along the adjacent subarachnoid space. RM occurred during disease-modifying therapy in four patients. In the acute episode, three patients improved on tocilizumab and the other three improved on pulse corticosteroids. For maintenance therapy, two patients received combined therapy of tocilizumab and other immunosuppressive agents, one received adalimumab and methotrexate, and two received low-dose oral corticosteroids with an immunosuppressive agent. Five patients had a good outcome, and one died of *Pneumocystis jirovecii* pneumonia after stabilization of his neurologic conditions. No relapse of RM occurred on immunotherapy during follow-up.

Conclusions: Chinese patients with RM share some remarkable clinical and neuroimaging features and respond well to appropriate immunotherapy. Tocilizumab could be a treatment option for this severe complication of RA.

KEYWORDS

rheumatoid arthritis, rheumatoid meningitis, immunotherapy, tocilizumab (TCZ), neuroimage

1 Introduction

Rheumatoid meningitis (RM) is a rare but severe neurological complication of rheumatoid arthritis (RA) and has a predilection for the meninges rather than the brain parenchyma (1). Neurological manifestations of RM include headache, cranial nerve palsy, seizure, altered mental status, and focal neurologic deficits. Some patients with RM present with acute focal neurologic deficits, which may be initially misdiagnosed as acute ischemic stroke (2–8). Some patients with RA develop these symptoms when still on disease-modifying therapies (DMTs), such as biological agents and immunosuppressive agents, thus necessitating differential diagnosis of central nervous system (CNS) opportunistic infections and drug-induced meningitis (7). RM can occur either before or many years after onset of arthritis (9), and the severity of CNS involvement correlates poorly with severity of systemic arthritis (2, 10), which renders the diagnosis of RM even more challenging. Meningeal biopsy may support a diagnosis of RM. However, it is invasive and sometimes has nonspecific findings (11). Some specific neuroimaging manifestations have been described but require further investigation.

RM is rarely reported in Chinese patients (12, 13). In this study, we retrospectively investigated the clinical, laboratory, and neuroimaging features and the treatment provided and outcomes in six patients with RM admitted to our hospital. Our aim was to characterize Chinese patients with RM and provide new evidence for the diagnosis and treatment of RM.

2 Materials and methods

This single-center retrospective case series study was conducted at the Peking Union Medical College Hospital, a tertiary referral center in Beijing, China. Patients with encephalitis or meningitis of unknown origin may be referred to our encephalitis center, known as the National Center for Autoimmune Encephalitis Quality Improvement.

We retrospectively analyzed patients admitted to our hospital with a diagnosis of "rheumatoid meningitis" between January 2013 and June 2021. The search string used for retrieving relevant medical information from the electronic health records was ("rheumatoid") AND ("meningitis" OR "encephalitis" OR "meningoencephalitis" OR "meninges" OR "brain"). The patients were required to fulfill the 2010 American College of Rheumatology/European League Against Rheumatism classification criteria for RA. The diagnosis of RM was reviewed by a panel of specialists, including neurologists and rheumatologists, based on clinical and laboratory findings. The Disease Activity Score-28 for Rheumatoid Arthritis with ESR (DAS 28) was evaluated and calculated by rheumatologists and the modified Rankin Scale (mRS) was evaluated by neurologists. In this study, pulse corticosteroids were defined as a methylprednisolone dosage of more than 250 mg per day (or its equivalent) for several days. Tocilizumab was administered intravenously at a dose of 8 mg/kg.

All individual-level medical information, including demographic, clinical, laboratory, and neuroimaging findings, treatments, and outcome data, were retrieved from the electronic health records.

The study was approved by the institutional review board of Peking Union Medical College Hospital (S-K1747). Written informed consent for treatment with tocilizumab or pulse corticosteroids was given by each patients' legal surrogate. Patient consent for publication was not required because de-identified data were used in the study.

2.1 Data availability

Data related to this study can be made available on request to the corresponding author.

3 Results

3.1 Clinical characteristics

Six patients (0.6%) with RM were identified among 933 patients admitted to hospital with RA during the study period. Their clinical features are summarized in Table 1 and Figure 1. Five of the six patients were female. The age at onset of RM ranged from 33 to 64 years. The time interval between onset of meningeal symptoms and the last follow-up ranged from 6 to 48 months. The symptoms of meningitis or meningoencephalitis occurred after the onset of arthritis in five patients and before the onset of arthritis in one patient (case 2). Meanwhile, active arthritis was absent in one patient (case 3, the DAS 28 score was 2.06) at onset of RM.

Headache occurred in all six patients and was the first neurological symptom in four. Four patients presented with hyperacute focal neurological deficits (onset within several minutes). One patient (case 5) initially presented with recurrent episodes of left-sided weakness and numbness and then progressed to persistent hemiplegia in the left upper and lower limbs. Three patients presented with epileptic seizures and one presented with cranial nerve palsy. The nadir mRS score was ≥3 in five patients.

The diagnosis of RM in these patients was delayed, with a range of 3 to 12 months from the onset of symptoms. The initial diagnosis was acute ischemic stroke in two patients (Cases 2 and 4), bacterial meningitis in one (Case 1), tuberculous meningitis in one (Case 5), viral meningitis in one (Case 3), and autoimmune encephalitis in one (Case 6).

3.2 Laboratory and neuroimaging findings

The laboratory findings in these patients are summarized in Table 2. Rheumatoid factor was elevated in all patients. Furthermore, all six patients were positive for anticitrullinated peptide antibody and five were positive for antiperinuclear factor and antikeratin antibody. The erythrocyte sedimentation rate was elevated in five patients. Blood interleukin (IL)-6 levels were normal in two of three tested patients.

10.3389/fimmu.2023.1065650

TABLE 1 The clinical features of the six patients with rheumatoid meningitis.

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6
Sex	Female	Female	Female	Female	Male	Female
Age at disease onset, y	43	53	33	52	64	47
RA duration before RM, y	12	0	2.5	10	30	20
DMT before RM	MTX, LEF	None	None	CS	CS, LEF, HCQ	LEF, MTX, SASP
DAS 28 at disease onset	6.21	NA	2.06	NA	NA	4.98
Neurologic symptoms	Headache, dizziness	Headache, recurrent episodes of slurred speech with left-sided numbness, diplopia	Headache, recurrent episodes of slurred speech with left-sided weakness and numbness, recurrent episodes of right-sided numbness, seizures	Cognitive decline, seizures, recurrent episodes of left-sided weakness and numbness, headache	Headache, recurrent episodes of left-sided weakness and numbness, persistent weakness in the left upper and lower limbs, altered mental status	Seizures, cognitive decline, hallucinations, headache, dizziness, tinnitus, unsteady gait, urinary incontinence
Other extra-articular symptoms	Fever	None	None	None	None	None
Time from Sx to Dx, m	4	5	6	12	3	4
mRS at nadir	2	3	3	3	5	4
DMT after RM	TCZ, HCQ	TCZ, LEF, MTX, ADM	CS, MMF, TCZ	CS, CTX, MTX	CS, LEF, HCQ, TCZ, MTX	CS, AZA
Antimicrobial drugs administered	Ceftriaxone	None	Acyclovir	None	MRP, LFX, RMP	None
F/U duration, m	17	16	36	48	6	10
mRS at last F/U	0	1	0	1	6	1

ADM, adalimumab; AZA, azathioprine; CS, corticosteroids; CTX, cyclophosphamide; DMT, disease-modifying therapy; Dx, diagnosis; F/U, follow-up; HCQ, hydroxychloroquine; LEF, leflunomide; LFX, levofloxacin; m, month; MMF, mycophenolate mofetil; MRP, Meropenem; mRS, modified Rankin Scale; MTX, methotrexate; RA, rheumatoid arthritis; RM, rheumatoid meningitis; RMP, rifampicin; SASP, sulfasalazine; Sx, symptoms; TCZ, tocilizumab; y, year.

Lumbar puncture was performed in all patients, and intracranial hypertension was detected in four. All patients showed mild to moderate pleocytosis in cerebrospinal fluid (CSF), and two patients had increased polymorphonuclear neutrophil counts. One patient tested negative for rheumatoid factor in CSF. IL-6 levels in CSF were tested in four patients and found to be dramatically elevated in three. IL-6 was higher in CSF than in serum in two tested patients (cases 2 and 5). Tests for other causes of meningitis yielded negative results (Table 2).

Magnetic resonance imaging (MRI) of the brain revealed that the meninges were affected in all patients, and that the cerebral parenchyma was affected in one patient (case 5). Enhancement of the pachymeninges (case 1, Figures 2A–D) or both the pachymeninges and leptomeninges (cases 2–6, Figures 2E–H) was shown in different patients. Lesions were generally located on the convex surface of the cerebral hemisphere with sparing of the meninges around the basal cisterns (cases 1–6). Dramatic asymmetric involvement of the meninges was observed in three patients (cases 3–5), all of whom had recurrent focal neurological deficits on the opposite side. Diffusion-weighted imaging (DWI) showed lesions with restricted diffusion along the adjacent subarachnoid space (cases 1–6, Figure 2). Hydrocephalus was shown in one patient (case 6). The meningeal and parenchymal lesions were significantly improved after immunotherapy, with gradual disappearance of hyperintensities on DWI. In case 5, repeated MRI of the brain showed a dynamic change. About one month after onset of meningeal symptoms, the meninges of the right parietal lobe were mainly affected with small (≤1cm) periventricular white matter lesions. About 2 months after onset, the meninges of the right frontal lobe were also affected with formation of confluent white matter lesions in the right parietal lobe. About 3 months after onset, most of the right parietal lobe was affected (Figures 2Q–T). After immunotherapy, the lesions were significantly reduced (Figures 2U–X).

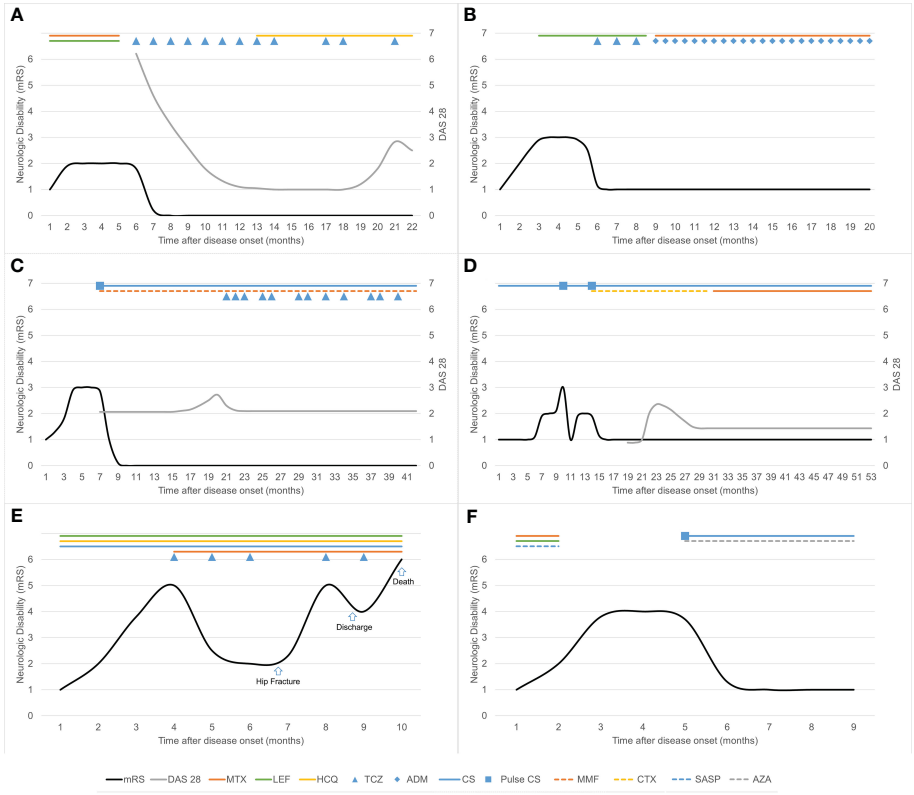


FIGURE 1
Clinical course and immunotherapy in six patients with rheumatoid meningitis. Neurologic disability was measured using the modified Rankin Scale (mRS). **(A)** In case 1, treatment with tocilizumab was transiently interrupted because of herpes zoster and surgery for gallbladder stones. **(B)** In case 2, treatment with tocilizumab was replaced by adalimumab after occurrence of skin rash. **(C)** In case 3, tocilizumab was administered once every 1–2 months because of recurrence of articular symptoms. Maintenance corticosteroid therapy was prednisone up to 10 mg daily orally. **(D)** In case 4, the maintenance corticosteroid dose was prednisone up to 10 mg daily (or its equivalent) orally. **(E)** In case 5, treatment with tocilizumab was transiently interrupted as a result of hip fracture surgery. **(F)** In case 6, the maintenance corticosteroid dose was methylprednisolone 12 mg daily orally. ADM, adalimumab; AZA, azathioprine; CS, corticosteroids; CTX, cyclophosphamide; DAS 28, Disease Activity Score-28 for Rheumatoid Arthritis with ESR; HCQ, hydroxychloroquine; LEF, leflunomide; MMF, mycophenolate mofetil; MTX, methotrexate; SASP, sulfasalazine; TCZ, tocilizumab.

3.3 Immunotherapy and outcomes

Details of the immunotherapy administered and outcomes are summarized in Figure 1 and Table 1. RM occurred during DMT in four patients. Case 1 had a medical history of central serous chorioretinopathy and case 2 had a history of stage 3 hypertension, diabetes mellitus, and dyslipidemia and was overweight. Both these patients refused steroids. Case 5 was already receiving steroids for severe arthritis. Three patients (cases 1, 2, and 5) improved on tocilizumab induction therapy, and the other three (cases 3, 4, and 6) improved on pulse methylprednisolone induction therapy during the acute episode. The neurological symptoms in these patients improved dramatically within the first week. For maintenance therapy, one patient received tocilizumab and hydroxychloroquine, one received tocilizumab, mycophenolate mofetil and low-dose oral corticosteroids, one received adalimumab and methotrexate, and two received low-dose oral corticosteroids with an immunosuppressive agent.

One patient (case 5) had complex medical conditions. Before immunotherapy, his level of consciousness decreased, and he developed complete hemiplegia on the left side. He showed significant improvement after treatment with tocilizumab and steroids. He was able to walk and live independently about 2 months after immunotherapy. However, he experienced a traumatic hip fracture during exercise, after which he became bedridden despite an artificial femoral head replacement. His neurological symptoms worsened, and he received prolonged treatment with steroids (≥1mg/kg daily) for 2 months and monthly tocilizumab. After discharge, he showed some improvement of neurological symptoms. However, he finally died of *Pneumocystis jirovecii* pneumonia, a serious infective complication of immunosuppressive therapy. The other five patients had a good outcome, with an mRS score of 0–1 at the last follow-up. No relapse of RM occurred while the patients were on immunotherapy during a median follow-up of 16.5 months.

4 Discussion

This retrospective investigation has characterized the clinical and neuroimaging features of Chinese patients with RM and adds some new insights into this condition. First, patients with RM exhibited a range of clinical manifestations, including hyperacute

10.3389/fimmu.2023.1065650

TABLE 2 The laboratory findings of the six patients with rheumatoid meningitis.

	Case 1	Case 2	Case 3	Case 4	Case 5	Case 6
Peripheral blood†						
RF (IU/mL)	52	74	75	36.4	234	76
ACPA	+	+	+	+	+	+
APF	+	+	+	+	+	–
AKA	+	+	+	+	+	–
ESR (mm/h)	58	85	8	29	89	36
hsCRP (mg/L)	11.31	33.44	4.27	3.67	166.63	NA
IgG (g/L)	8.41	8.64	13.65	7.89	9.25	9.96
CD19+ B cell count (/μL)	180	NA	237	110	60	NA
CD4+ T cell count (/μL)	680	NA	996	341	641	NA
CD8+ T cell count (/μL)	185	NA	693	206	536	NA
IL-6 (pg/mL)	NA	6.4	NA	3.6	48.1	NA
IL-8 (pg/mL)	NA	32	NA	41	22	NA
IL-10 (pg/mL)	NA	5.0	NA	5.0	5.0	NA
CSF†						
Opening pressure (mmH_20)	290	260	>330	155	290	NA
WBC count (/μL)	48	34	53	9	84	26
PMN count (/μL)	12	2	15	0	4	3
Protein (g/L)	0.63	0.62	NA	0.44	0.76	0.58
Glucose (mmol/L)	2.7	2.8	NA	3.2	3.3	2.9
IgG (mg/L)	49.2	138.0	NA	66.2	237.0	NA
IgG index	0.59	1.74	NA	1.01	2.16	NA
SOB	–	+	NA	+	+	NA
RF (IU/mL)	NA	NA	0	NA	NA	NA
IL-6 (pg/mL)	>1000	457.0	2.0	NA	244.0	NA
IL-8 (pg/mL)	288	294	42	NA	134	NA
IL-10 (pg/mL)	8.0	6.9	5.0	NA	7.7	NA
Important tests with negative results						
Peripheral blood	ANA, ANCA, APLA, ACE, IgG4, MOG, BAT	ANA, ANCA, APLA, ACE, IgG4, MOG, BAT, CrAg	ANA, ACE, IgG4, AQP4	ANA, ANCA, APLA, ACE, IgG4, AQP4	ANA, ANCA, APLA, ACE, IgG4, BAT, CrAg	ANA, ANCA, APLA, ACE, IgG4, BAT, CrAg
CSF	Cytology, Xpert, CrAg, bacterial culture, fungal culture, TB culture, Filmarray ME, mNGS	Cytology, Xpert, CrAg, bacterial culture, fungal culture, mNGS	Cytology, TB PCR, bacterial culture, fungal culture	Cytology, CrAg, bacterial culture	Cytology, Xpert, CrAg, bacterial culture, fungal culture, TB culture, mNGS	Cytology, TB PCR, Xpert, CrAg, bacterial culture, HSV PCR

ACE, angiotensin converting enzyme levels; ACPA, anticitrullinated peptide antibody; AKA, antikeratin antibody; ANA, antinuclear antibodies; ANCA, antineutrophil cytoplasmic antibodies; APF, antiperinuclear factor; APLA, antiphospholipid antibodies; AQP4, anti- aquaporin 4 antibody; BAT, Brucella agglutination test; CD, cluster of differentiation; CrAg, Cryptococcus antigen test; CSF, cerebrospinal fluid; ESR, erythrocyte sedimentation rate; hsCRP, high sensitivity C-reaction protein; HSV, herpes simplex virus; IgG, Immunoglobulin G; IgG4, immunoglobulin G4 levels; IL, interleukin; mNGS, metagenomic next-generation sequencing; MOG, anti-myelin oligodendrocyte glycoprotein antibody; NA, not available; PCR, polymerase chain reaction; PMN, polymorphonuclear neutrophil; RF, rheumatoid factor; SOB, specific oligoclonal bands; TB, tuberculosis; WBC, white blood cell. †, performed before the initiation of immunotherapy for RM.

10.3389/fimmu.2023.1065650

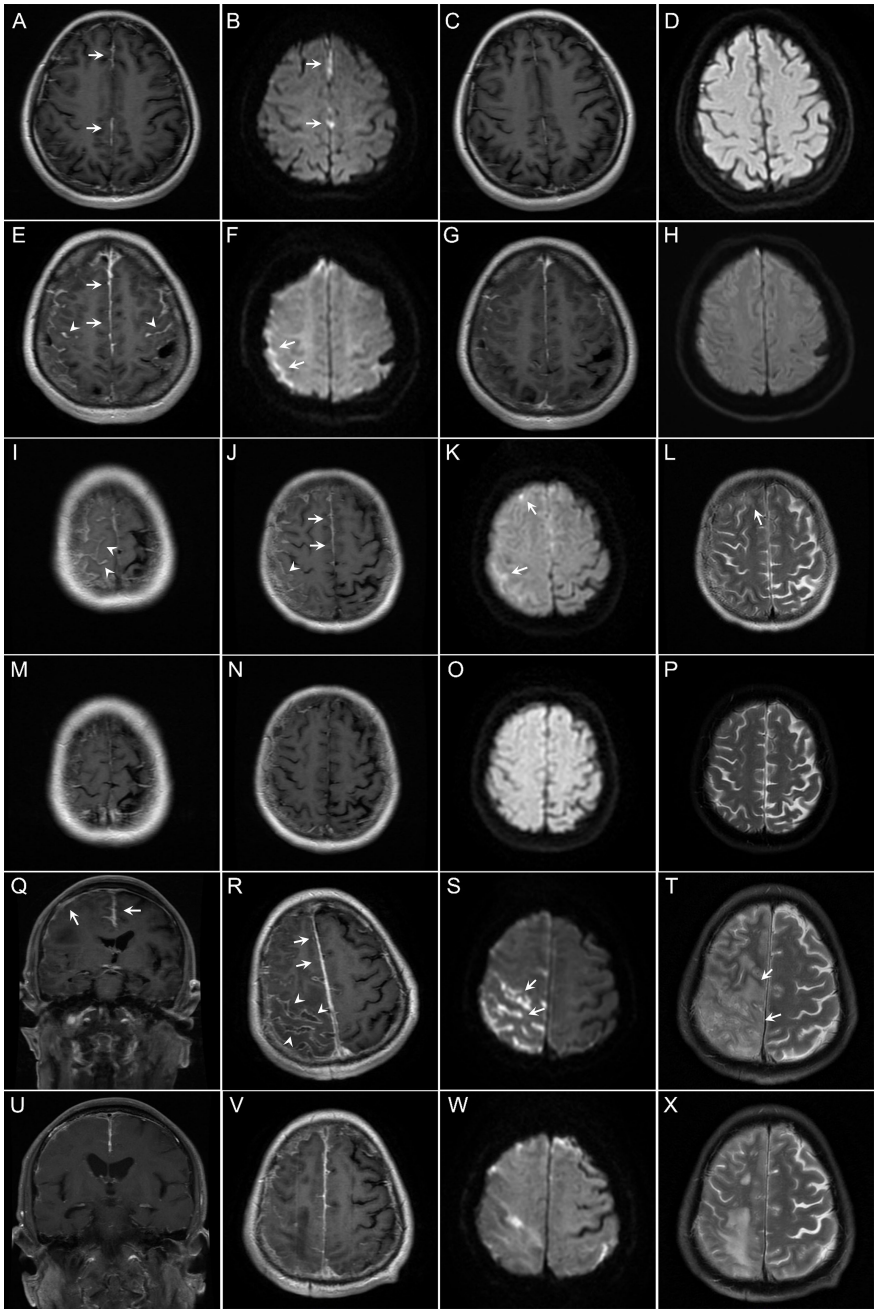


FIGURE 2
Findings on magnetic resonance imaging (MRI) of the brain in the patients with rheumatoid meningitis in this study. **(A–D)** In case 1, brain contrast MRI performed 4 months after onset of symptoms of meningitis showed pachymeningeal enhancement (**A**, arrows). Diffusion-weighted imaging (DWI) showed restricted diffusion (**B**, arrows). The patient experienced improvement, with resolution of the lesions 10 months after immunotherapy **(C, D)**. **(E–H)** In case 2, brain contrast MRI performed 5 months after onset of symptoms of meningitis showed enhancement of both the pachymeninges (**E**, arrows) and leptomeninges (**E**, arrowheads). Restricted diffusion was shown on DWI (**F**, arrows). Repeated MRI showed reduction of contrast enhancement and resolution of restricted diffusion 3 months after immunotherapy **(G, H)**. **(I–P)** In case 4, brain MRI performed 12 months after onset of symptoms of meningitis also showed involvement of both the pachymeninges (**J**, arrows) and leptomeninges (**I, J**, arrowheads) with restricted diffusion (**K**, arrows). T2-weighted MRI scans showed a small cortical lesion (**L**, arrow). Repeated MRI showed resolution of the lesions 28 months after immunotherapy **(M–P)**. **(Q–X)** In case 5, brain contrast MRI performed 3 months after onset of symptoms of meningitis revealed asymmetric involvement of both the pachymeninges (**Q, R**, arrows) and leptomeninges (**R**, arrowheads). Note the lesions are mainly located on the convex surface of the cerebral hemisphere. DWI showed sulcal restricted diffusion (**S**, arrows). T2-weighted MRI scans showed lesions in the parenchyma (**T**, arrows). Repeated MRI showed reduction of the lesions 3 months after immunotherapy **(U–X)**.

focal neurological deficits and seizures, which served as crucial clues for the diagnosis. Second, Chinese patients with RM and the previously reported patients shared some striking neuroimaging features. Third, the patients responded well to appropriate immunotherapy. Tocilizumab might be effective as both induction and maintenance therapy in patients with RM.

RM is a rare neurological complication of RA. A retrospective study by Parsons et al. in 2020 identified 14 patients with RM within

the previous 28 years at the Mayo Clinic (11). In 2021, Villa et al. conducted a systematic review in which they identified 130 patients with RM from 103 studies reported between 1954 and 2020 (14). In this study, we found that less than one percent of inpatients admitted with RA developed RM. This proportion of patients with RA who develop meningeal involvement might even be overestimated, given that some of our patients were specifically referred to our encephalitis center.

Diagnosis of RM is challenging. As in our patients, meningitis can occur as the initial clinical manifestation of RA or after decades of arthritis and can also occur in the absence of active arthritis. Furthermore, many of the patients developed meningitis when they are still on DMT, and the neuroimaging findings may resemble a subdural empyema (15), making CNS infection and drug-induced meningitis likely. Until now, no diagnostic criteria for RM have been established. Therefore, the diagnosis of RM relies on appropriate exclusion of infectious, neoplastic, and other autoimmune etiologies. Meningeal biopsy performed for the purposes of diagnosis and differential diagnosis might reveal three abnormal patterns: rheumatoid nodules, vasculitis, and nonspecific meningeal inflammation (16). A systematic review found that 72.5% of patients underwent biopsy, which showed rheumatoid nodules in 42.3% cases, nonspecific meningeal inflammation in 94.8%, and vasculitis in 16.5% (14). In a retrospective study of 10 patients who underwent biopsy, 90% showed nonspecific inflammation or granulomatous necrosis (11). Many patients lack the relatively specific pathological findings of rheumatoid nodules or vasculitis. Furthermore, meningeal biopsy is invasive. Therefore, RM requires other diagnostic clues for diagnosis.

Patients with RM have some distinctive clinical manifestations. The analysis of 130 patients with RM by Villa et al. revealed that the common clinical manifestations were focal neurological signs (64.6%), systemic symptoms (51.3%), episodic headache (50.4%), neuropsychiatric alterations (47.7%), seizure (40.2%), and joint manifestations (27.4%) (14). In our study, headache was the most common symptom in Chinese patients with RM. Focal neurological deficits, especially those of hyperacute onset (within several minutes), were striking symptoms and occurred in 67% of patients but were infrequent in patients with meningitis of other etiology. It is speculated that the underlying pathophysiology may involve cortical spreading depression induced by inflammation of the adjacent meninges (4). Meanwhile, epileptic seizures were also common symptoms and occurred in 50% patients. The presence of transient focal neurological deficits and epileptic seizures provide diagnostic clues for RM in patients with RA.

