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Influence of lifestyle factors in the management of diabetes mellitus

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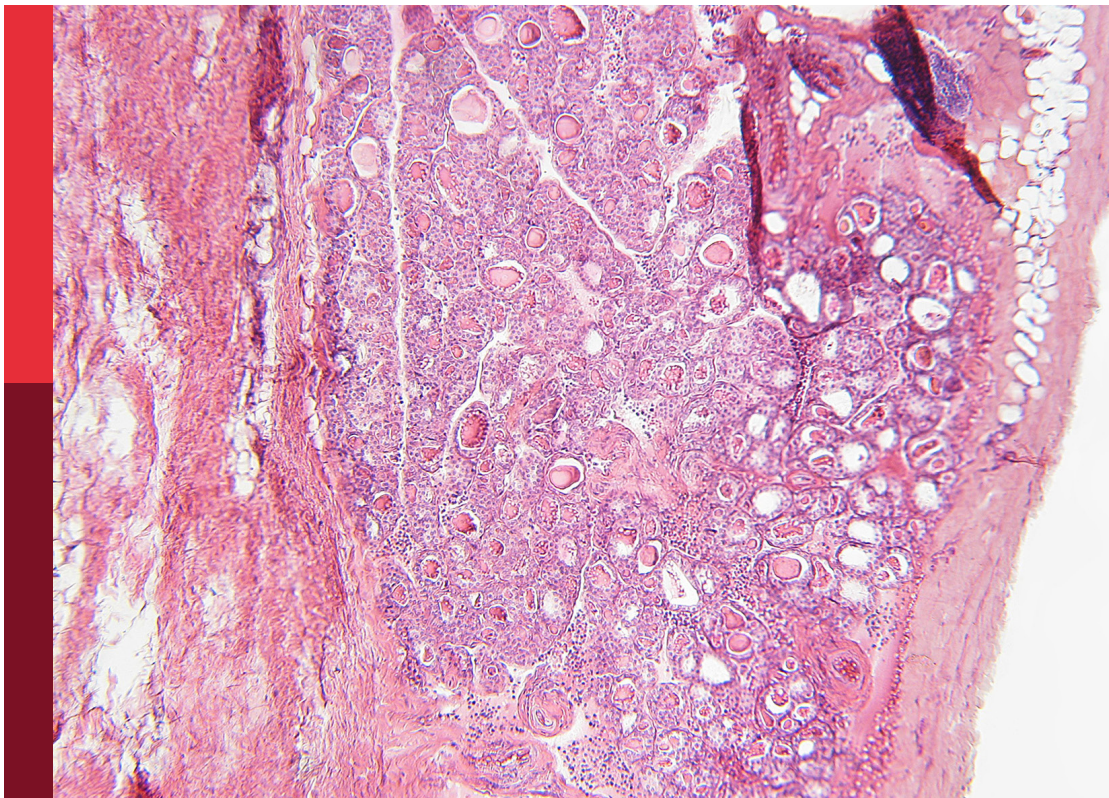

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Influence of lifestyle factors in the management of diabetes mellitus

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Editorial: Influence of lifestyle factors in the management of diabetes mellitus

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diabetes mellitus, inflammation, oxidative stress, drug therapy, lifestyle

Editorial on the Research Topic
Influence of lifestyle factors in the management of diabetes mellitus

Diabetes mellitus (DM) is a multi-factorial metabolic disease that affects approximately 462 million individuals, corresponding to 6.28% of the world's population (4.4% of those aged 15–49 years, 15% of those aged 50–69, and 22% of those aged 70+), with a prevalence rate of 6,059 cases per 100,000 (1). DM accounts for over 1 million deaths per year, making it the ninth leading cause of mortality (1). The DM burden is rising globally and at a much faster rate in developed regions, such as Western Europe, with equal gender distribution, and the incidence peaks at approximately 55 years of age (1).

In this scenario, we expect a global prevalence of DM of approximately 7,079 individuals per 100,000 by 2030 (1). To date, the rising burden of DM is a major concern in healthcare worldwide, and there is a need for urgent public health and clinical preventive measures (1). In non-Western countries, in a cohort of 215,041 elderly adults living in China (102,692 men and 112,349 women), authors reported a prevalence of self-reported diabetes of approximately 8.7%, with the highest prevalence in Beijing (20.8%) and the lowest prevalence in Xizang (0.9%), (Hu et al.). Notably, urban areas, older age, female, higher income, poor sleep quality, and some other factors were potential risk factors for diabetes (Hu et al.). Conversely, sociodemographic and behavioral factors could be linked to the low awareness of DM medication (Khoiry et al.). Indeed, irregular blood glucose monitoring without any comorbidity, never having any general medical checkup, 26–35 years of age, 36–45 years of age, and having no health insurance coverage were significantly associated with low awareness of diabetes medication (Khoiry et al.).