Patients with RM have some relatively specific neuroimaging features, which might be used as a diagnostic marker and help with differential diagnosis of other etiologies of meningitis. Our patients shared some remarkable neuroradiological manifestations (Figure 2), some have also been reported in other patients with RM (1, 15, 17, 18). First, RM can affect both the pachymeninges and leptomeninges, with the latter reported more frequently (60% vs 82.7%) (14). With disease progression, MRI might reveal involvement of the cerebral parenchyma. Repeated MRI in one patient showed dynamic changes from involvement of the meninges to involvement of the cerebral parenchyma. This propensity is different from that in some other rheumatology diseases, such as anti-neutrophil cytoplasm antibody-associated vasculitis and IgG4-related disease, in which involvement of the pachymeninges is predicted (19). Second, there is a hyperintensity signal (restricted diffusion) along the adjacent subarachnoid space on DWI (1, 15, 17, 18). In the previous studies, hyperintensity in the subarachnoid space on DWI was mainly observed in patients with bacterial or cryptococcal meningitis (20). However, all patients in our study had restricted diffusion on DWI. The CSF findings in patients with RM are markedly different from those in patients with bacterial or cryptococcal meningitis. Therefore, in patients with meningitis who show hyperintensity in the subarachnoid space on DWI, the CSF results will help to differentiate RM from bacterial meningitis and cryptococcal meningitis. Third, the lesions are located predominantly in the frontoparietal region, the convex surface of the brain, but spare the meninges around the basal cisterns (1, 15, 17, 18). A retrospective study by Parsons et al. showed that 12 (86%) of 14 patients had a frontoparietal predominance (11). Similarly, all patients in our study showed a frontoparietal predominance. This distribution is significantly different from that seen in tubercular meningitis, in which the meninges around the basal cisterns are usually affected. Fourth, involvement of the meninges can be unilateral or bilateral, when bilateral, there is usually lateral dominance. Unilateral involvement is more specific in RM. A retrospective study by Parsons et al. showed that asymmetric involvement was appreciated in 11 (78.6%) of 14 patients (11). Furthermore, three patients in this study showed asymmetric involvement. These neuroimaging features are helpful for the diagnosis of RM in patients with RA.

The evidence regarding treatment of RM is limited. It has been reported that CNS involvement occurs in patients with RA during immunotherapy (1, 9, 14, 21–25), such as corticosteroids (51%); non-biological DMTs—methotrexate, sulfasalazine, hydroxychloroquine, leflunomide, iguratimod, azathioprine, cyclosporine, bucillamine and tofacitinib (68%); biological DMTs—infliximab, etanercept and adalimumab (20%); and other agents (12%). However, RM has not been reported in patients on tocilizumab. In this study, RM occurred in four patients who showed active arthritis despite continuous corticosteroids and/or non-biological DMT for RA. IL-6 plays a role in induction and maintenance of the autoimmune process *via* B cell modulation and Th17 cell differentiation and in angiogenesis by upregulating the expression of intracellular adhesion molecules, which are important in the pathogenesis of RA (26). In patients with RA, high levels of IL-6/sIL-6R complex in synovial fluids are associated with joint destruction and disease progression (27). Tocilizumab is a monoclonal antibody that inhibits the IL-6 receptor, leading to inhibition of IL-6 signaling (28), and works rapidly and effectively in RA either as monotherapy or in combination with other agents (29–32). High IL-6 levels were also detected in the CSF of patients with RM in this study. Therefore, we speculated that tocilizumab might also be effective in the treatment of RM. In the previously reported cases, pulse corticosteroid therapy was the main induction therapy used for RM (14). Although application of tocilizumab in RM has rarely been reported, it has been used successfully alone or with methotrexate following corticosteroid therapy in four patients with RM (7, 22, 33).

However, use of tocilizumab as induction therapy has not been reported. In our study, two patients received tocilizumab alone, and one (case 5) received tocilizumab with low-dose corticosteroids as induction therapy; all experienced rapid improvement, suggesting that tocilizumab could be an effective induction therapy for RM. Meanwhile, three patients received tocilizumab with another immunosuppressive agents as maintenance therapy; all of these patients responded well to treatment. Therefore, tocilizumab might be an effective induction and maintenance therapy for RM.

RM is a severe neurological complication of RA. Disease relapse has been reported in 31.2% of patients and had a lethal outcome in 14% (14). In our study, no relapse was observed but one patient died of an opportunistic infection. Patients treated with biological agents should be closely monitored for infectious diseases.

This retrospective study has some limitations. First, no patient underwent meningeal biopsy for diagnosis. Second, some important evaluations, such as DAS-28 and laboratory tests were not performed in all patients. Third, the DMT regimen was not consistent across all the patients.

5 Conclusion

Chinese patients with RM share some striking clinical and neuroimaging features, including hyperacute focal neurological deficits, predominant involvement of the meninges in the frontoparietal region, and hyperintensity signals along the adjacent subarachnoid space on DWI. Patients with RM respond well to appropriate immunotherapy. Tocilizumab could be a promising option for induction and maintenance therapy in RM.

Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

Ethics statement

The studies involving human participants were reviewed and approved by institutional review board of Peking Union Medical College Hospital. Written informed consent for participation was not required for this study in accordance with the national legislation and the institutional requirements.

Author contributions

SF and JZ, design of the study, drafting and revising of the manuscript, major role in the acquisition of data, and analysis of the data. BH, major role in the acquisition of data, analysis of the data, and revising of the manuscript. MGL, major role in the acquisition of data, revising of the manuscript. JN, YZ, CM, HR, and YCZ, major role in the acquisition of data and analysis of the data. FF, MTL, and XZ, analysis of the data, revising of the manuscript. HG, design of the study, drafting and revising of the manuscript, and analysis of the data. All authors contributed to the article and approved the submitted version.

Funding

This study was supported by the CAMS Innovation Fund for Medical Sciences (CIFMS: 2021-I2M-C&T-A-002) and the National High Level Hospital Clinical Research Funding (2022-PUMCH-B-120).

Acknowledgments

The authors thanks Dr. Ke Li for his clinical work. We also thank Liwen Bianji for editing the English text.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Choi SJ, Ho Park Y, Kim JA, Han JH, Choe G, Kim S. Pearls & oy-sters: asymmetric meningeal involvement is a common feature of rheumatoid meningitis. *Neurology* (2017) 88(12):e108–e10. doi: 10.1212/wnl.0000000000003744

2. Luessi F, Sollors J, Müller H, Stoeter P, Sommer C, Vogt T, et al. Infliximab in the treatment of rheumatoid meningoencephalitis. *J Neurol* (2009) 256(12):2094–6. doi: 10.1007/s00415-009-5286-0

3. Matsushima M, Yaguchi H, Niino M, Akimoto-Tsuji S, Yabe I, Onishi K, et al. MRI And pathological findings of rheumatoid meningitis. *J Clin Neurosci* (2010) 17 (1):129–32. doi: 10.1016/j.jocn.2009.01.033

4. Bourgeois P, Rivest J, Bocti C. Rheumatoid meningitis presenting with stroke-like episodes. *Neurology* (2014) 82(17):1564–5. doi: 10.1212/wnl.0000000000000366

5. Roy B, Uphoff DF, Silverman IE. Rheumatoid meningitis presenting with multiple strokelike episodes. *JAMA Neurol* (2015) 72(9):1073–6. doi: 10.1001/jamaneurol.2015.1105

6. Akamatsu M, Maki F, Akiyama H, Hara D, Hoshino M, Hasegawa Y. Rheumatoid meningitis presenting with a stroke-like attack treated with recombinant tissue plasminogen activator: a case presentation. *BMC Neurol* (2018) 18(1):139. doi: 10.1186/s12883-018-1143-z

7. Schuster S, Braass H, Iking-Konert C, Schnoor U, Matschke J, Gerloff C, et al. Rheumatoid meningitis: a rare cause of aseptic meningitis with frequently stroke-like episodes. *Neurol Clin Pract* (2018) 8(5):451–5. doi: 10.1212/cpj.0000000000000504

8. Spinardi L, Muccioli L, Pastore Trossello M, Ciaffi J, Borlandelli E, Meliconi R, et al. Acute-onset focal neurological deficits in rheumatoid arthritis: consider rheumatoid meningitis. *Rheumatology* (2020) 59(11):3579. doi: 10.1093/rheumatology/keaa235

9. Starosta MA, Brandwein SR. Clinical manifestations and treatment of rheumatoid pachymeningitis. *Neurology* (2007) 68(13):1079–80. doi: 10.1212/01.wnl.0000257824.72457.91

10. Servioli MJ, Chugh C, Lee JM, Biller J. Rheumatoid meningitis. *Front Neurol* (2011) 2:84. doi: 10.3389/fneur.2011.00084

11. Parsons AM, Aslam F, Grill MF, Aksamit AJ, Goodman BP. Rheumatoid meningitis: clinical characteristics, diagnostic evaluation, and treatment. *Neurohospitalist* (2020) 10(2):88–94. doi: 10.1177/1941874419859769

12. Zhang LH, Wang XJ, Lin T, Jiao RY, Pang LX. Hypertrophic cranial pachymeningitis in rheumatoid arthritis: a case report. *Chin J Allergy Clin Immunol* (2017) 11(04):386–9. doi: 10.3969/j.issn.1673-8705.2017.04.014

13. Zheng RL, Lv H, Zhang W, Yu MX, Yuan Y. Rheumatoid meningitis: a case report. *J Peking Univ Health Sci* (2006) 03):324–5. doi: 10.3321/j.issn:1671-167X.2006.03.024

14. Villa E, Sarquis T, de Grazia J, Núñez R, Alarcón P, Villegas R, et al. Rheumatoid meningitis: a systematic review and meta-analysis. *Eur J Neurol* (2021) 28(9):3201–10. doi: 10.1111/ene.14904

15. Roques M, Tanchoux F, Calvière L, Cuinat L, Lubrano V, Uro-Coste E, et al. Mri with dwi helps in depicting rheumatoid meningitis. *J Neuroradiol* (2014) 41(4):275–7. doi: 10.1016/j.neurad.2013.10.005

16. Schmid L, Müller M, Treumann T, Arnold W, Möller B, Aeberli D, et al. Induction of complete and sustained remission of rheumatoid pachymeningitis by rituximab. *Arthritis Rheum* (2009) 60(6):1632–4. doi: 10.1002/art.24577

17. Koide R, Isoo A, Ishii K, Uruha A, Bandoh M. Rheumatoid leptomeningitis: rare complication of rheumatoid arthritis. *Clin Rheumatol* (2009) 28(9):1117–9. doi: 10.1007/s10067-009-1187-y

18. Hasiloglu ZI, Asik M, Erer B, Dikici AS, Altintas A, Albayram S. Magnetic resonance imaging of rheumatoid meningitis: a case report and literature review. *Rheumatol Int* (2012) 32(11):3679–81. doi: 10.1007/s00296-011-2105-6

19. Yonekawa T, Murai H, Utsuki S, Matsushita T, Masaki K, Isobe N, et al. A nationwide survey of hypertrophic pachymeningitis in Japan. *J Neurol Neurosurg Psychiatry* (2014) 85(7):732–9. doi: 10.1136/jnnp-2013-306410

20. Kawaguchi T, Sakurai K, Hara M, Muto M, Nakagawa M, Tohyama M, et al. Clinico-radiological features of subarachnoid hyperintensity on diffusion-weighted images in patients with meningitis. *Clin Radiol* (2012) 67(4):306–12. doi: 10.1016/j.crad.2011.10.001

21. Cavazzana I, Taraborelli M, Fredi M, Tincani A, Franceschini F. Aseptic meningitis occurring during anti-Tnf-Alpha therapy in rheumatoid arthritis and ankylosing spondylitis. *Clin Exp Rheumatol* (2014) 32(5):732–4.

22. Tsuzaki K, Nakamura T, Okumura H, Tachibana N, Hamano T. Rheumatoid meningitis occurring during etanercept treatment. *Case Rep Neurol Med* (2017) 2017:7638539. doi: 10.1155/2017/7638539

23. Huys AC, Guerne PA, Horvath J. Rheumatoid meningitis occurring during adalimumab and methotrexate treatment. *Joint Bone Spine* (2012) 79(1):90–2. doi: 10.1016/j.jbspin.2011.07.008

24. Finkelshtein V, Lampl Y, Lorberboym M, Kanner A, Ben-Ami Raichman D, Dabby R, et al. Self-limited rheumatoid meningitis as a presenting symptom of rheumatoid arthritis. *Isr Med Assoc J* (2018) 20(4):262–4.

25. Parsons AM, Zuniga LA, Hoxworth JM, Lyons M, Aslam F, Goodman BP. Rheumatoid meningitis: a case review. *Neurologist* (2018) 23(3):83–5. doi: 10.1097/nrl.0000000000000158

26. Kim GW, Lee NR, Pi RH, Lim YS, Lee YM, Lee JM, et al. Il-6 inhibitors for treatment of rheumatoid arthritis: past, present, and future. *Arch Pharm Res* (2015) 38(5):575–84. doi: 10.1007/s12272-015-0569-8

27. Ogata A, Kato Y, Higa S, Yoshizaki K. Il-6 inhibitor for the treatment of rheumatoid arthritis: a comprehensive review. *Mod Rheumatol* (2019) 29(2):258–67. doi: 10.1080/14397595.2018.1546357

28. Mihara M, Kasutani K, Okazaki M, Nakamura A, Kawai S, Sugimoto M, et al. Tocilizumab inhibits signal transduction mediated by both mil-6r and sil-6r, but not by the receptors of other members of il-6 cytokine family. *Int Immunopharmacol* (2005) 5(12):1731–40. doi: 10.1016/j.intimp.2005.05.010

29. Jones G, Ding C. Tocilizumab: a review of its safety and efficacy in rheumatoid arthritis. *Clin Med Insights Arthritis Musculoskelet Disord* (2010) 3:81–9. doi: 10.4137/cmamd.S4864

30. Singh JA, Beg S, Lopez-Olivo MA. Tocilizumab for rheumatoid arthritis: a cochrane systematic review. *J Rheumatol* (2011) 38(1):10–20. doi: 10.3899/jrheum.100717

31. Shetty A, Hanson R, Korsten P, Shawagfeh M, Arami S, Volkov S, et al. Tocilizumab in the treatment of rheumatoid arthritis and beyond. *Drug Des Devel Ther* (2014) 8:349–64. doi: 10.2147/dddt.S41437

32. Scott LJ. Tocilizumab: a review in rheumatoid arthritis. *Drugs* (2017) 77(17):1865–79. doi: 10.1007/s40265-017-0829-7

33. Qin Z, Kim J, Valencia D, Hamoodi L, Neltner J, Sizemore T, et al. Rheumatoid meningitis: a case report and review of the literature. *Neurol Clin Pract* (2020) 10(1):73–83. doi: 10.1212/cpj.0000000000000678

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 09 June 2023
DOI 10.3389/fneur.2023.1174116

OPEN ACCESS

EDITED BY
Beatrijs Wokke,
Erasmus Medical Center, Netherlands

REVIEWED BY
Marta Waliszewska-Prosół,
Wroclaw Medical University, Poland
Malik Ghannam,
University of Iowa Hospitals and Clinics, United States
Giacomo Cafaro,
University of Perugia, Italy

*CORRESPONDENCE
Stacey L. Clardy
✉ stacey.clardy@hsc.utah.edu

RECEIVED 25 February 2023
ACCEPTED 25 May 2023
PUBLISHED 09 June 2023

CITATION
Hoshina Y, Wong K-H, Galli J, Bacharach R, Klein J, Lebiedz-Odrobina D, Rose JW, Trump B, Hull C, Greenlee JE and Clardy SL (2023) Neurologic involvement in seronegative primary Sjögren's syndrome with positive minor salivary gland biopsy: a single-center experience.
Front. Neurol. 14:1174116.
doi: 10.3389/fneur.2023.1174116

Neurologic involvement in seronegative primary Sjögren's syndrome with positive minor salivary gland biopsy: a single-center experience

Yoji Hoshina[1], Ka-Ho Wong[1], Jonathan Galli[1,2], Rae Bacharach[1,3], Julia Klein[1], Dorota Lebiedz-Odrobina[4], John W. Rose[1,2], Bryan Trump[5], Christopher Hull[6], John E. Greenlee[1,2] and Stacey L. Clardy[1,2]*

[1]Department of Neurology, University of Utah, Salt Lake City, UT, United States, [2]George E. Wahlen Veterans Affairs Medical Center, Salt Lake City, UT, United States, [3]Department of Neurology, Penn State Health, Hershey, PA, United States, [4]Department of Medicine, Division of Rheumatology, University of Utah, Salt Lake City, UT, United States, [5]School of Dentistry, University of Utah, Salt Lake City, UT, United States, [6]Department of Dermatology, University of Utah, Salt Lake City, UT, United States

Objective: To assess the demographics, neurologic manifestations, comorbidities, and treatment of patients with seronegative primary Sjögren's syndrome (pSS).

Patients and methods: We conducted a retrospective chart review on patients with seronegative pSS evaluated by a neurologist at the University of Utah Health between January 2010 and October 2018. The diagnosis was based on characteristic symptoms, positive minor salivary gland biopsy according to the American-European Consensus Group 2002 criteria, and seronegative antibody status.

Results: Of 45 patients who met the study criteria, 42 (93.3%) were Caucasian, and 38 (84.4%) were female. The patients' mean age at diagnosis was 47.8±12.6 (range 13–71) years. Paresthesia, numbness and dizziness, and headache were noted in 40 (88.9%), 39 (86.7%), and 36 patients (80.0%), respectively. Thirty-four patients underwent brain magnetic resonance imaging. Of these, 18 (52.9%) showed scattered nonspecific periventricular and subcortical cerebral white matter T2/fluid-attenuated inversion recovery hyperintense foci. Twenty-nine patients (64.4%) presented to the neurology clinic prior to pSS diagnosis, and the median delay in diagnosis from the first neurology clinic visit was 5 (interquartile ranges 2.0–20.5) months. Migraine and depression were the most common comorbidities in 31 patients (68.9%). Thirty-six patients received at least one immunotherapy, and 39 were on at least one medication for neuropathic pain.

Conclusion: Patients often display various nonspecific neurological symptoms. Clinicians should express a high degree of skepticism regarding seronegative pSS and consider minor salivary gland biopsy to avoid delaying diagnosis, as undertreatment can affect patients' quality of life.

KEYWORDS

seronegative, primary Sjögren syndrome (pSS), nonspecific neurological symptoms, lip biopsy, neuropathic pain

1. Introduction

Primary Sjögren's syndrome (pSS) is a systemic autoimmune disorder, with a female predominance (9:1) and a peak incidence at approximately 50 years of age (1). It is commonly characterized by xerophthalmia and xerostomia, although other systemic and organ-specific manifestations may occur (1).

Neurologic involvement has been reported to range from 8.5 to 70% of pSS cases; this wide range is owing to the diverse diagnostic criteria of pSS, along with different definitions of neurologic symptoms and dissimilar availability of neurophysiological diagnostic testing resources (2–4). Although the prevalence of brain magnetic resonance imaging (MRI) abnormalities in patients with pSS remains unclear, MRI abnormalities have been reported with and without clinical evidence of central nervous system involvement (5). These include abnormalities typically visible on T2-weighted imaging (T2WI) or fluid-attenuated inversion recovery (FLAIR) sequences, raising concerns of possible multiple sclerosis (MS) (6). While neurologic involvement in pSS has been studied, it is still likely under-recognized, especially when initial symptoms lead to neurologic consultation prior to rheumatologic consultation or diagnosis. For example, neuropathic pain contributes to a delay in diagnosis, especially when sicca symptoms are mild (7), and neurologic manifestations may precede other pathognomonic Sjögren findings in 25–60% of cases with a mean delay of 24 months to pSS diagnosis (3).

Detection of anti-Ro/SSA and anti-La/SSB antibodies has long been used as the major diagnostic test for pSS and is included in the American-European Consensus Group 2002 criteria (8). However, these tests may be negative in 25–33% of patients (1, 9). pSS can also be diagnosed in the absence of anti-Ro/SSA or anti-La/SSB antibodies with positive histopathological evidence via minor salivary gland biopsy. In such cases, patients are diagnosed with seronegative pSS (7, 10, 11). Differences in clinical features between seropositive and seronegative populations are still controversial. For example, the Sjögren's Big Data Project of 10,500 patients reports that patients diagnosed with anti-Ro/SSA or anti-La/SSB antibodies have lower mean age at the time of diagnosis and a higher frequency of constitutional, renal, cutaneous, or hematological manifestations, which suggest that seronegative pSS may be a milder form of the disease (11). However, a more recent study comparing seronegative and seropositive patients reported similar demographic features between both groups (including age at diagnosis and sex distribution) and laboratory findings (except for antibodies) (7). None of these studies adequately characterize the neurologic manifestations observed in patients with seronegative pSS. Hence, we conducted this single-center study to assess the demographic and neurologic symptom profiles in this patient cohort.

2. Patients and methods

We performed an electronic database search of the University of Utah Health records between January 1, 2010, and October 31, 2018, to identify all patients with a positive minor salivary gland biopsy and seronegative antibody status, who were ultimately diagnosed with seronegative pSS, and were evaluated at least once by a neurologist. First, we extracted data for all patients diagnosed with SS using the International Classification of Diseases Clinical Modification, 9th and 10th revision codes 710.2 and M35.00. Thereafter, we reviewed their serological tests and clinic visit records. Patients who met all the following criteria were ultimately included in our analysis: (1) American-European Consensus Group criteria (8) for the diagnosis of pSS, (2) absence of anti-Ro/SSA and anti-La/SSB antibodies with a positive minor salivary gland biopsy result, and (3) at least one neurology clinic visit for any neurological complaint. All patients with SS associated with other established autoimmune diseases, such as that related to systemic lupus erythematosus or rheumatoid arthritis (RA), were excluded from the analysis. Positive minor salivary gland biopsy was confirmed based on the Chisholm and Mason classification (grades 1–4), and only grades 3 and 4 were included (12). Data collected for each patient included demographic information, clinical features focusing on neurologic symptoms, reason for neurology clinic visits, comorbidities, laboratory data, and radiologic data. Treatment and outcome data were also collected. The treatment outcome was assessed based on documentation during the follow-up appointment. The autonomic nervous system was assessed through a series of tests, including the quantitative sudomotor axon reflex test, heart rate response to the Valsalva maneuver and deep breathing, and blood pressure and heart rate response to the head-up tilt test. Cognitive impairment was defined as a Montreal Cognitive Assessment (MoCA) score ≤ 25.

A total of 286 patients were diagnosed with SS, of whom 94 tested negative for anti-Ro/SSA and anti-La/SSB antibodies. Of the seronegative patients, 56 patients presented to the neurology clinic with at least one neurologic symptom. Nine patients were excluded owing to a clinical diagnosis of pSS without minor salivary gland biopsy, and two patients were excluded owing to at least one other established rheumatologic condition (one patient had RA and the other patient had both RA and limited systemic sclerosis).

2.1. Statistical analysis

Values of quantitative variables are expressed as mean ± standard deviation or median and interquartile ranges (IQR), and values of qualitative variables are expressed as percentages. All statistical analyzes were performed using R Statistical Software (version 4.1.2; R Foundation for Statistical Computing, Vienna, Austria).

2.2. Standard protocol approvals, registrations, and patient consent

The study procedures were approved by the local institutional review board of the University of Utah (IRB_00108537).

3. Results

Forty-five patients were ultimately included in the analysis for the study. Table 1 summarizes the demographics of the patient population. The mean age was 47.8 ± 12.6 (range 13–71) years.

Abbreviations: FLAIR, fluid-attenuated inversion recovery; MRI, magnetic resonance imaging; MS, multiple sclerosis; pSS, primary Sjögren's syndrome; RA, rheumatoid arthritis; T2WI, T2-weighted imaging..

10.3389/fneur.2023.1174116

TABLE 1 Patient characteristics.

Characteristic	*n*	%
Sex		
Male	7	15.6
Female	38	84.4
Mean age at diagnosis of primary Sjögren's syndrome, years (range)	47.8 ± 12.6 (13–71)	
Median follow-up, months (interquartile ranges)	19 (3–46)	
Ethnicity		
Caucasian	42	93.3
African descent	2	4.4
Asian	1	2.2
Family history in first-degree relative		
Autoimmune conditions	12	26.7
Primary Sjögren's syndrome	2	4.4

TABLE 2 Neurologic clinical manifestations.

Neurological symptoms	*n* =45	%
Paresthesia	40	88.9
Numbness	39	86.7
Dizziness	39	86.7
Headache	36	80
Subjective cognitive decline	31	68.9
Subjective weakness	25	55.6
Imbalance	19	42.2
Neurological conditions diagnosed	***n*=45**	**%**
Peripheral neuropathy[a]	39	86.7
Small fiber neuropathy[b]	8	17.8
Dysautonomia[c]	10	22.2
Cognitive impairment[d]	12	26.7
Trigeminal neuralgia	3	6.7
Electroencephalogram abnormality[e]	2	4.4
Myelopathy[f]	1	2.2
Transient ischemic attack[g]	1	2.2

[a]Six patients had sensorimotor neuropathy and 33 had sensory neuropathy.
[b]Six patients showed positive skin biopsy. One patient was diagnosed based on clinical symptoms, evidence of small fiber neuropathy on a quantitative sensory test, and a normal nerve conduction test. One patient was diagnosed based on clinical symptoms, sudomotor dysfunction on autonomic tests, and a normal nerve conduction test.
[c]Ten patients were diagnosed using an autonomic test.
[d]Montreal Cognitive Assessment (MoCA) score ≤ 25/30. Of the 45 patients included in this study, 22 underwent MoCA testing.
[e]One patient had intermittent irregular delta activity in the left anterior-midtemporal region. One patient had intermittent focal theta to delta slowing in the left frontotemporal region.
[f]Cervical myelopathy with position sense loss.
[g]Likely due to a hypercoagulable state of unknown origin.

Thirty-eight patients (84.4%) were female, and 42 patients (93.3%) were Caucasian. Twelve patients (26.7%) had at least one first-degree relative with rheumatologic disease, including pSS in two cases (4.4%). All patients in this study had at least one of the sicca symptoms (40 patients had xerophthalmia, and 40 patients had xerostomia).

Neurologic manifestations are summarized in Table 2. The most common neurologic symptom was paresthesia (*n* = 40, 88.9%), followed by numbness and dizziness (n = 39, 86.7%), and headache (*n* = 36, 80.0%). Among the patients who had paresthesia, eight were diagnosed with small fiber neuropathy (six patients showed positive skin biopsy and two patients were diagnosed based on clinical symptoms, evidence of small fiber neuropathy on an electrophysiologic test, and normal nerve conduction test.) Among the 34 patients who underwent brain MRI during this period, 18 (52.9%) showed scattered T2WI/FLAIR hyperintensities in the periventricular and subcortical white matter, two of whom were eventually also diagnosed with MS, and one patient had a history of MS. Thirteen patients showed unremarkable results, and three patients showed other findings (one showed a finding of neurosarcoidosis, one showed global cortical atrophy, and one showed small punctuate focus of T2/FLAIR hyperintensity in both frontal lobes).

The reasons for neurology clinic referral are summarized in Table 3. The most common was neuropathy evaluation (*n* = 17, 37.8%), followed by consultations for abnormal MRI findings (n = 8, 17.8%), headache (*n* = 5, 11.1%), cognitive function (*n* = 5, 11.1%), and features of dysautonomia, particularly orthostatic symptoms (*n* = 4, 8.9%). One patient who was referred for myelopathy evaluation had position sense loss with electrophysiological evidence of spinal cord involvement. Of the eight patients who consulted for abnormal MRI findings, seven were referred to the neurology clinic with a concern of MS. Twenty-nine patients (64.4%) presented the neurology clinic prior to the diagnosis of seronegative pSS, with the median delay from the first neurology clinic presentation to the diagnosis being 5 (IQR 2.0–20.5) months.

The most common comorbidities observed were depression and migraine (*n* = 31, 68.9%). Twenty-one patients (46.7%) had been diagnosed as having fibromyalgia before presentation to our clinic, 13 patients (28.9%) had hypothyroidism, and 8 patients (17.8%) had irritable bowel syndrome.

Medication management is summarized in Table 4. Among the 45 examined patients, 36 had received at least one disease-modifying therapy. Thirty-one patients were on hydroxychloroquine, four were on steroids (one had a history of sarcoidosis, and one had a history of autoimmune hepatitis), two were on mycophenolate mofetil, and one was on methotrexate. Thirty-nine patients were on at least one medication for the treatment of neuropathic pain, including gabapentin (22 patients), duloxetine (20 patients), pregabalin (6 patients), venlafaxine (5 patients), baclofen (3 patients), and amitriptyline (1 patient). In addition, seven patients were on a selective serotonin reuptake inhibitor (three patients were on sertraline, two on fluoxetine, and one on escitalopram or citalopram). Thirty-seven patients had documented neuropathic symptoms at follow-up, of whom 25 (67.6%) had some symptomatic improvement after neuropathic medication initiation.

4. Discussion

We described the demographic, neurologic manifestations, and treatment of patients with seronegative pSS who presented to the neurology clinic. Our findings regarding age and female-to-male ratio

TABLE 3 Reasons for neurology clinic referral.

Neurological condition	*n* =45	%
Neuropathy	17	37.8
Magnetic resonance imaging abnormality	8	17.8
Headache	5	11.1
Cognitive dysfunction	5	11.1
Dysautonomia	4	8.9
Dystonia	1	2.2
Myelopathy	1	2.2
Seizure	1	2.2
Syncope	1	2.2
Transient ischemic attack	1	2.2
Multiple neurological conditions[a]	1	2.2

[a]Including bilateral hearing loss, right eye pain, weakness, dizziness, cognitive dysfunction, diffuse body pain, and headache.

TABLE 4 Medication management of seronegative primary Sjögren's syndrome with neurologic manifestations.

Treatment	*n* =45
Immunotherapy (total)[a]	36
Hydroxychloroquine	31
Immunosuppressant	7
Steroid	4
Mycophenolate mofetil	2
Methotrexate	1
Treatment of neuropathic pain (total)[b]	39
Gabapentin	22
Duloxetine	20
Pregabalin	6
Venlafaxine	5
Baclofen	3
Amitriptyline	1

[a]Two patients were on more than one immunotherapeutic agent.
[b]Seventeen patients were on more than one medication for neuropathic pain.

match previous reports (10); two patients had a family history of pSS. Although we could not find any genetic studies focusing on seronegative pSS, previous reports suggest a genetic predisposition to seropositive pSS with a complex mechanism involving both HLA and non-HLA genes (13). Most of these studies, however, discuss risk factors for the formation of anti-Ro/La antibodies, which may not apply to seronegative pSS. Further studies are needed to elucidate the link between seronegative pSS and genetic susceptibility.

In pSS, a variety of neurological problems have been reported (3–5), including peripheral neuropathy, which was the most common in our study, both from patients and as the documented reason for neurology referral (Tables 2, 3). While we did not compare seropositive and seronegative patients, other studies found that peripheral neuropathy was more prevalent in the seronegative group and had a greater impact on physical function outcomes (2, 9). Tani et al. (14) found that neuropathogenic effects mainly affected small nerve fibers instead of axon in seronegative pSS, which could explain the high frequency of paresthesia and small fiber neuropathy diagnosis in our patient cohort. The frequency of peripheral neuropathy in pSS patients ranges from 2 to 60%, depending on the detection methods used (5). The high proportion of neurologic conditions in our population is likely due to the study being conducted at a neurology clinic and including symptomatic patients regardless of quantitative assessments. Only 12 out of 31 patients reporting cognitive decline exhibited objective cognitive dysfunction. Previous research (15) showed that asymptomatic individuals with pSS displayed electrophysiological evidence of subtle cognitive dysfunction, while other studies indicated similar evidence of central nervous system dysfunction in asymptomatic patients with pSS using brainstem auditory evoked potentials and somatosensory evoked potentials (16, 17). This may explain part of the higher incidence of subjective neurologic clinical manifestations before a formal diagnosis is made.

MRI evaluation, particularly for concern of MS, was a common reason for neurology clinic referral. These concerns may be valid as pSS patients have a high prevalence of nonspecific T2WI/FLAIR white matter hyperintensities on brain MRI, even in the absence of focal neurologic symptoms (6, 18, 19). One study found that 49% of patients with pSS had white matter abnormalities, and 84% had multiple (≥3) lesions (19), which may lead to misdiagnosis of MS.

Several studies reported the relationship between migraine and pSS. Pal et al. (20) reported a significantly higher prevalence of migraine (46%) in patients with pSS. Escudero et al. (21) reported that the migraine-mimicking headache during pSS could be due to neurologic involvement and not just a comorbid migraine. Late-onset "migraine-like" episodes warrant evaluation to rule out pSS as a cause of headache. A previous study that demonstrated more severe physical function outcomes observed in seronegative pSS attributed the finding to concomitant fibromyalgia, which was more predominant in the seronegative pSS population (9). That study also reported that the fibromyalgia prevalence was twice as high in the seronegative group (33% vs. 17%). One large cohort study on the prevalence of fibromyalgia in pSS revealed a prevalence of 31% (22). Our study revealed that 46.7% of patients were documented to have fibromyalgia. This relatively high prevalence may reflect the symptom overlap between seronegative pSS and fibromyalgia, especially when patients have multiple symptomatic complaints, given that there is no specific laboratory examination to differentiate both conditions. Patients with seronegative pSS can be easily misdiagnosed or labeled with fibromyalgia and thus receive inappropriate treatment. Physicians should consider minor salivary gland biopsy in patients with suspected pSS, as serology can be negative in up to one-third of cases (1, 9).

Hydroxychloroquine is a commonly used immunomodulatory medication for musculoskeletal/joint pain associated with pSS (23). Although a randomized trial showed no significant improvement in pSS symptoms during 24 weeks of treatment (24), it is still commonly used as the first-line medication owing to its good safety profile and minimal side effects. All 40 patients with paresthesia received neuropathic pain medications, most commonly anticonvulsant calcium channel alpha 2-delta ligands, and serotonin-norepinephrine reuptake inhibitors for comorbid mood symptoms. Only seven patients received selective serotonin reuptake inhibitors for depression, highlighting the dual benefit of serotonin-norepinephrine reuptake inhibitors in managing chronic neuropathic pain, consistent with recommendations (25). Tricyclic

antidepressants, due to anticholinergic side effects, were avoided and only used in one patient who had an allergic reaction to duloxetine. Symptomatic therapy for neuropathic pain led to decreased pain symptoms in 67% of patients, indicating the need for its consideration in seronegative pSS patients with neuropathic symptoms.

Our study has some limitations. It includes data from a single center with a high Caucasian predominance, reflective of the regional referral area's racial and ethnic composition. Although the influence of race and ethnicity in this condition is still not well understood, one study from the Big Data Sjögren Project Consortium (an international multicenter registry that included 7,884 patients) also revealed a Caucasian predominance at 78.3% (10). Dedicated studies to determine if this accurately reflects racial/ethnic distribution or if other racial/ethnic groups are simply under-represented owing to healthcare disparities are beyond the scope of this retrospective review but are much needed in the future. Because this study was conducted in neurology clinics, our population had a high proportion of patients with neurologic complaints with a greater variety of neurologic symptoms than previously reported. While this study may not allow us to determine the frequency of neurologic complications in a general population, it aimed to characterize the clinical features in patients with seronegative pSS with neurologic involvement. Our study thus highlights the diversity of neurologic conditions seen in seronegative pSS and the importance of recognizing these manifestations, given their effect on patients' quality of life. Owing to the nature of the retrospective chart review, incomplete medical chart documentation can affect the ability to comprehensively interpret the data; for example, one patient had a medical history of type 2 diabetes mellitus (no documented diabetic neuropathy), and four patients had a medical history of Hashimoto's disease (data not presented). Similarly, the evaluation of treatment efficacy relied solely on the documentation provided by the treating physicians, with no utilization of standardized measurement tools, such as scales or scores. In comparison to existing literature, this study is among the largest to characterize the neurologic symptoms observed in patients with seronegative pSS and their response to treatment. Prospective clinical trials to evaluate neurologic symptoms and treatment outcomes in a multicenter setting would be more informative. Finally, the sensitivity of minor salivary gland biopsy as a diagnostic test varies between 60 and 86% (26, 27), and this represents an additional limitation. The specificity of minor salivary gland biopsy has, however, been reported to be relatively high (91–97%), making it useful for diagnosing pSS, especially in seronegative patients (26, 27). Our study included patients with Chisholm and Mason classification grades 3–4, which supports the diagnosis of seronegative pSS.

In summary, neurologic manifestations of seronegative pSS are heterogeneous and may precede or overshadow sicca symptoms, leading to difficulties in diagnosis, especially early in the course. Clinicians should be aware of the range of presentations of seronegative pSS and maintain a low threshold to perform diagnostic assessments, including minor salivary gland biopsy, especially since pSS without antibody positivity is common. It is also important to consider non-neurologic signs and symptoms—including sicca and joint pain—when evaluating patients with neuropathy to ensure that physicians do not miss a multisystemic disease such as pSS. This study also highlights that symptomatic therapy for neurologic symptoms in seronegative pSS is essential, as immunomodulatory therapy in isolation is rarely sufficient to manage symptoms and improve patient quality of life.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

YH: data acquisition, manuscript drafting, data analysis, and interpretation of the results. K-HW: data acquisition, conceptualization of the study, data analysis, interpretation of the results, and revising the manuscript for intellectual content. JGa, RB, JK, DL, JR, BT, CH, and JGr: revising the manuscript for intellectual content. SC: conceptualization of the study, data analysis, interpretation of the results, and revising the manuscript for intellectual content. All authors contributed to the article and approved the submitted version.

Acknowledgments

The authors thank Barbara Steinmetz Gural and the Siegal Rare Neuroimmune Association for their continued support.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Mariette X, Criswell LA. Primary Sjögren's syndrome. *N Engl J Med*. (2018) 378:931–9. doi: 10.1056/NEJMcp1702514

2. Yazisiz V, Aslan B, Erbasan F, Uçar İ, Öğüt TS, Terzioğlu ME. Clinical and serological characteristics of seronegative primary Sjögren's syndrome: a comparative study. *Clin Rheumatol*. (2021) 40:221–9. doi: 10.1007/s10067-020-05154-9

3. Fauchais AL, Magy L, Vidal E. Central and peripheral neurological complications of primary Sjögren's syndrome. *Presse Med*. (2012) 41:e485–93. doi: 10.1016/j.lpm.2012.06.002

4. Chai J, Logigian EL. Neurological manifestations of primary Sjogren's syndrome. *Curr Opin Neurol*. (2010) 23:509–13. doi: 10.1097/WCO.0b013e32833de6ab

5. Perzyńska-Mazan J, Maślińska M, Gasik R. Neurological manifestations of primary Sjögren's syndrome. *Reumatologia*. (2018) 56:99–105. doi: 10.5114/reum.2018.75521

6. Pierot L, Sauve C, Leger JM, Martin N, Koeger AC, Wechsler B, et al. Asymptomatic cerebral involvement in Sjögren's syndrome: MRI findings of 15 cases. *Neuroradiology*. (1993) 35:378–80. doi: 10.1007/BF00588375

7. Soliotis FC, Mavragani CP, Moutsopoulos HM. Central nervous system involvement in Sjogren's syndrome. *Ann Rheum Dis*. (2004) 63:616–20. doi: 10.1136/ard.2003.019497

8. Vitali C, Bombardieri S, Jonsson R, Moutsopoulos HM, Alexander EL, Carsons SE, et al. Classification criteria for Sjögren's syndrome: a. rev version of the European criteria proposed by the American-European consensus group. *Ann Rheum Dis*. (2002) 61:554–8. doi: 10.1136/ard.61.6.554

9. Segal BM, Pogatchnik B, Henn L, Rudser K, Sivils KM. Pain severity and neuropathic pain symptoms in primary Sjögren's syndrome: a comparison study of seropositive and seronegative Sjögren's syndrome patients. *Arthritis Care Res (Hoboken)*. (2013) 65:1291–8. doi: 10.1002/acr.21956

10. Brito-Zerón P, Acar-Denizli N, Zeher M, Rasmussen A, Seror R, Theander E, et al. Influence of geolocation and ethnicity on the phenotypic expression of primary Sjögren's syndrome at diagnosis in 8310 patients: a cross-sectional study from the big data Sjögren project consortium. *Ann Rheum Dis*. (2017) 76:1042–50. doi: 10.1136/annrheumdis-2016-209952

11. Brito-Zerón P, Acar-Denizli N, Ng WF, Zeher M, Rasmussen A, Mandl T, et al. How immunological profile drives clinical phenotype of primary Sjögren's syndrome at diagnosis: analysis of 10,500 patients (Sjögren big data project). *Clin Exp Rheumatol*. (2018) 36 Suppl 112:102–12.

12. Chisholm DM, Mason DK. Labial salivary gland biopsy in Sjogren's disease. *J Clin Pathol*. (1968) 21:656–60. doi: 10.1136/jcp.21.5.656

13. Peri Y, Agmon-Levin N, Theodor E, Shoenfeld Y. Sjögren's syndrome, the old and the new. *Best Pract Res Clin Rheumatol*. (2012) 26:105–17. doi: 10.1016/j.berh.2012.01.012

14. Tani J, Liao HT, Hsu HC, Chen LF, Chang TS, Shin-Yi Lin C, et al. Immune-mediated axonal dysfunction in seropositive and seronegative primary Sjögren's syndrome. *Ann Clin Transl Neurol*. (2020) 7:819–28. doi: 10.1002/acn3.51053

15. Dziadkowiak E, Sebastian A, Wiland P, Waliszewska-Prosół M, Wieczorek M, Zagrajek M, et al. Endogenous event-related potentials in patients with primary Sjögren's syndrome without central nervous system involvement. *Scand J Rheumatol*. (2015) 44:487–94. doi: 10.3109/03009742.2015.1032345

16. Waliszewska-Prosół M, Sebastian A, Wiland P, Budrewicz S, Dziadkowiak E, Ejma M. Brainstem auditory evoked potentials in patients with primary Sjögren's syndrome without central nervous system involvement. *Clin Rheumatol*. (2021) 40:991–7. doi: 10.1007/s10067-020-05344-5

17. Dziadkowiak E, Sebastian A, Wieczorek M, Kusińska E, Waliszewska-Prosół M, Wiland P, et al. Parameters of somatosensory evoked potentials in patients with primary Sjögren's syndrome: preliminary results. *J Immunol Res*. (2018) 2018:8174340. doi: 10.1155/2018/8174340

18. Morgen K, McFarland HF, Pillemer SR. Central nervous system disease in primary Sjogren's syndrome: the role of magnetic resonance imaging. *Semin Arthritis Rheum*. (2004) 34:623–30. doi: 10.1016/j.semarthrit.2004.07.005

19. Akasbi M, Berenguer J, Saiz A, Brito-Zerón P, Pérez-De-Lis M, Bové A, et al. White matter abnormalities in primary Sjögren syndrome. *Q J Med*. (2012) 105:433–43. doi: 10.1093/qjmed/hcr218

20. Pal B, Gibson C, Passmore J, Griffiths ID, Dick WC. A study of headaches and migraine in Sjögren's syndrome and other rheumatic disorders. *Ann Rheum Dis*. (1989) 48:312–6. doi: 10.1136/ard.48.4.312

21. Escudero D, Latorre P, Codina M, Coll-Cantí J, Coll J. Central nervous system disease in Sjögren's syndrome. *Ann Med Interne (Paris)*. (1995) 146:239–42.

22. Choi BY, Oh HJ, Lee YJ, Song YW. Prevalence and clinical impact of fibromyalgia in patients with primary Sjögren's syndrome. *Clin Exp Rheumatol*. (2016) 34:S9–S13.

23. Ramos-Casals M, Brito-Zerón P, Bombardieri S, Bootsma H, De Vita S, Dörner T, et al. EULAR recommendations for the management of Sjögren's syndrome with topical and systemic therapies. *Ann Rheum Dis*. (2020) 79:3–18. doi: 10.1136/annrheumdis-2019-216114

24. Gottenberg JE, Ravaud P, Puéchal X, Le Guern V, Sibilia J, Goeb V, et al. Effects of hydroxychloroquine on symptomatic improvement in primary Sjögren syndrome: the JOQUER randomized clinical trial. *JAMA*. (2014) 312:249–58. doi: 10.1001/jama.2014.7682

25. Moulin D, Boulanger A, Clark AJ, Clarke H, Dao T, Finley GA, et al. Pharmacological management of chronic neuropathic pain: revised consensus statement from the Canadian Pain Society. *Pain Res Manag*. (2014) 19:328–35. doi: 10.1155/2014/754693

26. Pijpe J, Kalk WW, van der Wal JE, Vissink A, Kluin PM, Roodenburg JLN, et al. Parotid gland biopsy compared with labial biopsy in the diagnosis of patients with primary Sjogren's syndrome. *Rheumatol (Oxf Engl)*. (2007) 46:335–41. doi: 10.1093/rheumatology/kel266

27. Giovelli RA, Santos MC, Serrano ÉV, Valim V. Clinical characteristics and biopsy accuracy in suspected cases of Sjögren's syndrome referred to labial salivary gland biopsy. *BMC Musculoskelet Disord*. (2015) 16:30. doi: 10.1186/s12891-015-0482-9

frontiers | Frontiers in Neurology

TYPE Review
PUBLISHED 22 June 2023
DOI 10.3389/fneur.2023.1099758

OPEN ACCESS

EDITED BY
Tatjana Pekmezovic,
University of Belgrade, Serbia

REVIEWED BY
Vincent Van Pesch,
Université Catholique de Louvain, Belgium
Abhay Ranjan,
Indira Gandhi Institute of Medical Sciences, India
Mattia Fonderico,
University of Florence, Italy
Karlo Toljan,
Cleveland Clinic, United States

*CORRESPONDENCE
Tamar Harel
✉ tamar.harel@va.gov;
✉ tharel@som.umaryland.edu

RECEIVED 16 November 2022
ACCEPTED 26 May 2023
PUBLISHED 22 June 2023

CITATION
Harel T, Gorman EF and Wallin MT (2023) New onset or relapsing neuromyelitis optica temporally associated with SARS-CoV-2 infection and COVID-19 vaccination: a systematic review.
Front. Neurol. 14:1099758.
doi: 10.3389/fneur.2023.1099758

New onset or relapsing neuromyelitis optica temporally associated with SARS-CoV-2 infection and COVID-19 vaccination: a systematic review

Tamar Harel[1,2]*, Emily F. Gorman[3] and Mitchell T. Wallin[1,2]

[1]Department of Veterans Affairs Multiple Sclerosis Center of Excellence (VA MSCoE), Baltimore VA Medical Center, Baltimore, MD, United States, [2]Department of Neurology, University of Maryland Medical Center, Baltimore, MD, United States, [3]Health Sciences and Human Services Library, University of Maryland, Baltimore, MD, United States

Background: Neuromyelitis optica spectrum disorder (NMOSD) is a rare chronic neuroinflammatory autoimmune condition. Since the onset of the COVID-19 pandemic, there have been reports of NMOSD clinical manifestations following both SARS-CoV-2 infections and COVID-19 vaccinations.

Objective: This study aims to systematically review the published literature of NMOSD clinical manifestations associated with SARS-CoV-2 infections and COVID-19 vaccinations.

Methods: A Boolean search of the medical literature was conducted between December 1, 2019 to September 1, 2022, utilizing Medline, Cochrane Library, Embase, Trip Database, Clinicaltrials.gov, Scopus, and Web of Science databases. Articles were collated and managed on Covidence® software. The authors independently appraised the articles for meeting study criteria and followed PRISMA guidelines. The literature search included all case reports and case series that met study criteria and involved NMOSD following either the SARS-CoV-2 infection or the COVID-19 vaccination.

Results: A total of 702 articles were imported for screening. After removing 352 duplicates and 313 articles based on exclusion criteria, 34 articles were analyzed. A total of 41 cases were selected, including 15 patients that developed new onset NMOSD following a SARS-CoV-2 infection, 21 patients that developed *de novo* NMOSD following COVID-19 vaccination, 3 patients with known NMOSD that experienced a relapse following vaccination, and 2 patients with presumed Multiple Sclerosis (MS) that was unmasked as NMOSD post-vaccination. There was a female preponderance of 76% among all NMOSD cases. The median time interval between the initial SARS-CoV-2 infection symptoms and NMOSD symptom onset was 14 days (range 3–120 days) and the median interval between COVID-19 vaccination and onset of NMO symptoms was 10 days (range 1 to 97 days). Transverse myelitis was the most common neurological manifestation in all patient groups (27/41). Management encompassed acute treatments such as high dose intravenous methylprednisolone, plasmapheresis, and intravenous immunoglobulin (IVIG) and maintenance immunotherapies. The majority of patients experienced a favorable outcome with complete or partial recovery, but 3 patients died.

Conclusion: This systematic review suggests that there is an association between NMOSD and SARS-CoV-2 infections and COVID-19 vaccinations. This association requires further study using quantitative epidemiological assessments in a large population to better quantify the risk.

KEYWORDS

neuromyelitis optica, COVID-19, severe acute respiratory syndrome coronavirus 2, COVID-19 vaccine, outcomes

Introduction

Novel coronavirus disease (COVID-19), a respiratory disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), was first detected in Wuhan, China in December 2019, and by March 2021 the World Health Organization (WHO) declared a worldwide pandemic (1). As of November 2, 2022, globally there have been over 628 million confirmed cases of COVID-19, including over 6.57 million deaths, and over 12.85 billion doses of the vaccine have been administered (2). Despite the rapid development and distribution of vaccinations, COVID-19 remains a prevalent and serious public health condition today.

SARS-CoV-2 has the ability to dysregulate the host immune system, producing various autoantibodies (3–5). This can induce a cascade of immune-mediated central nervous system (CNS) damage from either direct inoculation of the CNS or a systemic autoimmune response toward the virus (3–6). It has been shown that SARS-CoV2 can traverse the blood brain barrier and provoke CNS demyelination (3). Given this background, it is not surprising that a variety of case reports have linked the SARS-CoV-2 infection with an array of CNS autoimmune demyelinating disorders such as transverse myelitis (TM), acute demyelinating encephalomyelitis (ADEM), multiple sclerosis (MS), and neuromyelitis optica spectrum disorder (NMOSD) (6–8).

NMOSD is a chronic, relapsing, autoantibody-mediated astrocytopathy channelopathy that presents as severe CNS demyelination attacks commonly involving TM, optic neuritis (ON), area postrema syndrome (APS), and acute brainstem syndrome (BS) (9). The underlying pathogenic mechanism that leads to NMOSD is unclear, but mounting evidence suggests that there is an intricate interplay between environmental factors, such as vaccines and viral infection, and genetic susceptibility that leads to CNS inflammation (10–12).

As COVID-19 is likely to remain a prevalent infectious disease, it is essential that we elucidate the association between this SARS-CoV-2 infections and neuroinflammatory conditions such as NMOSD. Through this systematic review, we will assess the association between SARS-CoV-2 infections and the para and post-infectious manifestations of NMOSD. We will also investigate the potential association between COVID-19 vaccination and the development of *de novo* or relapsing NMOSD.

Methods

Design

Literature was retrieved from the following databases on September 13, 2022: Medline (Ovid), Cochrane Library (WileyOnline), Embase (Elsevier), Trip Database Pro, Clinicaltrials.gov, and Scopus (Elsevier). This systematic review was carried out in accordance with Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) guidelines. We aimed to identify relevant articles reporting on NMOSD manifestations following a SARS-CoV-2 infection or a *de novo* or relapsing forms of NMOSD presenting in association with any type of approved COVID-19 vaccine.

Search strategy

The search strategy combined keywords and controlled vocabulary related to NMOSD and COVID-19 and was tailored to the specifications of each database (see Supplementary appendix 1). A detailed search strategy can be found in Supplementary material. A manual search of bibliographies of relevant studies was also conducted. All citations for this review were required to be indexed in the peer-reviewed literature. Results were carefully verified to avoid duplicates or overlapping publications.

Inclusion criteria

We identified and triaged manuscripts and included all peer-reviewed, full-text, English language manuscripts that reported cases of NMOSD that met the 2015 International Panel for NMOSD Diagnosis (IPND) criteria in association with SARS-CoV-2 infection or a COVID-19 vaccination (13).

Exclusion criteria

The review was restricted to studies published in English. Poster and symposium abstracts, non-peer reviewed publications, and clinical trials were excluded from this report. We also excluded review papers, editorial, hypothesis reports, and commentaries, unless there was a report of a case of NMOSD following a SARS-CoV-2 infection or COVID-19 vaccine. Studies were also excluded if they contained insufficient clinical data, if the data was repeated from an article that had already been included, or if they addressed peripheral nervous system (PNS) demyelinating diseases or CNS demyelinating disorders other than NMOSD such as myelin oligodendrocyte glycoprotein antibody disease (MOGAD), TM, ON, MS, and acute disseminated encephalomyelitis (ADEM). Cases involving other types of coronaviruses (e.g., SARS-CoV/MERS-CoV) infections were also excluded.

Data extraction

Titles and abstracts of all identified studies were independently screened for relevance by two reviewers, MW and TH, to ensure they met criteria for inclusion. Following a full-text screening of eligible

articles, articles meeting criteria were retrieved, summarized, and managed on Covidence® software. Discordant abstract or article decisions and screening queries were resolved by consensus. The same reviewers then extracted data on the following parameters: article title, authors, publication year, country, age/gender of the patients, aquaporin-4 (AQP4) antibody status, SARS-CoV-2 infection presentation, NMOSD clinical presentation, COVID-19 vaccine related information, interval prior to onset of neurological symptoms, MRI findings, cerebrospinal fluid (CSF) analysis, SARS-COV-2 laboratory findings, treatment, and clinical outcome.

Statistical analysis

Quantitative data were described using range (minimum and maximum), mean and median, while qualitative data were described in percentages and numbers. Covidence software was used for evaluating and adjudicating articles for the systematic review and Microsoft Excel was used for statistical assessments.

Results

As seen in the PRISMA flowchart (Figure 1), our systematic search identified 702 potentially relevant articles through various databases. A total of 354 duplicate articles were discarded. The remaining 348 articles were screened by title and abstract, and 249 non peer reviewed or nonrelevant articles were removed. Thereafter, a total of 34 studies were deemed eligible by the authors after applying the inclusion/exclusion criteria to the full text documents, of which there were 24 single-case reports, 9 case-series, and 1 prospective cohort study. These 34 reports described 41 unique patients which were divided into three categories: NMOSD onset following a SARS-CoV-2 infection, NMOSD onset following COVID-19 vaccination, and relapses consistent with NMOSD following COVID-19 vaccination. The clinical characteristics for each of these categories are presented in Tables 1–4 which summarizing the demographic and clinical characteristics of patients with SARS-CoV-2 post-infection and COVID-19 post-vaccination NMOSD manifestations.

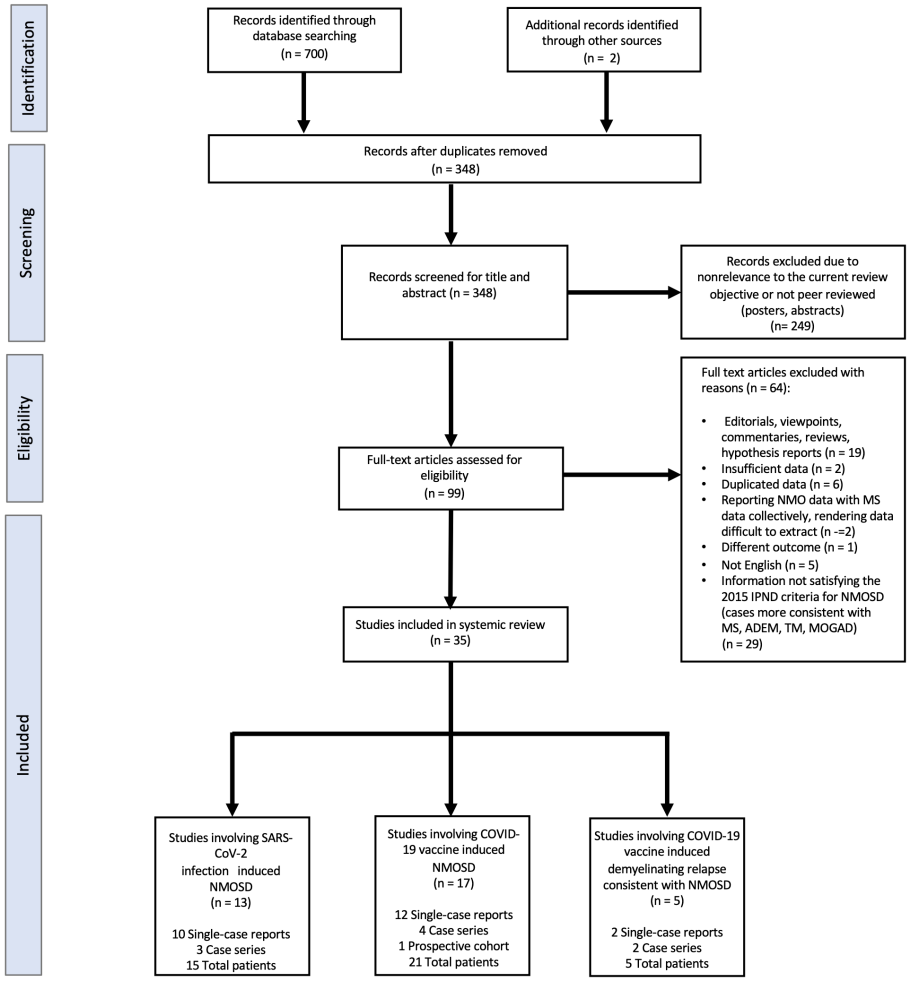


FIGURE 1
Flow chart of literature inclusion in accordance with PRISMA guidelines. ADEM, Acute Demyelinating Encephalomyelitis; IPND, International Panel for NMO Diagnosis; MOGAD, Myelin oligodendrocyte glycoprotein antibody-associated disease; MS, Multiple Sclerosis; NMOSD, Neuromyelitis Optica Spectrum Disorder; SARS-CoV-2, Severe Acute Respiratory Syndrome Coronavirus-2; TM, Transverse Myelitis.

New onset NMOSD following SARS-CoV-2 infection

Of the 15 patients that developed NMOSD following SARS-CoV-2 infection, 11 were female (73%), 3 were male (20%), and one was not identified (7%). The reported cases came from 12 countries; 2 cases each from France, India, and the United States of America (USA), and 1 case each from Italy, Korea, Pakistan, Qatar, Turkey, the United Arab Emirates, Australia, Brazil, and Egypt. Given a total of 626,337,158 world-wide COVID-19 cases as of October 31, 2022 (2), the global incidence based on reported cases of NMOSD following a SARS-CoV-2 infection is 0.02 per million.

The median age of the patients was 37.5 years (range 7.5–71 years). The latency period from the onset of COVID-19 symptoms to the first neurological manifestations followed a dual distribution: (i) *Short latency*: 3 to 14 days in 8/15 patients (53%) and (ii) *Long latency* (60 to 120 days) in 3/15 patients (20%). The median time interval between the initial SARS-CoV-2 infection symptoms and NMOSD symptom onset was 14 days (range 3–120 days).

Interestingly, 2/15 (13%) of patients had a history of a previously diagnosed immune-mediated condition; one patient had juvenile arthritis, and the other patient had a past episode of suspected ADEM. Comorbidities were present in 6/15 patients (40%) and are summarized with other clinical characteristics in Table 1.

In terms of the clinical presentation, TM was the most common neurological phenotype occurring in 10/15 (67%) patients. Two of the 10 had short-segment TM (STM) spanning over less than 3 vertebral segments and 8 were longitudinally extensive TM (LETM) spanning 3 or more vertebral segments. The second most common presentation was ON, found in 7 (47%) patients. APS, defined as intractable nausea, vomiting, or hiccups persisting for at least 48 h, was found in 2 (13%) patients. Brainstem involvement was found in 5 (33%). Ten patients (67%) tested positive for AQP4 antibody, while 4 (27%) were AQP4 antibody negative (one case not reported). CSF analysis in this group demonstrated pleocytosis in 5/15 (33%) patients while 2/15 (13%) had normal white blood cell (WBC) counts. CSF findings were not reported for 8/15 (53%) patients. High protein levels were reported in 2/15 (13%) patients.

Of the 13 cases that reported on acute treatment, all but one patient (92%) was initially treated with intravenous methylprednisolone. In additional to methylprednisolone, 5/13 (39%) were treated with plasmapheresis and 3/13 (23%) were treated with intravenous immunoglobulin (IVIG). Maintenance immunotherapy was provided to only four patients, including rituximab ($n = 3$) and azathioprine ($n = 1$). The treatment outcomes were reported for 13 of the 15 patients. Of these patients, 11/13 (84%) experienced complete or partial recovery following treatment, while 2/13 (15%) patients died. One death was caused by multiorgan failure and sepsis secondary to the SARS-CoV-2 infection. The second patient died from respiratory insufficiency, lymphopenia, and fever following cyclophosphamide treatment.

New onset and relapsing NMOSD following COVID-19 vaccination

Tables 2, 3 describe the clinical presentation, laboratory and imaging findings, and treatment outcomes of both *de novo* and relapsing NMOSD cases following the COVID-19 vaccine.

After receiving a COVID-19 vaccination, 26 patients developed a new demyelinating event related to NMOSD. A total of 21 of the 26 (81%) cases experienced an initial relapse of NMOSD following the COVID-19 vaccination, while 5 of the 26 (19%) cases had a recurrent exacerbation attributed to NMOSD following vaccination. Of the 5 relapsing cases, 3 of the patients had a known diagnosis of NMOSD, while two patients had been initially diagnosed with MS, which was unmasked as NMOSD post vaccination. Of note, one of the patients with known NMOSD had been stable and relapse free for 8 years, prior to their vaccine inducing a new relapse.

Based on data from the WHO, a total of 12,830,378,906 vaccine doses have been administered globally as of October 31, 2022 (2), the global incidence of an NMOSD demyelinating events among reported cases in the literature following vaccination is 0.002 per million.

Of the 26 cases developing NMOSD manifestations following a COVID-19 vaccination, 9 cases (35%) occurred after receiving the Pfizer-BioNTech BNT162b2 mRNA vaccine, 6 (23%) following the Oxford–AstraZeneca ChAdOx1 nCoV-19 viral vector vaccine, 5 (19%) following the Moderna mRNA-1273 vaccine, 4 (15%) after the Sinovac or Sinopharm inactivated COVID-19 vaccine, and 1 (4%) following the Sputnik V adenovirus viral vector vaccine. The specific vaccine involved in one case was unspecified, but it was a viral vector vaccine. In sum, 54% (14/26) of cases involved an mRNA vaccine, 31% (8/26) of cases involved a viral vector vaccine, and 15% (4/26) of cases involved an inactivated COVID-19 vaccine.

In terms of demographics, there was a female preponderance with a 3.3:1 ratio of female to male cases. The median age was 50 years with an age range of 19 to 80. The reported cases came from 13 countries with 4 cases each from Thailand and the USA, 3 cases each from Germany and Korea, 2 cases each from Italy and Turkey, and 1 case each from Brazil, Canada, China, France, India, Iran, Saudi Arabia, and the United Arab Emirates.

The median duration between vaccination and onset of NMOSD related clinical symptoms was 10 days (range 1–97 days). Figure 2, displays time intervals between vaccination and neurological symptom onset for each COVID-19 vaccine. Breaking down symptom onset with dose of the vaccine, 15/26 (58%) patients, experienced the onset of neurological symptoms following the first dose of the vaccine. A total of 6/26 (23%) patients had the onset of neurological symptoms following the second dose of the vaccine, and in 2/26 (8%) patients, the onset of neurological symptoms followed the third dose of the vaccine. One case did not specify, which dose induced the neurological symptoms.

Interestingly, 8/26 (31%) patients had a history of a previously diagnosed immune-mediated condition. In addition, 4/26 (15%) patients reported a family history of an immune-mediated condition including MS, AQP4-IgG positive NMOSD, myasthenia gravis, and systemic lupus erythematous.

Turning to the clinical presentation, TM was the most common phenotype, occurring in 17 (65%) patients. Four of those were STM spanning over less than 3 vertebral segments and 13 were LETM spanning 3 or more vertebral segments. The second most common presentation was ON, found in 5 (19%) patients. APS was found in 3 (12%) patients and brainstem involvement was found in 3 (12%) patients.

Of the 25 patients with a reported AQP4 antibody status, 22/25 (88%) patients tested positive for AQP4 antibody, while 3 (12%) were AQP4 antibody negative. Of the 20 patients with reported CSF results

TABLE 1 Characteristics of cases presenting with Neuromyelitis Optica in relation to a SARS-CoV-2 infection.

Reference/ country	Age/ sex	Comorbidities	Clinical presentation of COVID-19 infection	Clinical presentation of NMO	AQP4 antibody status*	Laboratory investigations	Time interval between COVID-19 and NMO (days)	MRI data	CSF findings	COVID-19 related findings	Treatment of NMO	Outcome
1. Aubart et al. (14) France	14/F	Juvenile arthritis	Asymptomatic	Monocular optic neuritis	+ (AQP4 Ab test not specified)	NR	NR	*Optic Nerves:* Optic Neuritis *Brain:* Spared Spine: Spared	NR	Positive SARS-CoV-2 nasal PCR	IVMP	Improvement
2. Barone et al. (15) Italy	35/M	None	NR	Monocular optic neuritis Myalgias	+ (AQP4 Ab test not specified)	ANA 1:640 Anti-TPO > 1,300 U/mL	30	*Optic Nerves:* Enhancing left optic nerve and optic chiasm lesion *Brain:* Spared Spine: Spared	NR	Negative Positive SARS-CoV-2 nasal PCR Positive serological IgG/IgM	IVIM IVIG Rituximab	Complete recovery
3. Batum et al. (16) Turkey	50/F	None	Fever Cough	Numbness, Urinary retention Weakness	+ (CSF AQP4-IgG)	Anti-CMV IgM negative, Brucella agglutination negative, EBV IgM negative, Anti-HAV IgM negative, Anti-HBc IgM negative, HIV negative, RF negative, ANA negative, ANCA negative, anti-mitochondrial antibody negative, Anti-smooth muscle antibody negative, Anti-Ro negative, Anti-La negative, Anti-ds DNA negative, Anti-nRNP negative, anti-Histon antibody negative, anti-MOG negative	NR	*Brain:* Spared Spine: LETM from C3 to Conus	Pleocytosis Protein 159 mg/dL OCB negative IgG index 1.2	Negative SARS-CoV-2 nasal PCR CXR: Bilateral Consolidation with ground-glass density	IVMP IVIG PLEX	Improvement
4. Correa et al. (7) Brazil	51/F	NR	Fever Myalgia Headache Anosmia Ageusia Cough	Myalgia Numbness Dysesthesias Weakness	+ (Serum and CSF cell-based assay for AQP4 antibodies positive)	ANA 1:320, Meningitis/ Encephalitis Panel negative	14	*Brain:* Anterior fornix and subfornical organ lesions *Spine:* Enhancing LETM	Pleocytosis Elevated Protein Positive IgG index	Positive serological IgM	IVMP PLEX Azathioprine	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 1 (Continued)

Reference/ country	Age/ sex	Comorbidities	Clinical presentation of COVID-19 infection	Clinical presentation of NMO	AQP4 antibody status*	Laboratory investigations	Time interval between COVID-19 and NMO (days)	MRI data	CSF findings	COVID-19 related findings	Treatment of NMO	Outcome
5. Das et al. (17) India	16/F	None	NR	Monocular optic neuritis Back and lower extremity stiffness	+ (Serum AQP4 IgG)	Vitamin B12 normal, thyroid hormone assay normal, serum anti-MOG negative, ANA positive, anti-Ro positive	~120	*Optic Nerves: Optic nerve lesion* *Brain: Fr*ontal subcortical area lesion Spine: LETM from C2 to C7	Normal WBC Normal Protein Elevated IgG index	Positive serological IgG/IgM	IVMP Rituximab	Improvement
6. Ghosh et al. (5) India	20/M	None	Fever Nausea/emesis Cough	Weakness Numbness Urinary retention Constipation Hiccups Nausea Vomiting Myalgias	+ (Transfected HEK293 cell-based assay serum AQP4 IgG positive)	CSF and paired sera: HIV, bacterial and parasitic infections, tuberculosis, autoimmune encephalitis and paraneoplastic encephalitis negative Serum studies: Systemic lupus erythematosus, Sjogren syndrome, Bechet's disease, sarcoidosis, and antiphospholipid antibody syndrome negative, Anti-MOG antibodies	5	*Brain:* Spared *Spine:* Non enhancing LETM from the medulla to T12	WBC 10 cells/uL Protein 80 mg/dL Negative OCB	Positive SARS-CoV-2 nasal PCR	IVMP Rituximab	Improvement
7. Jentzer et al. (18) France	71/F	Hereditary Hemorrhagic Telangiectasia	NR	Paraplegia	+ (Serum anti-AQP4 semi-quantitative cell- based assay)	NR	~90	*Spine:* LETM from C7 to T6	NR	Positive SARS-CoV-2 nasal PCR	NR	NR
8. Khair et al. (6) United States	13/F	Suspected ADEM ADHD	Fatigue Anosmia Ageusia	Diffuse weakness	+ (Serum and CSF)	SS-B IgG antibody positive, Anti-MOG negative, MBP, viral PCR panel and autoimmune encephalopathy panel negative	~60	*Brain:* Numerous non-enhancing lesions in the brain and brainstem *Spine: Numerous* non-enhancing lesions in the cervical and thoracic spinal cord	NR	positive SARS-CoV-2 nasal PCR	IVMP	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 1 (Continued)

Reference/ country	Age/ sex	Comorbidities	Clinical presentation of COVID-19 infection	Clinical presentation of NMO	AQP4 antibody status*	Laboratory investigations	Time interval between COVID-19 and NMO (days)	MRI data	CSF findings	COVID-19 related findings	Treatment of NMO	Outcome
9. Kim et al. (19) Korea	37/F	None	None	Bilateral lower extremity paraparesis Paresthesia Diminished deep tendon reflexes	+ (Serum AQP4 IgG)	Serum studies: C-reactive protein 7.09, Erythrocyte sedimentation rate 74 mm/h, VDRL negative, HIV negative, vitamins B1, B6, B12 normal, methylmalonic acid normal, thyroid-stimulating hormone normal, T3 normal, hemoglobin A1c normal, Jo-1 normal, SS-A/ Ro negative, SS-B/La negative, double-stranded DNA negative, paraneoplastic antibodies negative, anti-ganglioside antibodies negative, immunofixation negative CSF Studies: CMV negative, *Mycobacterium tuberculosis* negative, *Mycoplasma pneumoniae* negative, varicella-zoster virus negative, herpes simplex virus type I and II negative, *Streptococcus pneumoniae* negtaive, *Neisseria meningitidis* negative, Hemophilus influenzae type 1 negative, *Listeria monocytogenes* negative, Group B streptococcus negative, and Cryptococcus negative	3	*Spine:* Enhancing LETM from C1/2 to conus medullaris	WBC 602 cell/uL Proteins 188.4 mg/dL IgG index 0.98 Oligoclonal bands negative Myeline basic protein negative	Positive SARS-CoV-2 nasal PCR	IVMP	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 1 (Continued)

Reference/ country	Age/ sex	Comorbidities	Clinical presentation of COVID-19 infection	Clinical presentation of NMO	AQP4 antibody status*	Laboratory investigations	Time interval between COVID-19 and NMO (days)	MRI data	CSF findings	COVID-19 related findings	Treatment of NMO	Outcome
10. Mirmosayyeb et al. (20) UAE	43/F	None	Fatigue/asthenia Myalgias Anorexia	Urinary retention, Lower extremity numbness Thoracic sensory level Quadriplegia, Bilateral optic neuritis	– (AQP4 Ab test not specified)	NR	NR	*Optic Nerves:* Enhancing bilateral optic nerves *Brain:* Lesions in the thalami, brainstem, periaqueductal grey. Temporal lobe tumefactive lesion *Spine:* Enhancing LETM lesions throughout the cervical and thoracic cord	Mild pleocytosis Highly elevated myelin-basic protein Negative OCB	Positive SARS-CoV-2 PCR IgM/IgG	IVMP PLEX	Improvement
11. Mirmosayyeb et al. (20) United States	NR	NR	NR	Area postrema syndrome	+ (AQP4 Ab test not specified)	NR	NR	*Brain:* Dorsal medullary lesion *Spine:* LETM extending greater than 3 segments	NR	Positive SARS-CoV-2 nasal PCR Positive serological IgG/IgM	NR	NR
12. Mirmosayyeb et al. (20) Egypt	56/F	Surgically resected temporal meningioma	Fatigue Myalgias Anorexia Cough	Bilateral optic neuritis, Disorientation	NR (AQP4 Ab test not specified)	NR	14	*Brain:* Diencephalic, Thalami, Optic Chiasm, Optic Tracts lesions *Spine:* Spared	NR	Positive SARS-CoV-2 nasal PCR CXR: Bilateral Patchy Ground-Glass Opacification	IVMP	Died
13. Rafique et al. (21) Pakistan	7.5/F	None	None	Optic neuritis, Ataxia, Hypotonia, Hyporeflexia	– (Serum AQP4 IgG)	Anti-MOG antibody negative, anti-ganglioside antibody panel negative. CRP elevated, serum ferritin 497 ng/mL, LDH 376 U/L, ESR normal, D- Dimers 0.34 µg/mL	11	*Optic Nerves:* Optic nerve lesion *Brain:* Brain stem, area postrema, periaqueductal lesions *Spine:* Enhancing LETM cervical and thoracic lesions	NR	Positive serological IgG	IVMP IVIG PLEX	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 1 (Continued)

Reference/ country	Age/ sex	Comorbidities	Clinical presentation of COVID-19 infection	Clinical presentation of NMO	AQP4 antibody status*	Laboratory investigations	Time interval between COVID-19 and NMO (days)	MRI data	CSF findings	COVID-19 related findings	Treatment of NMO	Outcome
14. Sardar et al. (22) Qatar	38/F	Diabetes Obesity Obstructive sleep apnea, Migraine Gastritis	Headache Nausea/emesis	Bilateral optic neuritis, Holocephalic headache, Nausea	– (AQP4 Ab test not specified)	NR	14	*Optic nerves:* Bilateral optic nerve lesions *Brain:* Spared *Spine:* Spared	Normal WBC Normal Protein Oligoclonal bands negative	Positive SARS-CoV-2 nasal PCR	IVMP PLEX	Improvement
15. Shaw et al. (23) Australia	70/M	Hypertension Heart disease GERD Former smoker	Dyspnea	Visual blurring, Ptosis, Weakness, Urinary Incontinence, Fasciculation	– (AQP4 Ab test not specified)	C-reactive protein 282 mg/L	9	*Brain:* Spared *Spine:* Enhancing patchy multifocal T5 to T11 lesions	NR	Positive SARS-CoV-2 nasal PCR Positive serological IgG CXR: Bilateral patchy ground-glass opacification	None	Intubated/ died

Ab, antibody; ADEM, acute disseminated encephalomyelitis; ADHD, attention-deficit/hyperactivity disorder; ANA, anti-nuclear antibody; ANCA, anti-neutrophile cytoplasmic antibodies; AQP4, aquaporin-4; CMV, cytomegalovirus; CSF, cerebral spinal fluid; C, cervical; CXR, chest x-ray; EBV, epstein-Bar virus; F, female; HAV, hepatitis A virus; HBV, hepatitis B virus; HIV, human immunodeficiency virus; IgM, immunoglobulin M; IVIG, intravenous immunoglobulins; IVMP, intravenous methylprednisolone; LETM, longitudinally extensive transverse myelitis; M, male; NMO, neuromyelitis optica; PCR, polymerase chain reaction; MRI, magnetic resonance imaging; NR, not reported; PLEX, plasmapheresis; RF, rheumatoid factor; T, thoracic; VDRL, venereal disease research laboratory; WBC, white blood cells.

*This column documents whether the AQP4 antibody testing was completed in the serum, CSF or both and whether it was completed as a cell based assay or an Elisa study. If not specified in this column, the original manuscript did not list this information.

10.3389/fneur.2023.1099758

TABLE 2 Characteristics of cases presenting with *de novo* neuromyelitis optica in relation to COVID-19 vaccination.

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
1. Anamnart et al. (24) Thailand	26/F	None	Sinovac CoronaVac (Inactivated COVID-19 vaccine)	1	10	Leg monopoiesis, Decreased pinprick sensation in the arm, trunk, and leg, Generalized hyperreflexia	*Brain:* Spared *Spine:* Enhancing C4 to C5 lesion	Normal WBC Normal Protein Oligoclonal bands negative	+ (Serum AQP4- IgG by cell-based indirect immunofluorescence assay (CBA-IIF, Euroimmun®), titer 1:320)	NR	IVMP PLEX Rituximab	Improvement
2. Anamnart et al. (24) Thailand	46/F	None	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	1	9	Unilateral lower extremity weakness and hypesthesia, Hyperreflexia	*Brain:* Non-enhancing Medulla and subependymal periventricular area lesions *Spine: Enhancing* C2 to C3 lesion	Normal WBC Normal Protein Oligoclonal bands negative	+ (Serum AQP4- IgG by cell-based indirect immunofluorescence assay (CBA-IIF, Euroimmun®), titer 1:320)	NR	IVMP Azathioprine	Improvement
3. Arora et al. (25) India	50/M	None	NR (Vital vector vaccine)	1	20	Bilateral upper and lower extremity weakness, Urinary retention, Bilateral vision loss	*Brain:* Non-enhancing bilateral dorsolateral thalamic lesions *Spine:* C1, C2, T8 lesions	WBC 32 cells/ uL Protein 55 mg/ dL Oligoclonal bands negative	+ (Serum AQP4-IgG)	ANA negative, C-ANCA negative, P-ANCA negative, VDRL, negative. ACE levels normal. Anti- MOG antibodies negative	IVMP IVIG	Improvement
4. Badrawi et al. (26) United Arab Emirates	34/M	None	Sputnik V COVID-19 (Adenovirus viral vector vaccine)	2	21	Acute confusions, Dizziness, Headache, Imbalance	*Optic nerves:* Optic chiasm lesion *Brain:* Extensive periventricular and/or peri-ependymal lesions including along the third and fourth ventricles and periaqueductal gray mater. Lesions in the thalamus and corpus callosum `*Spine:* Spared	Lymphocystis Elevated protein. Oligoclonal bands negative HSV negative, Syphilis negative, cryptococcal antigen negative, VZV negative	+ (Serum AQP4-IgG Titer 1:40)	COVID-19 negative, adenovirus negative, Herpes Simplex virus (type I & II) negtaive, Epstein Barr virus negative, Cytomegalovirus, and Human Immunodeficiency virus negative	PLEX	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
5. Ballout et al. (27) United States	63/F	Hyperthyroid Hyperlipidemia	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	1	7	Weakness Urinary retention	*Brain: Enhancing* Thalamic lesion *Spine:* Non-enhancing central LETM from T6 to T12	WBC 33 cells/ uL Protein 57 mg/ dL	+ (Serum AQP-4 IgG Utilizing ELISA technique and CSF anti AQP4 Ab CBA with a titer of 1:16)	ANA 1:2560, Anti-DsDNA IU/mL, AE normal, C3 and C4 complement normal, paraneoplastic panel negative, CSF anti-MOG ab negative	IVMP PLEX	Improvement
6. Ballout et al. (27) United States	54/F	Immune thrombocytopenia purpura	Moderna SARS-CoV-2 mRNA-1,273 vaccine	2	3	Ascending numbness	*Brain:* Spared *Spine:* Enhancing central LETM from T2 to T9l	WBC 26 cells/u: Protein 71 mg/ dL MBP 27 Oligoclonal bands negative	+ (Serum AQP-4 IgG Utilizing ELISA technique with titers of 1,417.3 U/mL and CSF anti AQP4 Ab CBA)	ANA 1:320, ESR normal, CRP normal, c-ANCA normal, p-ANCA normal, ACE normal, SSA negative, SSB negative, serum and CSF anti-MOG negative, DsDNA antibodies negative	IVMP	Improvement
7. Caliskan et al. (28) Turkey	43/F	None	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	NR	1	Monocular optic, neuritis Hemiparesthesia, Hemiparesis, Urinary retention, Constipation	*Optic nerve:* Unliteral optic neuritis *Brain:* Enhancing periatrium lesion Non enhancing left crus cerebri *Spine:* Patchy enhancing lesion from C1 to mid-thoracic level	WBC 6 cels/uL Protein 40.1 mg/dL Oligoclonal bands positive	+ (Serum AQP-4 IgG Utilizing CBA with a titer of 1:320)	ANA negative, DsDNA antibody negative, lupus anticoagulant negative, RF negative, anti-cardiolipin antibody, and anti-beta2 glycoprotein levels normal, HIV negative, CMV negative, hepatitis viruses negative, VZV negative, CA 12–5 normal, CA 19–9 normal, CA 15–3, normal, human epididymis protein 4 normal, Anti-MOG ab negative	IVMP PLEX	Complete recovery

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
8. Chen et al. (29) China	Middle aged/F	None	Probable Sinovac CoronaVac or Sinopharm vaccine (Inactivated COVID-19 vaccine)	1	3	Emesis, Dizziness, Unsteady gait	*Brain:* Non enhancing area postrema and bilateral hypothalamus lesions *Spine:* Spared	WBC 31 cell/ uL Normal Protein Oligoclonal bands negative	+ (Serum AQP-4 IgG Utilizing CBA)	Vitamin B1 & B12 levels normal, tumor markers normal, ESR normal, CRP normal, immunoglobulins normal, complements normal, RF negative, antiphospholipid antibodies negative, GFAP IgG negative, Autoimmune encephalitis antibodies negative, paraneoplastic antibodies negative, serum cytokines (IFN-γ, IL-6, IL-4, IL-2, IL-10, IL-21, TNF-α) normal, ANA, positive SSA positive, SSB positive, Ro-52 positive, and p-ANCA positive	IVMP	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
9. Fujikawa et al. (30) United States	46/F	Vitamin B12 deficiency	Moderna SARS-CoV-2 mRNA-1,273 vaccine (mRNA vaccine)	1	2	Shooting back pian, Paresthesia distal to the T10 dermatome, Bilateral upper and lower extremity weakness Urinary retention	*Brain:* Spared *Spine:* Non-enhancing LETM from C6-T2	Normal WBC Normal Protein Oligoclonal bands negative	– (AQP-4 IgG test not specified)	Vitamin B12 level 245 pg./m, CRP normal, TSH normal, hemoglobin A1C normal, aldolase normal, methylmalonic acid normal, antinuclear antibody normal, Jo-1 normal, SS-A/ Ro negative, SS-B/La negative, ribonucleoprotein normal, scleroderma negative, DsDNA negative, anti-ribosomal, chromatin normal, centromere B antibodies negative, C3 & C4 compliments normal	IVMP	Improvement
10. Janarious et al. (31) United States	19/F	None	Moderna SARS-CoV-2 mRNA-1,273 vaccine (mRNA vaccine)	NR	15	Bilateral upper and lower extremity weakness and sensory changes, Urinary incontinence, T4 sensory level	*Brain:* NR *Spine:* LETM from Cervicomedullary junction to the conus medullaris	Pleocytosis Increased IgG synthesis rate	+ (CSF AQP-4 IgG positive, Serum AQP-4 Ab negative)	Serum Anti-MOG Ab negative	IVMP PLEX Rituximab	NR
11. Kim et al. (32) Korea	47/F	None	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	1	22	Intractable hiccups, Gait disturbance, Dysarthria, Dysphagia, Hoarseness	*Brain:* Enhancing medullary lesion, Non-enhancing parietal periventricular lesion *Spine:* Spared	WBC 0 cells/uL Protein 27 mg/ dL Oligoclonal bands negative IgG index 0.44	+ (Serum AQP-4 IgG)	NR	IVMP Azathioprine	Complete recovery

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
12. Kim et al. (32) Korea	57/F	Sjogren's syndrome	Moderna SARS-CoV-2 mRNA-1,273 vaccine (mRNA vaccine)	1	11	Constipation, Bilateral lower extremity paresthesia, T-12 hypoesthesia sensory level, Unilateral diminished position sensation, Bilateral lower extremity diminished vibration sensation, Spasticity	*Brain:* Non-specific white matter changes *Spine:* Enhancing LETM from T5–T9	WBC 0 cells/uL Protein 31 mg/dL Oligoclonal bands negative	+ (Serum AQP-4 IgG)	NR	IVMP Azathioprine	Improvement
13 Kim et al. (19) Korea	37/F	None	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	3	19	Bilateral lower extremity paraparesis, Paresthesia, Diminished deep tendon reflexes	*Brain:* Spared *Spine:* Enhancing intramedullary LETM from C1 to the conus medullaris	WBC 602 cells/uL Proteins 188.4 mg/dL IgG index 0.98 Oligoclonal bands negative	+ (AQP-4 IgG test not specified)	CRP 7.09, ESR 74 mm/h, VDRL negative, HIV negative, vitamins B1, B6, & B12 normal, methylmalonic acid normal, thyroid-stimulating hormone normal, T3 normal, hemoglobin A1c normal, Jo-1 negative, SS-A/Ro negative, SS-B/La negative, DsDNA negative, paraneoplastic antibodies negative, anti-ganglioside antibodies negative	IVMP	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
14. Khayat-Khoei et al. (33) Germany	64/M	Sjogren's disease	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	1	18	Pain, Paresthesia, Unilateral weakness, Urinary retention, Constipation, Balance/gait impairment, Saddle anesthesia	*Brain:* Non-enhancing corpus callosum, frontal white mater, parietal white mater lesions *Spine:* Enhancing central LETM from cervical spine to conus	WBC 1 cells/uL Protein 39 mg/dL Oligoclonal bands negative IgG index 0.68	+ (serum AGP-4 IgG titer > 1:100,000, CSF AQP-4 IgG titer1:128)	Positive SS-A/SS-B antibodies	IVMP PLEX	Improvement
15. Kuntz et al. (34) Canada	80/M	NR	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	2	2	Unilateral weakness, Unilateral numbness, Gait instability Urinary retention	*Brain:* Spared *Spine:* LETM from T3-T4 to T9-T10	WBC 39 cells/uL Protein Normal Oligoclonal bands negative	+ (serum AGP-4 IgG positive)	Anti-MOG Ab positive on initial test and negative on repetition, CRP 10.9, Serological screening for rheumatological and infectious diseases was unremarkable	IVMP PLEX, Mycophenolate mofetil	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
16. Lévi-Strauss et al. (35) France	72/F	None	Moderna SARS-CoV-2 mRNA-1,273 vaccine (mRNA vaccine)	1	7	Paresthesia, Hypoesthesia Weakness of the left arm and leg, Alteration of consciousness, Left sided choreoathetosis	*Brain:* Non-enhancing corpus callosum, area postrema, and periependymal lesions *Spine:* Spared	WBC 500 cells/ uL Protein 117 mg/dL Oligoclonal bands negative	+ (serum AGP-4 IgG positive via CBA)	HIV negative, No no immunodeficiency profile completed, ANA 1:160, anti-SSA/Ro antibody titer > 8 UI/mL, anti-DNA negative, anti-phospholipid antibodies negative, ANCA negative. Anti-MOG negative, anti-thyroid antibodies negative, CSF antiparaneoplastic panel (NMDA, anti-AMPA and anti-VGKC) negative, Serum antiparaneoplastic panel (anti-Yo, -Ri, -GAD, -Hu, -CV and -Tr antibodies) negative	IVMP PLEX Rituximab	Improvement
17. Motahharynia et al. (36) Iran	70/F	None	Sinovac CoronaVac (Inactivated COVID-19 vaccine)	3	7	Unilateral upper and lower extremity hypoesthesia, Quadriplegia	*Brain:* Spared *Spine: Enhancing rim shaped enhancing hemorrhagic LETM from C1 to C7. Lesion from T1 to T3*	WBC normal Protein normal Oligoclonal bands negative	+ (Serum AQP4- IgG via CBA)	NR	IVMP PLEX, Cyclo-phosphamide	Death

(Continued)

10.3389/fneur.2023.1099758

TABLE 2 (Continued)

Reference/ country	Age/ sex	Comorbidities	Name of vaccine (Vaccine type)	Dose #	Time interval between vaccination and NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigation	Treatment of NMO	Outcome
18. Shirah et al. (37) Saudi Arabia	31/F	Systemic Lupus Erythematosus	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	NR	14	Monocular optic neuritis	*Optic nerve:* Enhancing intraocular and intraorbital segments of the left optic nerve *Brain:* Spared *Spine:* Spared	WBC normal Protein normal Oligoclonal bands negative	+ (Serum AQP-4 IgG via immunofluorescence test with a titer of 1:1000)	ANA positive, DsDNA positive (968 IU/mL), ANCA positive, Anti-SSA positive (109 EU/mL) Anti-SSB positive (128 EU/mL), Low C3 (0.72 g/L) & C4 (0.08 g/L) compliments	IVMP PLEX, Rituximab	No recovery
19. Tasci et al. (38) Turkey	32/M	Graves' Disease Gastric neuroendocrine tumor	Sinovac CoronaVac (Inactivated COVID-19 vaccine)	1	14	Unilateral optic neuritis	*Optic nerves:* Right optic Neuritis *Brain;* Spared *Spine:* Spared	NR	+ (Serum AQP-4 IgG)	NR	IVMP Rituximab	Improvement
20. Tisavipat et al. (39) Thailand	50/M	None	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	2	4	Quadriparesis, Painful tonic spasms, Urinary retention	*Brain;* Spared *Spine:* Enhancing LETM from C2 to T1	NR	+ (Serum AQP-4 IgG)	NR	IVMP Rituximab	Improvement
21. Tisavipat et al. (39) Thailand	70/F	None	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	1	10	Lhermitte's sign, Unliteral arm weakness	*Brain;* Spared *Spine:* LETM from C1 to T1	NR	+ (Serum AQP-4 IgG)	NR	IVMP	Improvement

Ab, antibody; ACE, angiotensin converting enzyme; ANA, antinuclear antibodies; ANCA, anti-neutrophil cytoplasm antibodies; AQP4, aquaporin-4; ANCA, antineutrophil cytoplasmic antibodies; CBA, cell based assay; CMV, cytomegalovirus; CRP, C-reactive protein, CSF, cerebral spinal fluid; C, cervical; CXR, chest x-ray; DsDNA, double-stranded deoxyribonucleic acid; ELISA, Enzyme-linked immunosorbent assay; ESR, erythrocyte sedimentation rate; F, female; HSV, herpes Simplex virus; HIV, human immunodeficiency virus; IVIG, intravenous immunoglobulins; IVMP, intravenous methylprednisolone; LETM, longitudinally extensive transverse myelitis; M, male; MOG, myelin oligodendrocyte glycoprotein; MRI, magnetic resonance imaging; NMO, neuromyelitis optica; NR, not reported; PCR, polymerase chain reaction; PLEX, plasmapheresis; RF, rheumatoid factor; SSA, anti-sjogren's syndrome A; SSB, anti-sjogren's syndrome B; T, thoracic; TSH, thyroid-stimulating hormone; VDRL, venereal disease research laboratory; VZV, varicella zoster virus; WBC, white blood cells.

*This column documents whether the AQP4 antibody testing was completed in the serum, CSF or both and whether it was completed as a cell-based assay or an Elisa study. If not specified in this column, the original manuscript did not list this information.

10.3389/fneur.2023.1099758

TABLE 3 Characteristics of cases presenting with central nervous system relapses consistent with neuromyelitis optica in relation to COVID-19 vaccination.

Reference/ country	Age/ sex	Pre-existing history of CNS autoimmune disease	Name of vaccine (vaccine type)	Dose #	Time interval between vaccination & NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigations	Treatment of NMO	Outcome
1. Dinoto et al. (40) Italy	38/F	AQP4 + NMO on rituximab	*Pfizer-BioNTech COVID-19 Vaccine (BNT162b2)* (mRNA vaccine)	2	10	Optic neuritis	NR	NR	+ (AQP4 Ab test not specified)	NR	IVMP	Complete recovery
2. Dinoto et al. (40) Italy	61/F	AQP4 + NMO not on a DMT	*Pfizer-BioNTech* COVID-19 *Vaccine* (*BNT162b2*) (mRNA vaccine)	2	97	Myelitis	NR	NR	+ (AQP4 Ab test not specified)	NR	IVMP	No recovery
3. Fragoso et al. (41) Brazil	62/F	NMOSD DMT status not reported	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	1	7	Monocular vision loss	*Optic* nerve: Enhancing unliteral optic nerve lesion *Brain:* Spared *Spine:* Spared	NR	NR	NR	IVMP PLEX	Improvement

(Continued)

10.3389/fneur.2023.1099758

TABLE 3 (Continued)

Reference/ country	Age/ sex	Pre-existing history of CNS autoimmune disease	Name of vaccine (vaccine type)	Dose #	Time interval between vaccination & NMO (days)	Clinical presentation of NMO	MRI data	CSF findings	AQP4 antibody status*	Laboratory investigations	Treatment of NMO	Outcome
4. Helmchen et al. (42) Germany	40/F	Multiple sclerosis on natalizumab	Oxford–AstraZeneca ChAdOx1 nCoV-19 (Viral vector vaccine)	1	14	Binocular blindness, Lower extremity numbness, T5 sensory level, Back pain, Incontinence, Paraplegia	*Optic nerve:* Enhancing lesion in the chiasm and bilateral optic nerves and tracts *Brain:* Spared *Spine:* LETM from C7 - T1, LETM from T7 - T10, medullary conus lesion	WBC 524 cells/uL Protein 220 mg/dL	-	Anti-MOG negative (confirmed via indirect immuno-fluorescence testing with MOG-transfected HEK-293 cells), GFAP negative, flotillin negative, ANA negative, anti-phospholipids ab negative	IVMP	Improvement
5. Lohmann et al. (43) Germany	68/F	Secondary progressive multiple sclerosis Not on a DMT	*Pfizer-BioNTech COVID-19 Vaccine (BNT162b2)* (mRNA vaccine)	1	23	Sensorimotor paraparesis with a T8 level, Bowel and bladder incontinence	*Brain:* NR *Spine:* Enhancing LETM from C4 to T10	WBC 340 cells/uL Protein 259 mg/dL Oligoclonal bands negative	+ (CSF and serum AQP-4 IgG)		IVMP PLEX Eculizumab	Improvement

Ab, antibody; ANA, anti-nuclear antibody; AQP4, aquaporin-4; CSF, cerebral spinal fluid; C, cervical; CXR, chest x-ray; DMT, disease modifying therapy; F, female; GFAP, glial fibrillary acid protein; IVIG, intravenous immunoglobulins; IVMP, intravenous methylprednisolone; LETM, longitudinally extensive transverse myelitis; M, male; MOG, myelin oligodendrocyte glycoprotein; MRI, magnetic resonance imaging; NMO, neuromyelitis optica; NR, not reported; PCR, PLEX, plasmapheresis; T, thoracic; WBC, white blood cells.

*This column documents whether the AQP4 antibody testing was completed in the serum, CSF or both and whether it was completed as a cell based assay or an Elisa study. If not specified in this column, the original manuscript did not list this information.

10.3389/fneur.2023.1099758

11/20 (55%) had pleocytosis, 9/20 (45%) had elevated CSF protein levels. Only one patient's CSF findings were positive for oligoclonal bands (OCB) out of the 16 cases that explicitly documented OCB status.

In terms of therapy, all but one patient (96%) was initially treated with intravenous methylprednisolone. Subsequently, 12/26 (46%) patients were treated with pulses of plasmapheresis, and 1/26 (4%) patients was treated with IVIG. Maintenance immunotherapy was documented in 12/26 (46%) patients, including rituximab ($n = 6$), azathioprine ($n = 3$), cyclophosphamide ($n = 1$), eculizumab ($n = 1$), and mycophenolate mofetil ($n = 1$). The treatment outcomes were reported for 25/26 patients. Of these patients, 22/25 (88%) experienced complete or partial recovery following treatment, 2/25 (8%) patients did not improve with treatment, and 1/25 (4%) patients died. The cause of death was not discussed in the case series.

Comparison of demographic and clinical characteristics of patients with SARS-CoV-2 post-infection and COVID-19 post-vaccination NMOSD

Table 4 compares the demographic and clinical characteristics of NMOSD following SAR-CoV-2 infection and COVID-19 vaccination. The COVID-19 vaccine exposure group and the SARS-CoV-2 viral infection group had similar sex ratios with a female preponderance, but the vaccine group's age was on average over a decade older than the SARS-CoV-2 infected group. Both groups had a similar percentage of comorbidities, but the COVID-19 vaccine group (31% vs. 13%) was more likely to present with a comorbid autoimmune condition. Both groups had a similar rate of transverse myelitis, but the SARS-CoV-2 infected group were more likely to present with optic neuritis and brain stem involvement. The COVID-19 vaccinated group was also more likely to present with positive AQP4 antibodies than the SARS-CoV-2 infected group (85% vs. 65%). Both groups demonstrated a similar mortality rate.

Discussion

As the COVID-19 pandemic has continued to persist, a mounting number of neurological manifestations and complications related to this disease have been described. Para and post infectious and post vaccination autoimmune CNS demyelination is a rare, but well documented phenomena. A small but accumulating base of literature suggests an association between the SARS-CoV-2 infection, the COVID-19 vaccine, and NMOSD. This systemic review contributes to this growing literature, including 41 worldwide cases of NMOSD temporally associated with the SARS-CoV-2 infection or COVID-19 vaccination. The analysis revealed that the NMOSD cases met standardized criteria, neurological symptoms developed within 2 weeks in most cases, the majority responded to standard immune therapies and overall neurological morbidity was moderate with 7% mortality.

The theory that a viral infection can trigger NMOSD pathogenesis is supported by several case series and case reports demonstrating an association between NMOSD and various viral infections including epstein barr virus, influenza virus, human immunodeficiency viruses (HIV), and varicella zoster virus (9, 44–47). SARS-CoV-2 infection has joined these other viral agents as a potential risk factor for PNS and CNS demyelinating disease (48, 49). In fact, TM, acute necrotizing encephalopathy, acute inflammatory demyelinating polyneuropathy (AIDP), and ADEM events have been associated with SARS-CoV-2 para and post infections, demonstrating that this emergent viral disease is associated with other CNS demyelinating disorders (50–53). Additionally, case reports have demonstrated an association between COVID-19 vaccinations and the onset of ADEM, TM, and MS following the COVID-19 vaccination (33, 54, 55).

The pathological mechanism explaining how the COVID-19 vaccine or the SARS-COV-2 infection induce NMOSD is not fully understood, but it is hypothesized that there is an interplay between viral and vaccine-related features and individual susceptibility factors (56). SARS-CoV-2 is thought to infect its host via the angiotensin-converting enzyme-2 (ACE-2) receptors on the cell surface of type II alveolar epithelial cells in the lung (57, 58). ACE-2 receptors are also expressed on the glial cells and the neurons (59). Therefore, in addition to infecting the respiratory system, SARS-CoV-2 can impact the central and peripheral nervous system. Once the host is exposed to either the COVID-19 vaccine or SARS-CoV-2 infection, NMOSD development may be mediated by either neurotropism or via aberrant immune mediated injury (5). Once SARS-CoV-2 has accessed the nervous system, several proposed pathological mechanisms have been suggested including bystander activation, spreading of the epitope, molecular/immunological mimicry involving cross-reactive autoantibodies targeting SARS-CoV-2 antigens, and amplified blood–brain barrier (BBB) permeability allowing antibody (i.e., AQP-4 peptides) entry into the CNS (5, 10). Evidence indicates that SARS-CoV-2 crosses the blood brain barrier (BBB) along with other cytokines including IL-1β, IL-2, IL-4, IL-6, IL-8, IL-10, TNF-α, and IFN-γ. This impacts macrophages, microglia, and astrocytes, which mediate a cytokine storm leading to the death of neurons and oligodendrocytes. This produces a cytokine storm and a proinflammatory state. Of these cytokines, IL-6 has particular significance as it has been implicated in playing a critical role in regulating the immune response in MS by promoting pathogenic T helper (Th) 17 cells generation (60). Disruption of Th 17 and regulatory T cell responses caused by SARs-CoV-2 exposure can induce inflammation and mitochondrial dysfunction that amplifies the inflammatory process, resulting in immune-metabolic constraints on neural energy metabolism (61). Additional mechanisms include activation of toll-like receptors (TLRs), antibody production against myelin via molecular mimicry, and the affinity for angiotensin-converting enzyme 2 (ACE2) receptors, which can induce myelin destruction (62). Furthermore, neuro-invasion by SAR-CoV-2 or its antigens may cause leakage of CNS antigens such as AQP-4 peptides into the systemic circulation, triggering the bystander immune cascade (5).

Several case reports have indicated that cytotoxic lesion of the corpus callosum (CLOCCs) are also associated with COVID-19 vaccinations (63–65). CLOCCs is caused by an influx of water into

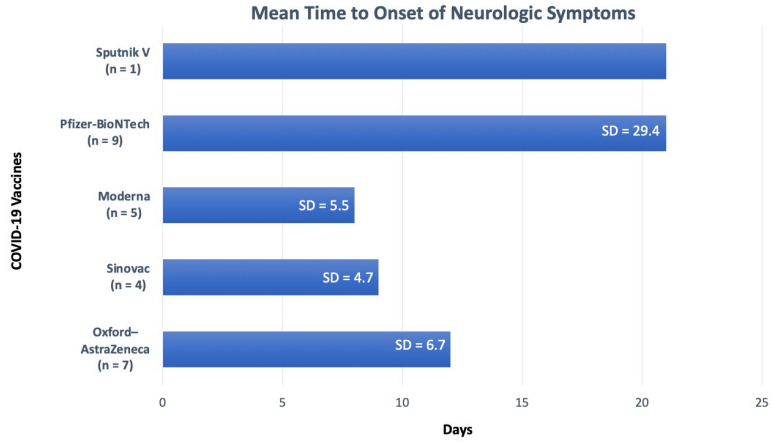


FIGURE 2
Duration from vaccination to symptom onset for each vaccine. SD, Standard deviation.

the cells due to cytokine induced glutamate release from astrocytes (63). The proposed underlying pathogenic mechanism between COVID-19 vaccine triggered NMOSD and CLOCCS is similar. For example, the CSF of CLOCCS patients is notable for elevated IL-6 and IL-10 and these cytokines are also implicated in NMOSD induction. Similarly, Toll-like receptors, which are activated by mRNA vaccines, have been implicated in both processes and both disorders respond to intravenous IV IgG and corticosteroids. Cytokine storm pathology is a central mechanism of both vaccine induced NMOSD and CLOCCs (65).

In terms of SARS-CoV-2 variants in our case series, it is difficult to assess which types were most often implicated. None of the individual case reports discussed which variant was responsible for the reported COVID-19 case associated with NMOSD onset. Except in one case, the original case reports and case series, did not document the date of infection, rendering it difficult to assess which variant was the dominant strain at the time. Furthermore, the publications that reviewed cases of SARS-Cov-2 associated NMOSD were published in 10 different countries across a 3 three-year time span. Using either the date of publication or the date the paper was received to determine the latest possible date that each case of SARS-CoV-2 infection, we found 8 cases reported in 2020, 3 cases in 2021, and 4 case in 2022. Given the diversity of locations and the range of dates of publication and the failure of these publications to document the date of infection, it is not possible to provide reliable data on which variants were represented in this case series. That said, most cases would have contracted the earlier pre-Omicron variants of SARS-CoV-2 (66).

The latency period between vaccine or infection exposure and NMOSD clinical onset ranged from 1 to 120 days but the majority of patients developed neurological symptoms within 1–2 weeks following exposure to the virus or the vaccine. In order for a disorder to be considered vaccine induced, the WHO suggests that there should be a clear temporal relationship between exposure and disease onset. The latency period between the exposure and the adverse event, however, was not defined by the WHO (67). Other studies that attempted to demonstrate a causal link between vaccination and disease onset included various latency time ranges from 8 weeks to 5 months (68, 69). For example, Karussis et al. (68) completed a PubMed search from 1979 to 2013 reviewing 71 documented cases of post-vaccination CNS demyelination secondary to various vaccines including influenza, HPV, and hepatitis A or B vaccines. In their review, symptoms typically manifested within 2 weeks (mean: 14.2 days), however, they also included delayed presentations from 4 weeks to 5 months post-vaccination (68). One study assessing the association between hepatitis B vaccination and the development of MS between 1991 and 1997 utilized an 8-week latency period between vaccination exposure and disease onset (69). Given the rarity of NMOSD, in our study, we included a delayed latency period of up to 120 days to ensure completeness of the data. However, the majority of the cases presented with a latency period of less than 30 days. The mean latency period between SARS-CoV-2 infection and NMOSD development was 34 days [Standard deviation (SD) 39 days]. Of the 11 cases that reported the latency period, only 3 were over 30 days. Of the 21 patients that developed *de novo* NMOSD following the COVID-19 vaccine, all patients had a latency period of less than 30 days (mean: 10 days). Of the patients that developed a relapsing CNS demyelination consistent with NMO following exposure to the COVID-19 vaccine, only one of the 5 cases presented with a latency period of more than 30 days (mean: 30 days). This short-term association, however, should be considered with reservations as there are no controls or quantitative risk outcomes (e.g., odds ratios).

The cases presenting with a long latency distribution, in which NMOSD occurred more than 28 days after the exposure, may represent coincidental NMOSD manifestations. In the long latency cases, the vaccine or infectious exposure and NMOSD disease onset may be causally related rather than causative. These cases of prolonged latency may represent sporadic NMOSD that may have occurred regardless of the exposure, especially as both the SARS-CoV-2 infections and COVID-19 vaccinations were wide spread over a brief interval and a large portion of the population encountered at least one of these exposures. The cases with a short latency distribution of less than 28 days are less likely to be coincidental,

TABLE 4 Comparison of demographic and disease characteristics of patients with SARS-CoV-2 post-infection and COVID-19 post-vaccination NMOSD.

Characteristics	NMOSD following a SARS-CoV-2 infection	*De novo* and relapsing NMOSD following COVID-19 vaccination
Age in years, mean (SD)	37.5 (21)	50 (16)
Sex		
Female (%)	11 (73%)	20 (77%)
Male (%)	3 (20%)	6 (33%)
Not reported	1 (7%)	0 (0%)
Patients with a reported comorbid autoimmune condition (%)	2 (13%)	8 (31%)
Patients with a comorbid condition	6 (40%)	12 (46%)
Days between exposure to SARS-CoV-2 infection vs. COVID-19 vaccination & NMOSD onset (range)	14 (3–120)	10 (1–97)
Neurological manifestations		
Transverse myelitis	10 (67%)	17 (65%)
Short-segment transverse myelitis	2 (13%)	4 (15%)
Longitudinally extensive transverse myelitis	8 (53%)	13 (50%)
Optic neuritis	7 (47%)	5 (19%)
Area postrema syndrome	2 (13%)	3 (12%)
Brainstem involvement	5 (33%)	3 (12%)
AQP-4 antibody status		
Positive (%)	10 (67%)	22 (85%)
Negative (%)	4 (27%)	3 (12%)
Unknown (%)	1 (7%)	1 (4%)
Outcome		
Complete or partial recovery	11 (73%)	22 (85%)
No recovery	0 (0%)	2 (8%)
Death (%)	2 (13%)	1 (4%)
Not reported	2 (13%)	1 (4%)

although causation cannot be proven. Both short and long latency periods were included, however, for completeness as this is a hypothesis generating study. We advise a case-controlled study for a more rigorous investigation.

The current data, spanning from December 2019 to the present provides too brief of an overview to give insight into the long-term risks of para-post infectious and post vaccine associated NMOSD. The data suggests, however, that if SARS-CoV-2 or COVID-19 vaccine exposed patients meet the diagnostic criteria for NMOSD, they should be managed like any other NMOSD patient to optimize the clinical outcome.

Females comprised the majority (76%) of cases in this series. This female preponderance corresponds with data in the literature that indicates a 2-fold higher incidence among females with NMOSD compared with males (70). The female preponderance found in our series may be secondary to a heightened immune response against self and foreign antigens in females compared to males.

With 24% of cases having a prior immune-mediated condition, *de novo* and relapsing NMOSD manifestations may be more prevalent among those with a pre-existing autoimmune disease. The results of this review suggest that in some susceptible individuals, exposure to the SARS-CoV-2 infection or COVID-19 vaccine may introduce a short-term risk of CNS demyelination.

Although this review indicates that there is a plausible association between the COVID-19 vaccination and NMOSD, the number of cases appears to be rare, and vaccination is still strongly encouraged. Currently, epidemiological and clinical data suggests that the benefits of vaccination conferred to both the individual and the public supersedes the possible risk of NMOSD associated with vaccination (34, 71). Furthermore, given the large number of patients that have received the COVID-19 vaccination, only a few reports have documented NMOSD manifestations following the vaccine, indicating that this is an uncommon occurrence.

This is a comprehensive systemic review of NMOSD cases associated with SARS-CoV-2 infections and the COVID-19 vaccine, including 34 published reports and 41 individual cases. The majority of cases reported in the existing literature were presented as case reports, and the few case series available were often more broadly focused on a variety of CNS demyelinating disorders rather than exclusively discussing NMOSD.

Given the established temporal relationship between SARS-CoV-2 infections and COVID-19 vaccination and the onset of NMOSD, our systemic review adds the current literature that underscores a potential link between viral infections and vaccinations and the development of *de novo* and relapsing NMOSD. This review suggests a probable association between post-infectious or vaccine triggered autoimmune mediated CNS demyelinating astrocytopathy. Our findings also suggest that vaccine and viral triggered CNS autoimmune demyelination may be more common among individuals with a pre-existing autoimmune disorder or a family history of autoimmune disease. However, the heterogeneity of the clinical data prevents a metanalysis from being performed. Although a causative relationship cannot be established on a temporal association alone, raising awareness of this potential correlation may influence the diagnosis and management of future patients presenting with demyelinating sequalae in the setting of infectious or vaccine mediated triggers. The lack of a control group prevented our ability to generate standard risk outcomes and future studies involving a control group are merited. This paper provides evidence for hypothesis generation that can be further tested with a case-control study allowing for a more detailed characterization of demographic, clinical characteristic, and genetic data to prove causality.

Strengths of this review include the comprehensive search of the literature, the detailed adjudication of cases and the comparison of COVID-19 vaccine and SARS-CoV-2 infection. Limitations included the small number of cases with retrospective observations. Several

cases included incomplete workups and there was heterogeneity of clinical data available, impairing the ability to complete a meta-analysis.

Conclusion

This systematic review comprehensively demonstrates a temporal association between *de novo* and relapsing forms of NMOSD and SARS-CoV-2 infections and COVID-19 vaccinations. Association, however, does not away imply causation. We would also emphasize that the protective benefits that the COVID-19 vaccine conveys to both the individual and society as a whole far outweigh any hypothetical risk that would be implied from this review. Our report suggests, a link between the COVID-19 virus or vaccine exposure and the pathological cascade that may induce clinical NMOSD symptoms. Furthermore, given the brief duration of the study, the potential long-term effects of exposure are unknown. This systematic review does suggest that NMO manifestations following a COVID-19 viral or vaccine exposure may be more common than currently recognized, particularly among high-risk demographic groups. This association requires further study using quantitative epidemiological assessments in representative populations to better quantify the risk of developing clinical symptoms of NMOSD.

Author contributions

MW and TH contributed to conceptualization, study design, literature search, obtaining data, data management and analyses, data verification, drafting the manuscript and figures, manuscript revisions, statistical analysis, administrative oversight, study supervision, and validation. All authors contributed to the article and approved the submitted version.

Acknowledgments

The authors are grateful the University of Maryland Health Sciences and Human Services Library staff for their assistance on our review and development of the Covidence database.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2023.1099758/full#supplementary-material

References

1. Ismail II, Salama S. Association of CNS demyelination and COVID-19 infection: an updated systematic review. *J Neurol.* (2021) 1–36.

2. World Health Organization. (2022). *WHO COVID-19 dashboard*. Available at: https://covid19.who.int (Accessed November 2, 2022).

3. Desforges M, Le Coupanec A, Dubeau P, Bourgouin A, Lajoie L, Dubé M, et al. Human coronaviruses and other respiratory viruses: underestimated opportunistic pathogens of the central nervous system? *Viruses.* (2019) 12:14. doi: 10.3390/v12010014

4. Kim JE, Heo JH, Kim HO, Song SH, Park SS, Park TH, et al. Neurological complications during treatment of Middle East respiratory syndrome. *J Clin Neurol (Seoul, Korea).* (2017) 13:227–33. doi: 10.3988/jcn.2017.13.3.227

5. Ghosh R, De K, Roy D, Mandal A, Biswas S, Biswas S, et al. A case of area postrema variant of neuromyelitis optica spectrum disorder following SARS-CoV-2 infection. *J Neuroimmunol.* (2021) 350:577439. doi: 10.1016/j.jneuroim.2020.577439

6. Khair AM, Nikam R, Husain S, Ortiz M, Kaur G. Para and post-COVID-19 CNS acute demyelinating disorders in children: A case series on expanding the Spectrum of clinical and radiological characteristics. *Cureus.* (2022) 14:e23405. doi: 10.7759/cureus.23405

7. Corrêa DG, de Souza Lima FC, da Cruz Bezerra D, Coutinho AC, Hygino da Cruz LC. COVID-19 associated with encephalomyeloradiculitis and positive anti-aquaporin-4 antibodies: cause or coincidence? *Mult Scler J.* (2021) 27:973–6. doi: 10.1177/1352458520949988

8. Yavari F, Raji S, Moradi F, Saeidi M. Demyelinating changes alike to multiple sclerosis: a case report of rare manifestations of COVID-19. *Case Rep Neurol Med.* (2020) 2020:1–4. doi: 10.1155/2020/6682251

9. Lana-Peixoto MA, Talim N. Neuromyelitis optica spectrum disorder and anti-MOG syndromes. *Biomedicine.* (2019) 7:42. doi: 10.3390/biomedicines7020042

10. Koga M, Takahashi T, Kawai M, Fujihara K, Kanda T. A serological analysis of viral and bacterial infections associated with neuromyelitis optica. *J Neurol Sci.* (2011) 300:19–22. doi: 10.1016/j.jns.2010.10.013

11. Zhong X, Zhou Y, Lu T, Wang Z, Fang L, Peng L, et al. Infections in neuromyelitis optica spectrum disorder. *J Clin Neurosci.* (2018) 47:14–9. doi: 10.1016/j.jocn.2017.10.005

12. Langer-Gould A, Qian L, Tartof SY, Brara SM, Jacobsen SJ, Beaber BE, et al. Vaccines and the risk of multiple sclerosis and other central nervous system demyelinating diseases. *JAMA Neurol.* (2014) 71:1506–13. doi: 10.1001/jamaneurol.2014.2633

13. Wingerchuk DM, Banwell B, Bennett JL, Cabre P, Carroll W, Chitnis T, et al. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. *Neurology.* (2015) 85:177–89. doi: 10.1212/WNL.0000000000001729

14. Aubart M, Roux CJ, Durrleman C, Gins C, Hully M, Kossorotoff M, et al. Neuroinflammatory disease following severe acute respiratory syndrome coronavirus 2 infection in children. *J Pediatr.* (2022) 247:22–28.e2. doi: 10.1016/j.jpeds.2022.05.018

15. Barone S, Rapisarda L, Manzo L, Mechelli A, Pascarella A, Bruno P, et al. A case of neuromyelitis optica spectrum disorder (NMOSD) and acute myositis following SARS-CoV-2 infection. *J Neurol Sci.* (2021) 429:119862. doi: 10.1016/j.jns.2021.119862

16. Batum M, Kisabay Ak A, Mavioğlu H. Covid-19 infection-induced neuromyelitis optica: a case report. *Int J Neurosci.* (2022) 132:999–1004. doi: 10.1080/00207454.2020.1860036

17. Das D, Bhattacharjee H, Rehman O, Deori N, Magdalene D, Bharali G, et al. Neuromyelitis optica spectrum disorder post-COVID-19 infection: A rare case report from Northeast India. *Indian J Ophthalmol.* (2022) 70:1833–6. doi: 10.4103/ijo.IJO_61_22

18. Jentzer A, Carra-Dallière C, Lozano C, Riviere S, Darmon O, Ayrignac X, et al. Neuromyelitis optica spectrum disorder following COVID-19 infection with increase in pre-existing anti-aquaporin-4 antibodies. *J Neurol.* (2022) 269:2850–3. doi: 10.1007/s00415-022-10972-9

19. Kim Y, Heo D, Choi M, Lee JM. A case presenting with Neuromyelitis Optica Spectrum disorder and infectious Polyradiculitis following BNT162b2 vaccination and COVID-19. *Vaccine*. (2022) 10:1028. doi: 10.3390/vaccines10071028

20. Mirmosayyeb O, Ghaffary EM, Bagherieh S, Barzegar M, Dehghan MS, Shaygannejad V. Post COVID-19 infection neuromyelitis optica spectrum disorder (NMOSD): A case report-based systematic review. *Mult Scler Relat Disord*. (2022) 60:103697. doi: 10.1016/j.msard.2022.103697

21. Rafique S, Wasim A, Sultan T, Ahmad A. Post-COVID neuromyelitis optica spectrum disorder. *J Coll Physicians Surg Pak*. (2021) 31:138–40. doi: 10.29271/jcpsp.2021.Supp2.S138

22. Sardar S, Safan A, Okar L, Sadik N, Adeli G. The diagnostic dilemma of bilateral optic neuritis and idiopathic intracranial hypertension coexistence in a patient with recent COVID-19 infection. *Clin Case Rep*. (2021) 9:e04347. doi: 10.1002/ccr3.4347

23. Shaw VC, Chander G, Puttanna A. Neuromyelitis optica spectrum disorder secondary to COVID-19. *Br J Hosp Med*. (2020) 81:1–3. doi: 10.12968/hmed.2020.0401

24. Anamnart C, Tisavipat N, Owattanapanich W, Apiwattanakul M, Savangned P, Prayoonwiwat N, et al. Newly diagnosed neuromyelitis optica spectrum disorders following vaccination: case report and systematic review. *Mult Scler Relat Disord*. (2022) 58:103414. doi: 10.1016/j.msard.2021.103414

25. Arora S, Sehgal V, Bansal P. Neuromyelitis Optica like presentation following COVID vaccination. *Eur J Mol Clin Med*. (2021) 8:909–16.

26. Badrawi N, Kumar N, Albastaki U. Post COVID-19 vaccination neuromyelitis optica spectrum disorder: case report & MRI findings. *Radiol Case Rep*. (2021) 16:3864–7. doi: 10.1016/j.radcr.2021.09.033

27. Ballout AA, Babaie A, Kolesnik M, Li JY, Hameed N, Waldman G, et al. A single-health system case series of new-onset CNS inflammatory disorders temporally associated with mRNA-based SARS-CoV-2 vaccines. *Front Neurol*. (2022) 13:264. doi: 10.3389/fneur.2022.796882

28. Caliskan I, Bulus E, Afsar N, Altintas A. A case with new-onset neuromyelitis optica spectrum disorder following COVID-19 mRNA BNT162b2 vaccination. *Neurologist*. (2022) 27:147–50. doi: 10.1097/NRL.0000000000000420

29. Chen S, Fan XR, He S, Zhang JW, Li SJ. Watch out for neuromyelitis optica spectrum disorder after inactivated virus vaccination for COVID-19. *Neurol Sci*. (2021) 42:3537–9. doi: 10.1007/s10072-021-05427-4

30. Fujikawa P, Shah FA, Braford M, Patel K, Madey J. Neuromyelitis optica in a healthy female after severe acute respiratory syndrome coronavirus 2 mRNA-1273 vaccine. *Cureus*. (2021) 13:e17961. doi: 10.7759/cureus.17961

31. Janarious A., Ullah S., (2022). Severe presentation of Neuromyelitis Optica with positive antibody following COVID-19 vaccination (P 7–1.006). *Neurology*, [online] 98 (18 Supplement). Available at: https://n.neurology.org/content/98/18_Supplement/1666

32. Kim KH, Kim SH, Park NY, Hyun JW, Kim HJ. Onset of various CNS inflammatory demyelination diseases following COVID-19 vaccinations. *Mult Scler Relat Disord*. (2022) 68:104141. doi: 10.1016/j.msard.2022.104141

33. Khayat-Khoei M, Bhattacharyya S, Katz J, Harrison D, Tauhid S, Bruso P, et al. COVID-19 mRNA vaccination leading to CNS inflammation: a case series. *J Neurol*. (2022) 269:1093–106. doi: 10.1007/s00415-021-10780-7

34. Kuntz S., Saab G., Schneider R., (2022). Antibody-positive Neuromyelitis Optica Spectrum disorder after second COVID-19 vaccination: a case report. SN Comprehensive Clinical Medicine, 4:130.

35. Lévi-Strauss J, Provost C, Wane N, Jacquemont T, Mélé N. NMOSD typical brain lesions after COVID-19 mRNA vaccination. *J Neurol*. (2022) 269:5213–5. doi: 10.1007/s00415-022-11229-1

36. Motahharynia A, Naghavi S, Shaygannejad V, Adibi I. Fulminant neuromyelitis optica spectrum disorder (NMOSD) following COVID-19 vaccination: A need for reconsideration? *Mult Scler Relat Disord*. (2022) 66:104035. doi: 10.1016/j.msard.2022.104035

37. Shirah B, Mulla I, Aladdin Y. Optic neuritis following the BNT162b2 mRNA COVID-19 vaccine in a patient with systemic lupus erythematosus uncovering the diagnosis of Neuromyelitis Optica Spectrum disorders. *Ocul Immunol Inflamm*. (2022) 18:1–3. doi: 10.1080/09273948.2022.2089901

38. Tascı YY, Nalcacoglu P, Gumusyayla S, Vural G, Toklu Y, Yesılırmak N. Aquaporin-4 protein antibody-associated optic neuritis related to neuroendocrine tumor after receiving an inactive COVID-19 vaccine. *Indian J Ophthalmol*. (2022) 70:1828–31. doi: 10.4103/ijo.IJO_2494_21

39. Tisavipat N, Anamnart C, Owattanapanich W, Apiwattanakul M, Savangned P, Prayoonwiwat N, et al. Author's response to the commentary: Neuromyelitis optica complicating COVID vaccinations and additional case reports. *Mult Scler Relat Disord*. (2022) 66:104055. doi: 10.1016/j.msard.2022.104055

40. Dinoto A, Sechi E, Ferrari S, Gajofatto A, Orlandi R, Solla P, et al. Risk of disease relapse following COVID-19 vaccination in patients with AQP4-IgG-positive NMOSD and MOGAD. *Mult Scler Relat Disord*. (2022) 58:103424. doi: 10.1016/j.msard.2021.103424

41. Fragoso YD, Gomes S, Gonçalves MVM, Junior EM, de Oliveira BES, Rocha CF, et al. New relapse of multiple sclerosis and neuromyelitis optica as a potential adverse event of Astra Zeneca AZD1222 vaccination for COVID-19. *Mult Scler Relat Disord*. (2022) 57:103321. doi: 10.1016/j.msard.2021.103321

42. Helmchen C, Buttler GM, Markewitz R, Hummel K, Wiendl H, Boppel T. Acute bilateral optic/chiasm neuritis with longitudinal extensive transverse myelitis in longstanding stable multiple sclerosis following vector-based vaccination against the SARS-CoV-2. *J Neurol*. (2022) 269:49–54. doi: 10.1007/s00415-021-10647-x

43. Lohmann L, Glaser F, Möddel G, Lünemann JD, Wiendl H, Klotz L. Severe disease exacerbation after mRNA COVID-19 vaccination unmasks suspected multiple sclerosis as neuromyelitis optica spectrum disorder: a case report. *BMC Neurol*. (2022) 22:1–4. doi: 10.1186/s12883-022-02698-y

44. Machado C, Amorim J, Rocha J, Pereira JM, Lourenço E, Pinho J. Neuromyelitis optica spectrum disorder and varicella-zoster infection. *J Neurol Sci*. (2015) 358:520–1. doi: 10.1016/j.jns.2015.09.374

45. Mathew T, Avati A, D'Souza D, Therambil M, Baptist AA, Shaji A, et al. HIV infection associated neuromyelitis optica spectrum disorder: clinical features, imaging findings, management and outcomes. *Mult Scler Relat Disord*. (2019) 27:289–93. doi: 10.1016/j.msard.2018.11.014

46. Sellner J, Hemmer B, Mühlau M. The clinical spectrum and immunobiology of parainfectious neuromyelitis optica (Devic) syndromes. *J Autoimmun*. (2010) 34:371–9. doi: 10.1016/j.jaut.2009.09.013

47. Ghosh R., De K., Roy D., Mandal A., Biswas S., Biswas S., et al., (2007). A case of area postrema variant of neuromyelitis optica spectrum disorder following SARS-CoV-2 infection. *J. Neuroimmunol*. (2021) 350:577439. doi: 10.1016/j.jneuroim.2020.577439

48. Schirinzi T, Landi D, Liguori C. COVID-19: dealing with a potential risk factor for chronic neurological disorders. *J Neurol*. (2021) 268:1171–8. doi: 10.1007/s00415-020-10131-y

49. Shabani Z. Demyelination as a result of an immune response in patients with COVID-19. *Acta Neurol Belg*. (2021) 121:859–66. doi: 10.1007/s13760-021-01691-5

50. Nolen LT, Mukerji SS, Mejia NI. Post-acute neurological consequences of COVID-19: an unequal burden. *Nat Med*. (2022) 28:20–3. doi: 10.1038/s41591-021-01647-5

51. Al-Ramadan A, Rabab'h O, Shah J, Gharaibeh A. Acute and post-acute neurological complications of COVID-19. *Neurol Int*. (2021) 13:102–19. doi: 10.3390/neurolint13010010

52. Moreno-Escobar MC, Kataria S, Khan E, Subedi R, Tandon M, Peshwe K, et al. Acute transverse myelitis with Dysautonomia following SARS-CoV-2 infection: A case report and review of literature. *J Neuroimmunol*. (2021) 353:577523. doi: 10.1016/j.jneuroim.2021.577523

53. Lahiri D, Ardila A. COVID-19 pandemic: a neurological perspective. *Cureus*. (2020) 12:e7889. doi: 10.7759/cureus.7889

54. Román GC, Gracia F, Torres A, Palacios A, Gracia K, Harris D. Acute transverse myelitis (ATM): clinical review of 43 patients with COVID-19-associated ATM and 3 post-vaccination ATM serious adverse events with the ChAdOx1 nCoV-19 vaccine (AZD1222). *Front Immunol*. (2021) 12:879. doi: 10.3389/fimmu.2021.653786

55. Havla J, Schultz Y, Zimmermann H, Hohlfeld R, Danek A, Kümpfel T. First manifestation of multiple sclerosis after immunization with the Pfizer-BioNTech COVID-19 vaccine. *J Neurol*. (2022) 269:55–8. doi: 10.1007/s00415-021-10648-w

56. Ismail II, Salama S. A systematic review of cases of CNS demyelination following COVID-19 vaccination. *J Neuroimmunol*. (2022) 362:577765. doi: 10.1016/j.jneuroim.2021.577765

57. Wan Y, Shang J, Graham R, Baric RS, Li F. Receptor recognition by the novel coronavirus from Wuhan: an analysis based on decade-long structural studies of SARS coronavirus. *J Virol*. (2020) 94:e00127–0. doi: 10.1128/JVI.00127-20

58. Hirano T, Murakami M. COVID-19: a new virus, but a familiar receptor and cytokine release syndrome. *Immunity*. (2020) 52:731–3. doi: 10.1016/j.immuni.2020.04.003

59. Reyfman PA, Walter JM, Joshi N, Anekalla KR, McQuattie-Pimentel AC, Chiu S, et al. Single-cell transcriptomic analysis of human lung provides insights into the pathobiology of pulmonary fibrosis. *Am J Respir Crit Care Med*. (2019) 199:1517–36. doi: 10.1164/rccm.201712-2410OC

60. Ismail II, Salama S. Association of CNS demyelination and COVID-19 infection: an updated systematic review. *J Neurol*. (2022) 269:541–76. doi: 10.1007/s00415-021-10752-x

61. Kappelmann N, Dantzer R, Khandaker GM. Interleukin-6 as potential mediator of long-term neuropsychiatric symptoms of COVID-19. *Psychoneuroendocrinology*. (2021) 131:105295. doi: 10.1016/j.psyneuen.2021.105295

62. Feizi P, Sharma K, Pasham SR, Nirwan L, Joseph J, Jaiswal S, et al. Central nervous system (CNS) inflammatory demyelinating diseases (IDDs) associated with COVID-19: A case series and review. *J Neuroimmunol*. (2022) 371:577939. doi: 10.1016/j.jneuroim.2022.577939

63. Youn T, Yang H. Cytotoxic lesion of the corpus callosum (CLOCCs) after SARS-CoV-2 mRNA vaccination. *J Korean Med Sci*. (2021) 36:e228. doi: 10.3346/jkms.2021.36.e228

64. Poussaint TY, LaRovere KL, Newburger JW, Chou J, Nigrovic LE, Novak T, et al. Multisystem inflammatory-like syndrome in a child following COVID-19 mRNA vaccination. *Vaccine*. (2021) 10:43. doi: 10.3390/vaccines10010043

65. Ohara H, Shimizu H, Kasamatsu T, Kajita A, Uno K, Lai KW, et al. Cytotoxic lesions of the corpus callosum after COVID-19 vaccination. *Neuroradiology*. (2022) 64:2085–9. doi: 10.1007/s00234-022-03010-y

66. Young M, Crook H, Scott J, Edison P. Covid-19: virology, variants, and vaccines. *BMJ Med*. (2022) 1:e000040. doi: 10.1136/bmjmed-2021-000040

67. World Health Organization (2018). Causality assessment of an adverse event following immunization (AEFI): user manual for the revised WHO classification. (2nd ed.). Geneva: World Health Organization; Licence: CC BY-NC-SA 3.0 IGO. Available at: https://apps.who.int/iris/bitstream/handle/10665/259959/9789241513654-eng.pdf?sequence=1&isAllowed=y

68. Karussis D, Petrou P. The spectrum of post-vaccination inflammatory CNS demyelinating syndromes. *Autoimmun Rev*. (2014) 13:215–24. doi: 10.1016/j.autrev.2013.10.003

69. Wraith DC, Goldman M, Lambert PH. Vaccination and autoimmune disease: what is the evidence? *Lancet*. (2003) 362:1659–66. doi: 10.1016/S0140-6736(03)14802-7

70. Papp V, Magyari M, Aktas O, Berger T, Broadley SA, Cabre P, et al. Worldwide incidence and prevalence of neuromyelitis optica: a systematic review. *Neurology*. (2021) 96:59–77. doi: 10.1212/WNL.0000000000011153

71. Malhotra HS, Gupta P, Prabhu V, Kumar Garg R, Dandu H, Agarwal V. COVID-19 vaccination-associated myelitis. *QJM: Int J Med*. (2021) 114:591–3. doi: 10.1093/qjmed/hcab069

frontiers | Frontiers in Neurology

TYPE Original Research
PUBLISHED 26 October 2023
DOI 10.3389/fneur.2023.1266067

OPEN ACCESS

EDITED BY
Ilka Kleffner,
University Hospital Bochum GmbH, Germany

REVIEWED BY
Elena Grebenciucova,
Northwestern University, United States
Takao Kiriyama,
Nara Medical University, Japan

*CORRESPONDENCE
Limei Wang
✉ lylimeiwang@126.com

RECEIVED 24 July 2023
ACCEPTED 09 October 2023
PUBLISHED 26 October 2023

CITATION
Sun M, Liu H, Zhu B, Liu Y, Li A and Wang L (2023) Comparison of glial fibrillary acidic protein-immunoglobulin G-associated myelitis with myelin oligodendrocyte glycoprotein-immunoglobulin G-associated myelitis.
Front. Neurol. 14:1266067.
doi: 10.3389/fneur.2023.1266067

Comparison of glial fibrillary acidic protein-immunoglobulin G-associated myelitis with myelin oligodendrocyte glycoprotein-immunoglobulin G-associated myelitis

Mengyang Sun[1], Hao Liu[1], Bingqing Zhu[1], Yang Liu[1], Aijia Li[2] and Limei Wang[1]*

[1]Department of Neurology, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, China, [2]Zhengzhou University Medical College, Zhengzhou, China

Objective: Glial fibrillary acidic protein-immunoglobulin G (GFAP-IgG)-associated myelitis and myelin oligodendrocyte glycoprotein-IgG (MOG-IgG)-associated myelitis have rarely been compared. Therefore, this study aimed to explore the clinical, laboratory, and imaging features of them to identify the differences.

Methods: Overall, 14 and 24 patients with GFAP-IgG-and MOG-IgG-associated myelitis, respectively, were retrospectively screened and included in the study.

Results: Among the 14 patients with GFAP-IgG-associated myelitis, the condition was more common in males (71.4%), with a median age of onset of 36.5 years, and more common in adults than in children (35.7%). In contrast, among the 24 patients with MOG-IgG-associated myelitis, the condition was equally divided between males and females, with a median age of onset of 9.5 years and more in children (66.7%) than in adults. The median age of onset of GFAP-IgG-associated myelitis was later than that of the MOG-IgG group. Isolated myelitis was rare in both groups. Elevated cerebrospinal fluid (CSF) protein levels were more prevalent in patients with GFAP-IgG-associated myelitis (64.3%) than in those with MOG-IgG-associated myelitis (16.7%) ($p < 0.05$), whereas patchy gadolinium enhancement of the cerebral lesion site was less common in patients with GFAP-IgG-associated myelitis than in those with MOG-IgG associated myelitis ($p < 0.05$). Six patients had a combination of other neurological autoantibodies, the specific mechanism of the overlapping antibodies remains unclear.

Conclusion: Cerebrospinal fluid analysis and gadolinium enhanced MRI examination may help to distinguish the two kinds of myelitis.

KEYWORDS

glial fibrillary acidic protein (GFAP), myelin oligodendrocyte glycoprotein (MOG), myelitis, cerebrospinal fluid, overlapping antibodies

Introduction

Autoimmune glial fibrillary acidic protein astrocytopathy (GFAP-A) is an autoimmune encephalomyelitis mainly affecting the central nervous system (CNS) caused by a new autoantibody that can detect antibodies against GFAP-immunoglobulin G (GFAP-IgG) (1). The major manifestations of this disease include inflammation of the meninges, brain, spinal cord, and optic nerves (ON). Additionally, it frequently presents with a subacute onset of memory loss, confusion, one or more meningeal symptoms, and myelitis manifestations (2). Cranial imaging reveals periventricular radially oriented perivascular enhancement, and myelitis commonly manifests as longitudinally extensive transverse myelitis (LETM) (2–4). Anti-myelin oligodendrocyte glycoprotein-IgG (MOG-IgG)-associated disorders (MOGAD) are immune-mediated inflammatory demyelinating disorders of the CNS that manifest as ON, transverse myelitis, or acute disseminated encephalomyelitis (ADEM); they are usually recurrent and can lead to functional impairment due to recurrence (5, 6). MOG-IgG-associated myelitis usually presents as LETM (7). Although the clinical manifestations of myelitis usually include a combination of motor weakness, sensory symptoms, and bowel and bladder dysfunctions and are somewhat disabling, timely identifying the etiology is essential to reducing the harmful effects of inflammation (8). Furthermore, both diseases are associated with spinal cord lesions and have rarely been compared, and exploring their clinical, laboratory, and imaging features can help us understand them. Therefore, this study retrospectively analyzed and compared the clinical features of GFAP-IgG-and MOG-IgG-associated myelitis to better understand the clinical diagnosis and management processes.

Materials and methods

Patients

Overall, 35 and 91 patients with positive serum/cerebrospinal fluid (CSF) GFAP-IgG and MOG-IgG, respectively, who visited the First Affiliated Hospital of Zhengzhou University from October 2019 to October 2022 were retrospectively recruited, and the inclusion criteria for patients with autoimmune GFAP-A were as follows: (1) positive serum or CSF GFAP-IgG level; (2) presence of spinal cord lesions; (3) negative serum or CSF MOG-IgG and aquaporin-4-IgG (AQP4-IgG) levels; and (4) complete clinical data. In contrast, the inclusion criteria for patients with MOGAD were as follows: (1) positive serum MOG-IgG level, (2) presence of spinal cord lesions, (3) negative serum or CSF GFAP-IgG and AQP4-IgG levels, and (4) complete clinical data. Furthermore, the following were the exclusion criteria: other diseases such as traumatic brain injury, brain tumors, lead exposure, and multiple sclerosis. Based on the inclusion and exclusion criteria, 14 and 24 patients (MOG-IgG titers are presented in the Supplementary Material) with GFAP-IgG-and MOG-IgG-associated myelitis, respectively, were reported, describing their age, sex, clinical characteristics, CSF findings, brain and spinal cord magnetic resonance imaging (MRI) features, and treatment.

Laboratory and imaging examinations

Cell-based assays were used to detect GFAP-IgG, MOG-IgG, and AQP4-IgG levels in the serum or CSF of the patients. All patients underwent spinal cord and brain imaging using 3.0 T MRI with spinal cord lesions of ≥3 and <3 segments as long- and short-segment lesions, respectively. Additionally, some patients received intravenous gadolinium injections to assess potential contrast enhancement. MRIs of the brain and spinal cord were performed by a neurologist and a neuroradiologist, respectively. Furthermore, the normal reference ranges for each laboratory index were as follows: CSF pressure: 80–180 mmH_2O, CSF leukocytes: $(0–5)\times10^6$/L, lymphocyte ratio: 60–70%, CSF protein: 150–450 mg/L, and CSF adenosine deaminase (ADA): 0–10 ng/mL.

Ethics statement

This study followed the ethical guidelines and received ethical approval from the Ethics Committee of the First Affiliated Hospital of Zhengzhou University (2022-KY-1205-002).

Statistical analysis

Statistical analyses and data visualization were performed using SPSS version 26.0. Normally and non-normally distributed continuous variables were expressed as mean ± standard deviation ($x \pm s$) and median, respectively. Frequencies (percentages) were used for categorical variables. Furthermore, continuous and categorical variables were compared using the Wilcoxon rank-sum test and the chi-square or Fisher's exact probability test, respectively. Statistical significance was considered at $p < 0.05$.

Result

General information

Among the 35 patients with positive CSF or serum GFAP-IgG levels, 20 and 15 were males and females, respectively. In total, 18 (51.4%) patients had spinal cord lesions, and four had overlapping antibodies. Among the 14 patients with GFAP-IgG-associated myelitis who were included, nine, two, and three were positive for GFAP-IgG in CSF, CSF and serum, and serum, respectively. Additionally, among the 91 patients with positive serum MOG-IgG levels, 47 and 44 were males and females, respectively. Thirty (33.0%) patients had spinal cord lesions, five had overlapping antibodies, and one did not undergo a CSF examination. Finally, 24 patients with MOG-IgG-associated myelitis were included, of whom 11 and 13 were positive for MOG-IgG in the serum and in both CSF and serum, respectively.

Demographic and clinical characteristics

Among the 14 patients with GFAP-IgG-associated myelitis, 2 (14.3%) had isolated myelitis in adults. Six (42.9%) patients had a

presumed or confirmed infection with prodromal symptoms; one had varicella-zoster virus (VZV) infection 2 months before the onset of neurological symptoms; two had Epstein–Barr virus (EBV) infection; one had human herpes virus type 7 infection; and two had influenza-like symptoms. Additionally, one case of combined tuberculosis and another patient with a previous diagnosis of acute Guillain-Barre syndrome 6 months earlier were identified. One patient had a history of rheumatoid arthritis and tested positive for anticyclic citrullinated peptide and rheumatoid factor. Four patients exhibited elevated levels of thyroid-associated antibodies. Furthermore, 3 (12.5%) of 24 patients with MOG-IgG-associated myelitis had isolated myelitis, all of whom were adults. In total, 10 (41.7%) patients had prodromal symptoms before the disease onset, and nine had upper respiratory tract infection symptoms, including two mycoplasma infections and one respiratory syncytial virus infection. Two cases were associated with hepatitis B virus and *Helicobacter pylori* infections. Moreover, five patients had elevated thyroid-associated antibodies; one had previous erythema multiforme and positive antinuclear antibodies; one was diagnosed with Hashimoto's thyroiditis; and two had a history of allergic purpura. However, no tumors were found in any of the 38 patients.

Common clinical symptoms in patients with GFAP-IgG-and MOG-IgG-associated myelitis are presented in Figure 1—limb weakness is the most common in both types of myelitis. Table 1 lists the clinical characteristics of 38 patients with myelitis. Fourteen patients with GFAP-IgG-associated myelitis were more likely to be males (71.4%), with a median age of onset of 36.5 years, and more were adults than children (35.7%). In contrast, 24 patients with

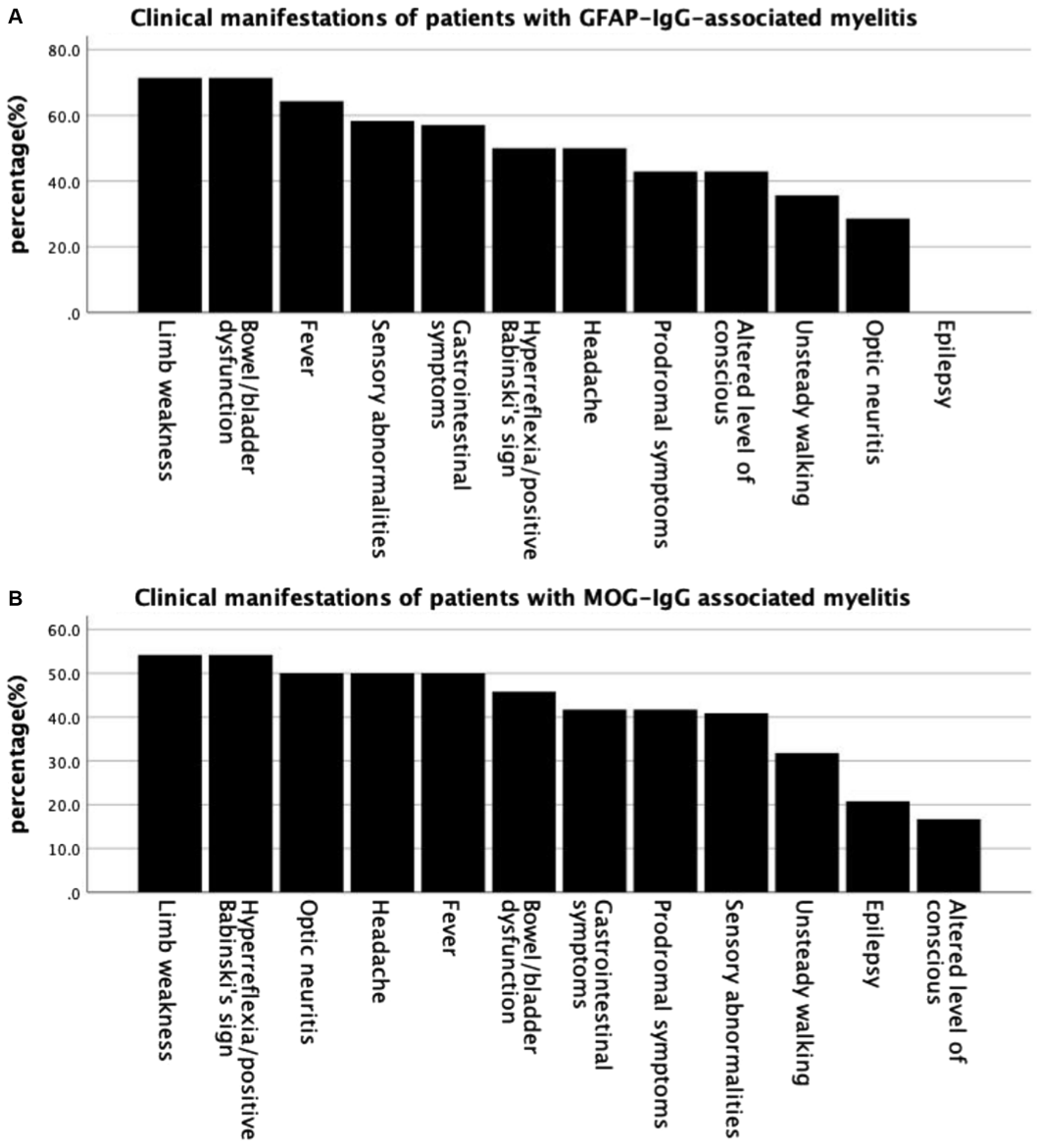


FIGURE 1
Common clinical symptoms in patients with GFAP-IgG-and MOG-IgG-associated myelitis. **(A)** Clinical manifestations of patients with GFAP-IgG associated myelitis. **(B)** Clinical manifestations of patients with MOG-IgG associated myelitis. GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.

TABLE 1 Demographic and clinical characteristics of patients with GFAP-IgG and MOG-IgG-associated myelitis [*n* (%)].

Characteristics	GFAP-IgG, *n* = 14	MOG-IgG, *n* = 24	*p*-values
Female	4 (28.6%)	12 (50.0%)	0.309
Children	5 (35.7%)	16 (66.7%)	0.094
Age (range), years	36.5 (9–71)	9.5 (1–79)	0.060
Prodromal symptoms	6 (42.9%)	10 (41.7%)	1.000
Extraspinal symptoms			
Headache	7 (50.0%)	11/22 (50.0%)	1.000
Fever	9 (64.3%)	12 (50.0%)	0.506
Gastrointestinal symptoms	8 (57.1%)	10 (41.7%)	0.503
Optic neuritis[a]	4 (28.6%)	12 (50.0%)	0.309
Epilepsy	0 (0.0%)	5 (20.8%)	0.137
Altered level of conscious	6 (42.9%)	4 (16.7%)	0.127
Symptoms/signs of myelopathy			
Sensory abnormalities[b]	7/12 (58.3%)	9/22 (40.9%)	0.475
Bowel/bladder dysfunction	10 (71.4%)	11 (45.8%)	0.181
Hyperreflexia/positive Babinski's sign	7 (50.0%)	13 (54.2%)	1.000
Limb weakness	10 (71.4%)	13 (54.2%)	0.329
Unsteady walking	5 (35.7%)	7/22 (31.8%)	1.000
Treatment			
Intravenous methylprednisolone	12 (85.7%)	23 (95.8%)	0.542
Intravenous immunoglobulin	5 (35.7%)	8 (33.3%)	1.000
Plasma exchange	2 (14.3%)	0 (0.0%)	0.129
Immunosuppressants	1 (7.1%)	2 (8.3%)	1.000

GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.
[a]Optic neuritis manifestations include vision loss, blurred vision, double vision, oculomotor pain or retrobulbar pain, optic papillary edema, and abnormal visual evoked potentials (VEP).
[b]Sensory abnormalities such as pain, painful tonic spasm, pruritus, numbness, and hypesthesia.

MOG-IgG-associated myelitis had an equal number of males and females, with a median age of onset of 9.5 years, and more were children (<18 years) (66.7%) than adults. Intravenous methylprednisolone (IVMP) was the most commonly used treatment for both myelitis types. No significant differences were found between the 14 and 24 cases of GFAP-IgG-and MOG-IgG-positive groups based on age of onset, sex ratio, proportion of children, extraspinal symptoms, symptoms/signs of myelopathy, and treatment ($p > 0.05$) (Table 1).

CSF analysis

A CSF examination was performed in 38 patients during the acute phase, among whom elevated CSF protein levels were more prevalent in the GFAP-IgG group than in the MOG-IgG group, with significant differences ($p < 0.05$). Additionally, three patients in the GFAP-IgG group had CSF protein >1 g/L rather than in the MOG-IgG group. In both groups, patients had elevated CSF leukocyte count and lymphocyte ratio, types of oligoclonal bands (OCB), and increased CSF pressure (adults) without a significant difference ($p > 0.05$). Furthermore, elevated CSF ADA levels were rare in both groups (Table 2).

TABLE 2 CSF findings of GFAP-IgG and MOG-IgG-associated myelitis [*n* (%)].

Findings	GFAP-IgG, *n* = 14	MOG-IgG, *n* = 24	*p*-values
Elevated leukocyte	12 (85.7%)	20 (83.3%)	1.000
Significantly elevated leukocyte[a]	8 (57.1%)	9 (37.5%)	0.318
Elevated protein	9 (64.3%)	4 (16.7%)	0.005
Elevated lymphocyte ratio	12 (85.7%)	14 (58.3%)	0.147
Elevated pressure[b]	3/9 (33.3%)	2/8 (25.0%)	1.000
OCB in CSF (type 2)	2 (14.3%)	7 (29.2%)	0.438
OCB in CSF and serum (type 3)	2 (14.3%)	1 (4.2%)	0.542
Elevated ADA	1/13 (7.7%)	0/15 (0.0%)	0.464

OCB, oligoclonal band; CSF, cerebrospinal fluid; ADA, adenosine deaminase; GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.
[a]Significantly elevated leukocyte count >50 × 10*6 /L.
[b]Elevated pressure: >180 mm H_2O. (The pressure values in this table are for adult patients only).

Imaging characteristics

Cranial and spinal MRIs were performed in 38 patients. We depicted the two patient groups' imaging performance to visualize their differences (Figure 2). Among the 14 patients with GFAP-IgG-associated myelitis, 11 (78.6%) had intracranial lesions, which mainly manifested as high signals on T2 weighted image/fluid-attenuated inversion recovery (T2WI/FLAIR) sequences (Figures 3A1,A2). Nine patients underwent gadolinium enhancement scans, of whom three and two had meningeal and cerebellar enhancements, respectively (Figures 3A3). LETM was present in 10 cases (76.9%) (Figures 3C1,C2), and 13 cases (92.9%) had the involvement of central gray matter. Additionally, 21 of 24 patients (87.5%) with MOG-IgG-associated myelitis had intracranial lesions, which were characterized by abnormal signals on T2WI/FLAIR (Figures 3B1,B2). Eleven cranial gadolinium-enhanced scans were performed, 6 (54.5%) showed patchy enhancement, 12 (57.1%) with LETM (Figures 3D1,D3), and 18 (75.0%) with involvement of the central gray matter (1 case was confined to the gray matter of the spinal cord, with an axial appearance of the "H sign"; Figures 3D2). In cranial MRI, patchy gadolinium enhancement was less common in patients with GFAP-IgG-associated myelitis than in those with MOG-IgG-associated myelitis, with a significant difference ($p < 0.05$). No significant differences were found between the two groups regarding cranial MRI lesion site, spinal cord

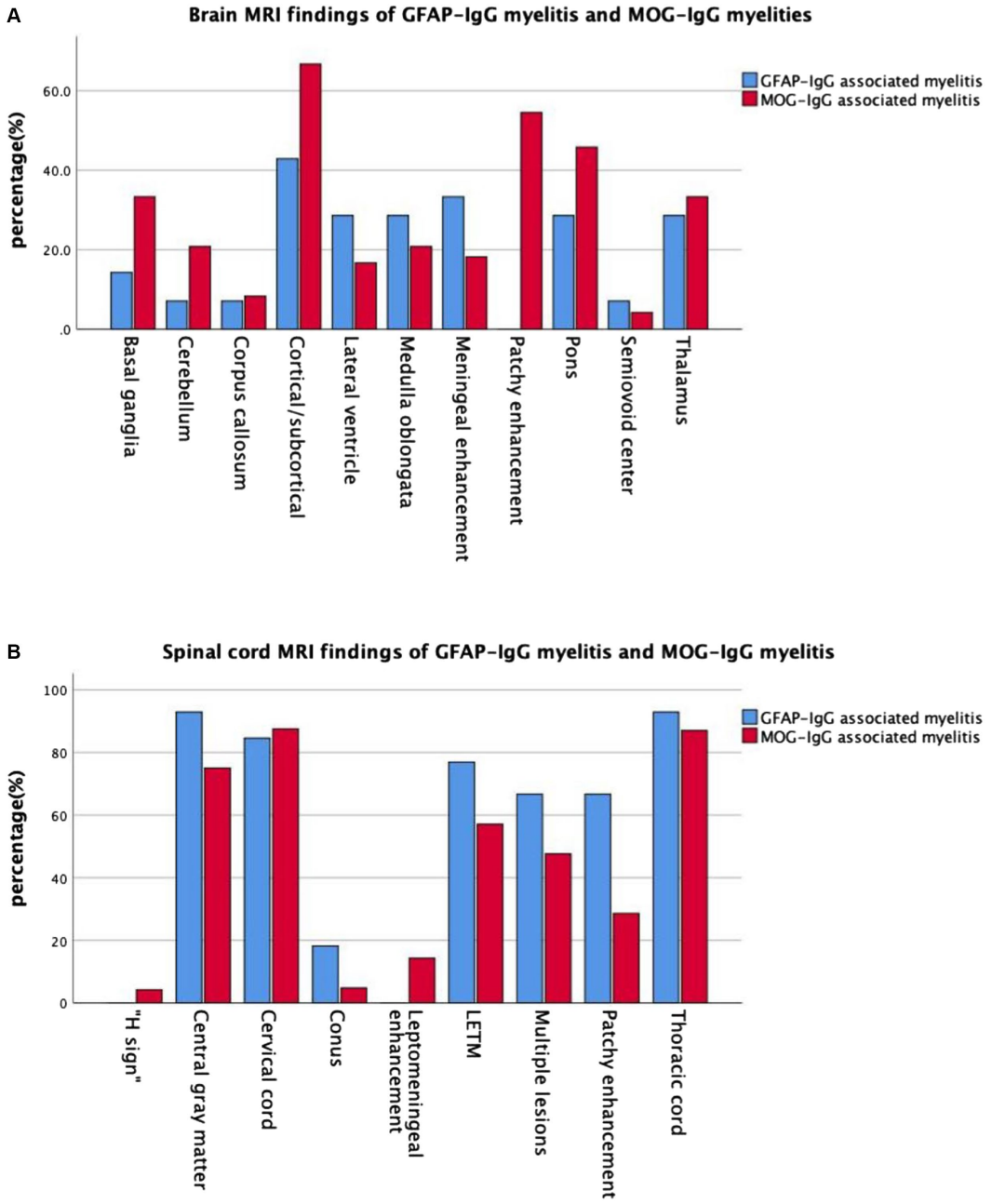


FIGURE 2
Imaging manifestations in the two groups of patients with myelitis. **(A)** Brain MRI and **(B)** spinal MRI manifestations in the two groups of patients with myelitis. MRI, magnetic resonance imaging; GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.

long-segment lesions, lesion distribution, or site of involvement ($p > 0.05$) (Table 3).

Overlapping antibodies

In addition to the 38 patients included, six with myelitis had other neurological autoantibodies in combination (Table 4). Patient 1 was admitted to the hospital with weakness in the limbs and unsteady walking and was administered IVMP, intravenous immunoglobulin (IVIG), and plasma exchange (PE). She was discharged from the hospital with improved symptoms compared to the previous visit. Two months later, she was revisited, and the relevant antibodies turned negative. Moreover, no obvious discomfort was noted. Patient 2 was admitted to the hospital because of vomiting and unsteady walking, and the test results suggested an EBV infection. Additionally, a vision examination suggested decreased vision in both eyes and abnormal visual evoked potential (VEP) in the right eye. The patient was administered IVMP, IVIG, and immunosuppressive treatments and was reexamined with a negative GFAP-IgG level. Her symptoms improved, and she was discharged from the hospital with mycophenolate mofetil (MMF) and a small dose of steroid. Subsequently, the patient experienced recurrence

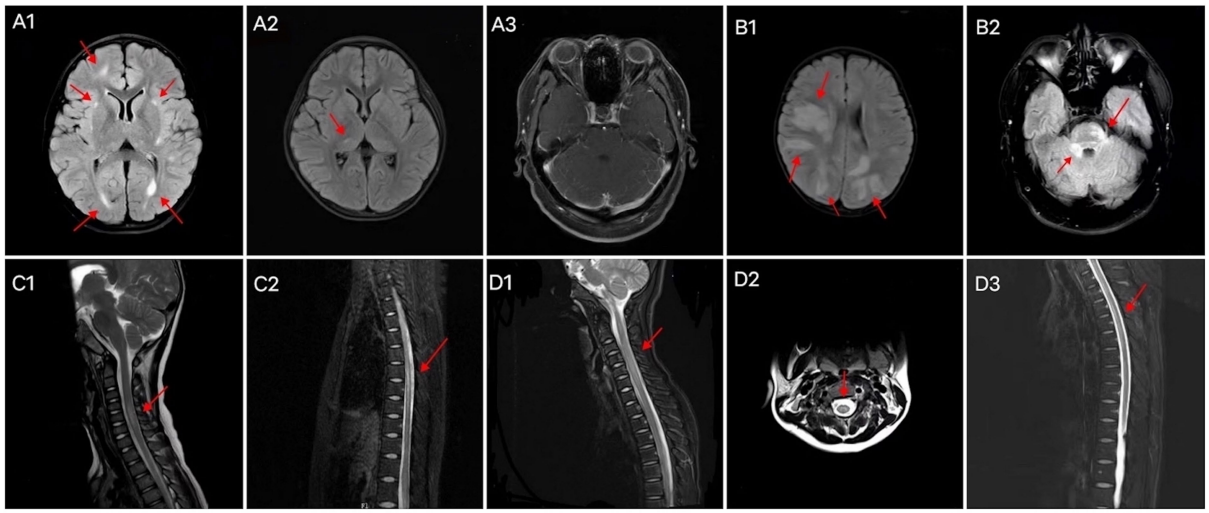


FIGURE 3
Brain MRI characteristics of patients with **(A)** GFAP-IgG-and **(B)** MOG-IgG-associated myelitis. Abnormal signals in the right frontal lobe, bilateral lateral posterior horn of the ventricle, and bilateral basal ganglia areas **(A1)**; abnormal signals in the right thalamus **(A2)**; bilateral cerebellar multiple strip enhancement **(A3)**; multiple abnormal signals in the cortex/subcortex **(B1)**; abnormal signal in the right pontocerebellar commissure and pons **(B2)**. Spinal cord MRI characteristics of patients with GFAP-IgG-associated myelitis (**C1, C2**) and MOG-IgG-associated myelitis (**D1–D3**). LETM of the cervical cord **(C1)**, LETM of the thoracic cord **(C2)**, LETM of the cervical cord **(D1)**, "H sign" **(D2)**, and LETM of the thoracic cord **(D3)**. LETM, longitudinally extensive transverse myelitis; MRI, magnetic resonance imaging; GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.

TABLE 3 MRI characteristics of GFAP-IgG and MOG-IgG-associated myelitis [*n* (%)].

Characteristics	GFAP-IgG, *n* = 14	MOG-IgG, *n* = 24	*p*-values
Intracranial lesion	11 (78.6%)	21 (87.5%)	0.650
Meningeal enhancement	3/9 (33.3%)	2/11 (18.2%)	0.617
Patchy enhancement	0/9 (0.0%)	6/11 (54.5%)	0.014
Cortical/subcortical	6 (42.9%)	16 (66.7%)	0.187
Medulla oblongata	4 (28.6%)	5 (20.8%)	0.699
Lateral ventricle	4 (28.6%)	4 (16.7%)	0.433
Thalamus	4 (28.6%)	8 (33.3%)	1.000
Pons	4 (28.6%)	11 (45.8%)	0.329
Basal ganglia	2 (14.3%)	8 (33.3%)	0.268
Corpus callosum	1 (7.1%)	2 (8.3%)	1.000
Semiovoid center	1 (7.1%)	1 (4.2%)	1.000
Cerebellum	1 (7.1%)	5 (20.8%)	0.383
Spinal cord MRI			
LETM	10/13 (76.9%)	12/21 (57.1%)	0.292
Cervical cord	11/13 (84.6%)	21 (87.5%)	1.000
Thoracic cord	13 (92.9%)	20/23 (87.0%)	1.000
Conus	2/11 (18.2%)	1/21 (4.8%)	0.266
Central gray matter	13 (92.9%)	18 (75.0%)	0.227
"H sign"*	0 (0.0%)	1 (4.2%)	1.000
Multiple lesions	8/12 (66.7%)	10/21 (47.6%)	0.469
Leptomeningeal enhancement	0/6 (0.0%)	1/7 (14.3%)	1.000
Patchy enhancement	4/6 (66.7%)	2/7 (28.6%)	0.286

*Spinal cord MRI T2-hyperintensity is confined to gray matter on axial sequences, forming the "H sign." LETM, longitudinally extensive transverse myelitis; MRI, magnetic resonance imaging; GFAP, glial fibrillary acidic protein; IgG, immunoglobulin G; MOG, myelin oligodendrocyte glycoprotein.

TABLE 4 Clinical, CSF, and imaging characteristics of patients with overlapping antibodies.

Case number / sex/age, y	Clinical symptoms	Antibody (CSF)	Antibody (serum)	CSF leukocytes	CSF protein	MRI	Treatment
NO.1 Female 11	Weakness of both lower limbs, unstable walking, drowsiness, loss of appetite, vomiting, urinary and bowel disorders, seizure, hallucinations, and change in temperament	GFAP MOG NMDAR	GFAP MOG NMDAR	18×10^6/L	262.0 mg/L	Frontal lobe, C3-5, T10	IVMP, IVIG, and PE
NO.2 Female 12	Vomiting, unsteady walking, blurred vision, fever, itchy skin, crooked mouth, and incomplete eyelid closure	GFAP AQP4	AQP4	11×10^6/L	443.0 mg/L	Pontine brain medulla oblongata C1–5, T1, T7, T10–11	IVMP, IVIG, and immunosuppressants
NO.3 Male 36	Blurred vision in the right eye, numbness and weakness in both lower limbs, episodic limb spasms and convulsions, fever, unsteady walking, and bowel/bladder dysfunction	GFAP MOG	MOG	38×10^6/L	342.0 mg/L	Frontal lobe, thalamus, cerebral peduncle, periventricular area, cerebellar, C1–T3	IVMP, IVIG, and immunosuppressants
NO.4 Female 4	Blurred vision, fever, and itchy skin	MOG	GFAP MOG	22×10^6/L	407.0 mg/L	C4-T8	IVMP and immunosuppressants
NO.5 Male 26	Visual confusion, fever, headache, seizure, dizziness, abnormal sensation in both lower limbs, and intermittent speech confusion	NMDAR	MOG NMDAR	40×10^6/L	342.0 mg/L	Cortical/subcortical, pons, pontine arms, cerebral peduncles, and thalamus C5-T10	IVMP and IVIG
NO.6 Female 6	Seizures, fever, tremors, headache, loss of vision, drowsiness, and urinary disturbances	MOG	MOG IgLON5	71×10^6/L	340.0 mg/L	Optic nerve C3-T1	IVMP

C, cervical spinal cord; T, thoracic spinal cord; IVMP, intravenous methylprednisolone; IVIG, intravenous immunoglobulin; PE, plasma exchange; MRI, magnetic resonance imaging; NMDAR, N-methyl-d-aspartate receptor; GFAP, glial fibrillary acidic protein; MOG, myelin oligodendrocyte glycoprotein; AQP4, aquaporin-4; CSF, cerebrospinal fluid.

and left-side blindness in the right eye. Patient 3 was admitted to the hospital with blurred vision in the right eye and numbness and weakness in both lower limbs. The patient had previous symptoms of an antecedent infection and was treated with IVMP, IVIG, and MMF. He had three recurrences, and the antibody was negative on follow-up. The patient's muscle strength was better than before, although he walked unsteadily. Patient 4 was admitted to the hospital with blurred vision in the right eye. She had a fever, dizziness, and drowsiness 1 month prior. The patient was

treated with IVMP and was discharged from the hospital after her vision improved. Subsequently, she was treated with low-dose steroid and reviewed regularly. The patient had blurred vision in the left eye and was discharged from the hospital after being treated with MMF. She relapsed several times and used other immunosuppressive agents; however, vision loss persisted in both eyes. Patient 5 was admitted to the hospital with blurred vision in the right eye and experienced headaches, fever, and seizures 20 days before the disease's onset. A physical examination revealed sensory abnormalities in both lower limbs. After admission, the patient had intermittent speech disorganization and was treated with IVMP and IVIG. He was discharged from the hospital after his psychiatric symptoms had improved and was readmitted with diplopia 3 months later. Finally, he continued receiving IVMP and was administered small doses of steroid and MMF outside the hospital. Patient 6 was admitted to the hospital with intermittent seizures and fever, and laboratory tests suggested EBV infection, blurred vision, IVMP treatment, and improved vision. Finally, she was discharged for convulsive episodes with lethargy and dysuria.

Discussion

This study retrospectively compared the clinical features, CSF and imaging features, and overlapping antibodies in 14 and 24 patients with GFAP-IgG-and MOG-IgG-associated myelitis, respectively.

In our study, GFAP-IgG-associated myelitis was prevalent in adults, consistent with the results of previous studies (4, 9, 10). Two patients (14.3%) had isolated myelitis, and all children had a combined presentation of encephalitis. Four patients (28.6%) were females, while 4 (31%) were females in a previous study (9). However, other studies reported a female majority (4, 10). There are few studies on myelitis; the number of cases is small, and there is some variation in the findings. Therefore, a large-scale study is required to further confirm whether there are sex differences. Patients with prodromal symptoms in GFAP-IgG-associated myelitis are relatively common (42.9% in our study), viral infections are common, and some studies have shown that autoimmune GFAP-A may be associated with herpes simplex virus infection (3, 11, 12). One patient in our study had a VZV infection 2 months before the disease onset; Dubey et al. (13) reported one patient who developed autoimmune GFAP-A a few weeks after VZV encephalitis. Therefore, further studies are needed to determine whether this disease is associated with viral infections. In contrast, compared to previous studies [16/54 (30%)] (14), MOG-IgG-associated myelitis was more prevalent in children (66.7%). In a study of 54 patients with MOG-IgG-associated myelitis, 24 (44%) were females (14), while 15 (39.5%) were females in another study (15). In our study, 12 (50%) patients were female. No significant sex differences were observed between groups. Although 41.8% of patients with MOG-IgG-associated myelitis have a prodromal event, such as infection or vaccination, before disease onset (15), our study revealed such cases in 41.7% of patients; however, no vaccination was found. Four patients had other autoimmune diseases, suggesting that the appearance of MOGAD may be related to an immune disorder. A Chinese study showed that 29.2% (38/130) of patients with MOGAD had myelitis (15). Myelitis is the second most common manifestation of MOGAD in adults, accounting for 18–52% of cases (16). In our study, 30 patients (33.0%) had spinal cord lesions on MRI with a younger age of onset. Limb weakness, dysuria, and sensory abnormalities were common in the GFAP-IgG group, which could also have manifested as unsteady walking (35.7%). Xu et al. (4) showed that 1 of 19 patients exhibited unsteady walking and dysuria. Furthermore, in our study, ON symptoms were relatively uncommon in the GFAP-IgG group than MOG-IgG group, and no epilepsy was found, nor were any of the patients in the study by Fang et al. (1) presented with epilepsy. Nine (69%) patients in the study by Sechi et al. (9) presented with tremors, which were not observed in our patients. Moreover, the 38 patients included showed that most of them with myelitis in both groups also had extraspinal symptoms, and nonspecific symptoms, such as headache and fever, were common in both groups. In both groups, the treatment was generally IVMP. Therefore, compared to MOG-IgG-associated myelitis, GFAP-IgG-associated myelitis appears to have a later age of onset, is relatively rare in patients with optic neuritis, and is relatively common in those with impaired consciousness.

CSF findings in both groups showed inflammatory changes and a predominance of elevated lymphocytes, and OCB was observed in both the serum and CSF. In our study, 12 (85.7%) and 9 (64.3%) patients with GFAP-IgG-associated myelitis had elevated CSF leukocytes and CSF proteins, respectively, and all 13 patients in a previous study had increased CSF leukocytes (9). In another retrospective study of 16 patients with GFAP-IgG-associated myelitis, the median CSF protein concentration was 729 mg/L and approximately 2,344 mg/L (10). Our CSF protein was approximately 1837 mg/L, including three cases >1 g/L; therefore, we hypothesize that protein elevation is more pronounced in the CSF of patients with GFAP-IgG-associated myelitis. However, future sample sizes should be expanded to provide more evidence. Most of the patients (83.3%) in the MOG-IgG group had elevated CSF leukocytes, with nine (37.5%) having $>50 \times 10^6$/L; other studies have shown that 45–55% of patients can have significantly elevated CSF leukocytes (14, 17). A study reported elevated CSF protein levels in 77% of the 35 patients with MOG-IgG-associated myelitis (18). Contrary to our study (16.7%), maybe the elevation of CSF proteins was not significant in pediatric patients with MOGAD compared to adults. No significant elevation in CSF ADA levels was observed in either group. Previous studies have suggested that most patients with autoimmune GFAP-A show a transient elevation in CSF ADA levels during the first month of onset (19). Therefore, we hypothesized that GFAP-IgG-associated myelitis is more likely to be associated with elevated CSF protein levels and is more significantly increased than MOG-IgG-associated myelitis, which may indicate a severe inflammatory response in the CNS.

Autoimmune GFAP-A has been shown to exhibit typical periventricular radially oriented perivascular enhancement in approximately 40–50% of cases (1–3, 20). While intracranial lesions in patients with GFAP-IgG-associated myelitis in our study were found in the cortical/subcortical, medulla oblongata, lateral ventricles, thalamus, pons, and basal ganglia, among others, nine of them had cranial MRI enhancement scans, although this typical enhancement was not observed. However, three cases (33.3%) were abnormal and showed meningeal enhancement, and cerebellar meningeal enhancement was observed in two cases. In a Chinese cohort, cranial MRI of this disease was extremely rare in patients with periventricular radially oriented perivascular

enhancement, whereas cerebellar meningeal enhancement was noteworthy (21). In another study, no cases of this typical enhancement were observed (22). Intracranial lesions in MOGAD tend to present as periventricular lesions extending from nearby cortical lesions with large lesion areas. Nodular enhancement has been observed on enhancement scans (23, 24). Eleven of our patients with MOG-IgG-associated myelitis underwent cranial enhancement scans, and six (54.5%) showed patchy enhancement. In our study, LETM was common in patients with GFAP-IgG-associated myelitis. Additionally, the cervical and thoracic spinal cords were susceptible; 2/11 (18.2%) involved the conus, >90% involved the central gray matter, and multiple lesions were common, consistent with a report of 19 patients by a Chinese author (4). One study reported that the spinal cord central canal, punctate, or leptomeningeal enhancement is reportedly typical of GFAP-IgG-associated myelitis (9). Among the six patients with gadolinium enhancement in this study, four (66.7%) showed patchy enhancement. None of our 14 patients with GFAP-IgG myelitis had spinal cord lesions confined to gray matter on axial MRI sequences (expressed as the "H sign"). LETM and short-segment lesions were found in 12 (57.1%) and 9 (42.9%) patients with MOG-IgG-associated myelitis, similar to previous reports, which showed that LETM is the predominant pattern in MOG-IgG-associated myelitis; however, short-segment lesions are also common (16, 17). MRI lesions of the spinal cord in patients with MOG-IgG-associated myelitis, predominantly involving the central gray matter, were limited to the axial sequence of only one patient (4.2%) exhibiting a more pronounced "H sign," which differs from the findings of previous studies (14). A Chinese cohort study showed that only 3 out of 29 (10.3%) patients with MOGAD had spinal cord enhancement that was limited to gray matter (24). Our study showed that patients with MOG-IgG-associated myelitis frequently had involvement of the cervical and thoracic spinal cords, while conus involvement was rare. Another study showed that myelitis was common in the thoracic and lumbar spinal cord (25), and conus involvement may have been relatively rare in the Chinese cohort. Cerebellar meningeal enhancement appeared to occur more frequently in patients with GFAP-IgG-associated myelitis. Based on a previous study (14), the "H sign" of spinal MRI, which is confined to the gray matter, may also be an imaging feature that distinguishes the two types of myelitis.

Overlapping antibodies were observed for both autoimmune GFAP-A and MOGAD (26). Six patients in our study with a combination of other neurological autoantibodies had a younger age of onset (maximum age, 35 years). Three (50%) patients had prodromal symptoms or co-infection with a viral infection; however, no tumors were detected. Patients with autoimmune encephalitis (AE) usually present with rapidly progressive cognitive dysfunction, refractory epilepsy, and psychiatric abnormalities (27). In our study, all three patients with positive autoimmune antibodies showed related symptoms (patients 1, 5, and 6). Therefore, patients with overlapping antibodies can have both disease manifestations, such as symptoms of AE and demyelinating disease, which may complicate the diagnosis. Whether patients with overlapping antibodies are more likely to have tumors remains unclear; however, no tumors were found in the six patients in this study, allowing for long-term follow-up. Moreover, the specific mechanism of the overlap syndrome has not been fully elucidated; therefore, more studies are needed to further elucidate the underlying mechanism.

MOG is a glycoprotein that is specifically expressed in CNS oligodendrocytes, and anti-MOG-IgG is a pathogenic antibody against MOGAD (28). Although GFAP is an intracellular protein, antibodies against intracellular antigens are unlikely to reach their targets *in vivo* and are not usually considered pathogenic, and a T-cell-mediated inflammatory response is considered the primary mechanism of neuronal destruction (29). Some pathological biopsies support a CD8+ T-cell-mediated autoimmune response (30). However, the pathogenesis of these diseases remains unclear, and more research is needed to elucidate them and better understand and treat them.

Limitations

First, there may be bias due to the retrospective design of the study, as well as a case selection bias because of its single-center nature. Therefore, future prospective cohort studies with larger sample sizes should further clarify the differences between the two groups. Second, the specificity was low when only serum GFAP-IgG was positive, and 3 of 14 GFAP-IgG myelitis cases were only serum positive; however, all cases had characteristic clinical syndromes, and other diseases were reasonably excluded. Lastly, this study focused on comparing clinical features between the two groups of patients rather than on clinical outcomes or long-term follow-up.

Conclusion

This study provides clinical, CSF, and MRI evidence for recognizing and differentiating GFAP-IgG-and MOG-IgG-associated myelitis. Therefore, these findings will help clinicians better recognize these diseases.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving humans were approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University (2022-KY-1205-002). The studies were conducted in accordance with the local legislation and institutional requirements. Written informed consent for participation in this study was provided by the participants' legal guardians/next of kin.

Author contributions

MS: Writing – original draft, Data curation, Formal analysis. HL: Data curation, Formal analysis, Writing – review & editing. BZ: Data curation, Formal analysis, Writing – review & editing. YL: Data curation, Formal analysis, Writing – review & editing. AL: Data curation, Formal analysis, Writing – review & editing. LW: Writing – review & editing.

Funding

The author(s) declare that no financial support was received for the research, authorship, and/or publication of this article.

Acknowledgments

The authors are very grateful to the participants involved in this study.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2023.1266067/full#supplementary-material

References

1. Fang B, McKeon A, Hinson SR, Kryzer TJ, Pittock SJ, Aksamit AJ, et al. Autoimmune glial fibrillary acidic protein astrocytopathy: a novel meningoencephalomyelitis. *JAMA Neurol*. (2016) 73:1297–307. doi: 10.1001/jamaneurol.2016.2549

2. Flanagan EP, Hinson SR, Lennon VA, Fang B, Aksamit AJ, Morris PP, et al. Glial fibrillary acidic protein immunoglobulin G as biomarker of autoimmune astrocytopathy: analysis of 102 patients. *Ann Neurol*. (2017) 81:298–309. doi: 10.1002/ana.24881

3. Long Y, Liang J, Xu H, Huang Q, Yang J, Gao C, et al. Autoimmune glial fibrillary acidic protein astrocytopathy in Chinese patients: a retrospective study. *Eur J Neurol*. (2018) 25:477–83. doi: 10.1111/ene.13531

4. Xu H, Huang Q, Xiao X, Liu T, Chen B, Yang H, et al. Magnetic resonance imaging of the spinal cord and clinical characteristics in patients with autoimmune glial fibrillary acidic protein astrocytopathy. *Chin J Neurol*. (2019) 3:92–7. doi: 10.3760/cma.j.issn.1006-7876.2019.02.003

5. Banwell B, Bennett JL, Marignier R, Kim HJ, Brilot F, Flanagan EP, et al. Diagnosis of myelin oligodendrocyte glycoprotein antibody-associated disease: international MOGAD panel proposed criteria. *Lancet Neurol*. (2023) 22:268–82. doi: 10.1016/S1474-4422(22)00431-8

6. Jurynczyk M, Messina S, Woodhall MR, Raza N, Everett R, Roca-Fernandez A, et al. Clinical presentation and prognosis in MOG-antibody disease: a UK study. *Brain*. (2017) 140:3128–38. doi: 10.1093/brain/awx276

7. Mariano R, Messina S, Kumar K, Kuker W, Leite MI, Palace J. Comparison of clinical outcomes of transverse myelitis among adults with myelin oligodendrocyte glycoprotein antibody vs aquaporin-4 antibody disease. *JAMA Netw Open*. (2019) 2:e1912732. doi: 10.1001/jamanetworkopen.2019.12732

8. Greenberg BM. Treatment of acute transverse myelitis and its early complications. *Continuum*. (2011) 17:733–43. doi: 10.1212/01.CON.0000403792.36161.f5

9. Sechi E, Morris PP, McKeon A, Pittock SJ, Hinson SR, Weinshenker BG, et al. Glial fibrillary acidic protein IgG related myelitis: characterisation and comparison with aquaporin-4-IgG myelitis. *J Neurol Neurosurg Psychiatry*. (2019) 90:488–90. doi: 10.1136/jnnp-2018-318004

10. Yao H, Li H, Jiang L, Long Y, Yang X. Coexisting autoimmune glial fibrillary acidic protein (GFAP)-IgG and aquaporin 4 (AQP4)-IgG in patients with myelitis. *J Sun Yat-Sen Univ*. (2022) 43:607. doi: 10.13471/j.cnki.j.sun.yat-sen.univ(med.sci).20220414.001

11. Li J, Xu Y, Ren H, Zhu Y, Peng B, Cui L. Autoimmune GFAP astrocytopathy after viral encephalitis: a case report. *Mult Scler Relat Disord*. (2018) 21:84–7. doi: 10.1016/j.msard.2018.02.020

12. Handoko M, Hong W, Espineli E, Saxena K, Muscal E, Risen S. Autoimmune glial fibrillary acidic protein astrocytopathy following herpes simplex virus encephalitis in a pediatric patient. *Pediatr Neurol*. (2019) 98:85–6. doi: 10.1016/j.pediatrneurol.2019.05.010

13. Dubey D, Hinson SR, Jolliffe EA, Zekeridou A, Flanagan EP, Pittock SJ, et al. Autoimmune GFAP astrocytopathy: prospective evaluation of 90 patients in 1 year. *J Neuroimmunol*. (2018) 321:157–63. doi: 10.1016/j.jneuroim.2018.04.016

14. Dubey D, Pittock SJ, Krecke KN, Morris PP, Sechi E, Zalewski NL, et al. Clinical, radiologic, and prognostic features of myelitis associated with myelin oligodendrocyte glycoprotein autoantibody. *JAMA Neurol*. (2019) 76:301–9. doi: 10.1001/jamaneurol.2018.4053

15. Zhang Bao J, Huang W, Zhou L, Wang L, Chang X, Lu C, et al. Myelitis in inflammatory disorders associated with myelin oligodendrocyte glycoprotein antibody and aquaporin-4 antibody: a comparative study in Chinese Han patients. *Eur J Neurol*. (2021) 28:1308–15. doi: 10.1111/ene.14654

16. Kim KH, Kim SH, Hyun JW, Kim HJ. Clinical and radiological features of myelin oligodendrocyte glycoprotein-associated myelitis in adults. *J Clin Neurol*. (2022) 18:280–9. doi: 10.3988/jcn.2022.18.3.280

17. Ciron J, Cobo-Calvo A, Audoin B, Bourre B, Brassat D, Cohen M, et al. Frequency and characteristics of short versus longitudinally extensive myelitis in adults with MOG antibodies: a retrospective multicentric study. *Mult Scler*. (2020) 26:936–44. doi: 10.1177/1352458519849511

18. Sechi E, Buciuc M, Flanagan EP, Pittock SJ, Banks SA, Lopez-Chiriboga AS, et al. Variability of cerebrospinal fluid findings by attack phenotype in myelin oligodendrocyte glycoprotein-IgG-associated disorder. *Mult Scler Relat Disord*. (2021) 47:102638. doi: 10.1016/j.msard.2020.102638

19. Kimura A, Takekoshi A, Yoshikura N, Hayashi Y, Shimohata T. Clinical characteristics of autoimmune GFAP astrocytopathy. *J Neuroimmunol*. (2019) 332:91–8. doi: 10.1016/j.jneuroim.2019.04.004

20. Shan F, Long Y, Qiu W. Autoimmune glial fibrillary acidic protein astrocytopathy: a review of the literature. *Front Immunol*. (2018) 9:2802. doi: 10.3389/fimmu.2018.02802

21. Liu L, Fang B, Qiao Z, Di X, Ma Q, Zhang J, et al. Clinical manifestation, auxiliary examination features, and prognosis of GFAP autoimmunity: a Chinese cohort study. *Brain Sci*. (2022) 12:1662. doi: 10.3390/brainsci12121662

22. Iorio R, Damato V, Evoli A, Gessi M, Gaudino S, Di Lazzaro V, et al. Clinical and immunological characteristics of the spectrum of GFAP autoimmunity: a case series of 22 patients. *J Neurol Neurosurg Psychiatry*. (2018) 89:138–46. doi: 10.1136/jnnp-2017-316583

23. Salama S, Khan M, Shanechi A, Levy M, Izbudak I. MRI differences between MOG antibody disease and AQP4 NMOSD. *Mult Scler*. (2020) 26:1854–65. doi: 10.1177/1352458519893093

24. Chen C, Liu C, Fang L, Zou Y, Ruan H, Wang Y, et al. Different magnetic resonance imaging features between MOG antibody-and AQP4 antibody-mediated disease: a Chinese cohort study. *J Neurol Sci*. (2019) 405:116430. doi: 10.1016/j.jns.2019.116430

25. Sato DK, Callegaro D, Lana-Peixoto MA, Waters PJ, de Haidar Jorge FM, Takahashi T, et al. Distinction between MOG antibody-positive and AQP4 antibody-positive NMO spectrum disorders. *Neurology*. (2014) 82:474–81. doi: 10.1212/WNL.0000000000000101

26. Zhu B, Sun M, Yang T, Yu H, Wang L. Clinical, imaging features and outcomes of patients with anti-GFAP antibodies: a retrospective study. *Front Immunol*. (2023) 14:1106490. doi: 10.3389/fimmu.2023.1106490

27. Mittal MK, Rabinstein AA, Hocker SE, Pittock SJ, Wijdicks EF, McKeon A. Autoimmune encephalitis in the ICU: analysis of phenotypes, serologic findings, and outcomes. *Neurocrit Care.* (2016) 24:240–50. doi: 10.1007/s12028-015-0196-8

28. Lerch M, Bauer A, Reindl M. The potential pathogenicity of myelin oligodendrocyte glycoprotein antibodies in the optic pathway. *J Neuroophthalmol*. (2023) 43:5–16. doi: 10.1097/WNO.0000000000001772

29. Irani SR, Gelfand JM, Al-Diwani A, Vincent A. Cell-surface central nervous system autoantibodies: clinical relevance and emerging paradigms. *Ann Neurol*. (2014) 76:168–84. doi: 10.1002/ana.24200

30. Yuan Z, Li H, Huang L, Fu C, Chen Y, Zhi C, et al. CD8+ T-cell predominance in autoimmune glial fibrillary acidic protein astrocytopathy. *Eur J Neurol*. (2021) 28:2121–5. doi: 10.1111/ene.14778

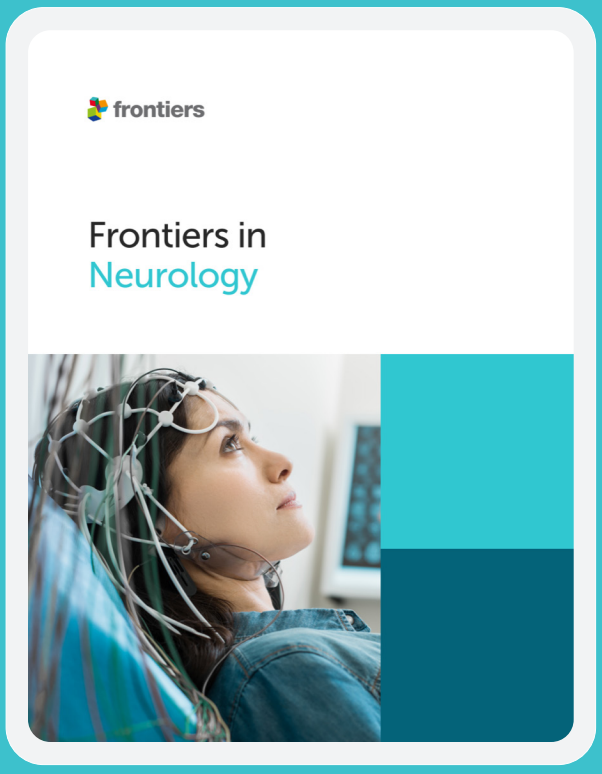
Frontiers in Neurology

Explores neurological illness to improve patient care

The third most-cited clinical neurology journal explores the diagnosis, causes, treatment, and public health aspects of neurological illnesses. Its ultimate aim is to inform improvements in patient care.

Discover the latest Research Topics

See more →



Frontiers

Avenue du Tribunal-Fédéral 34
1005 Lausanne, Switzerland
frontiersin.org

Contact us

+41 (0)21 510 17 00
frontiersin.org/about/contact