Notably, the complexity of oral anti-diabetic drug (OAD) regimens could affect the quality of life (QOL) and treatment satisfaction (Chang et al.). Indeed, a study conducted in Taiwan showed a significantly greater effect on QOL among patients with fewer OAD classes and higher treatment satisfaction (Chang et al.). Parallelly, new anti-DM treatments could effectively ameliorate the glucose and blood lipid metabolism via effects on the intestinal flora in type 2 DM (T2DM) patients (Peng, X. et al.). In this context, the

Cyclocarya paliurus leaves' extracts (CP) display a more beneficial effect in the alleviation of T2DM-associated metabolic phenotypes than glipizide by regulating gut microbiota and metabolites in T2DM patients, with no significant effects on liver and kidney function (Peng, X. et al.). The liver activity could itself play a relevant role in the metabolic abnormalities of DM by controlling lipid and glucose homeostasis and feeding metabolites (Chen et al.). Indeed, authors observed that feeding could induce the release of hepatokines, which regulates glucose and lipid metabolism, and these feeding-induced hepatokines act on multiple organs to regulate glucolipotoxicity and thus influence the development of T2DM (Chen et al.).

Regarding the negative role played by lipid metabolism in DM patients, authors found that clonal hematopoiesis could be evaluated as a novel risk factor for T2DM patients with hypercholesterolemia (Kim et al.). Indeed, the clonal hematopoiesis of indeterminate potential (CHIP) is associated with atherosclerosis, cardiovascular disease (CVD), and new-onset T2DM (Kim et al.). Notably, the subjects with CHIP and hyper-LDL-cholesterolemia had approximately twice the risk of diabetes than subjects without CHIP and with low LDL cholesterol (Kim et al.). Thus, there could be a synergism between CHIP and high LDL cholesterol as a high-risk factor for diabetes (Kim et al.).

In this context, it is well known that DM causes an increase in inflammatory and oxidative stress, negatively affecting glucose homeostasis and insulin resistance and worsening clinical outcomes (2, 3). Indeed, over-inflammation/oxidative stress is a leading cause of atherosclerosis, atherosclerotic plaque fissuration, and CVDs in DM patients (2, 3), such as in the overall population (4, 5) and in gender-specific cohorts of patients (Wu et al.). Moreover, abnormalities of oxidative balance score (OBS) and high OBS are negatively associated with diabetes risk in a gender-dependent manner (Wu et al.).

Conversely, personality factors and health status influence resilience, and coping strategies could influence diabetic subjects (Rivera-Picón et al.). Indeed, concerning health status, the absence of pathology is related to using rational strategies more than to diagnosing diabetes (Rivera-Picón et al.). In the clinical setting, although diabetes care is improving, there are still cases that are poorly managed with adverse clinical outcomes (2–4). Indeed, DM is a leading cause of CVDs, and hospitalizations and deaths in developed countries (5). Conversely, DM could negatively affect other clinical conditions, such as cognitive decline (Wang et al.). Indeed, people with DM described misconceptions about their cognitive decline and suffered from them during disease management (Wang et al.). Intriguingly, authors found that poor glycemic control could impair the brain networks responsible for learning, memory, and controlled reactivity to food in adolescents with type 1 diabetes whose glycemic control is poor (Litmanovitch et al.). Thus, we need to support disease management with cognitive decline in clinical practice (Wang et al.), and improve glycemic control to ameliorate brain functions (Litmanovitch et al.).

However, from here, we could say that the DM prevention necessitates an integrated and holistic strategy to ameliorate glycemic control based on the condition's cause. Indeed, inadequate glycemic management impacts the usage of healthcare resources, medical expenses, and death rates dramatically. Furthermore, in the current topic, we focused, on one side, on the prevention of DM (lifestyle modification) and its complications (best glycemic control and anti-diabetic medications) and, on the other side, on the early treatment of CVDs linked to DM (drug and interventional treatments). As an example, regarding lifestyle modification, we could promote weight loss and physical activity (Brinkmann et al.). Indeed, obesity is a major risk factor for DM, which is, in turn, a significant risk factor for CVDs such as coronary artery disease and stroke (Chung et al.). Thus, in a study population of 24,346 participants, of whom 8,334 (mean age, 50.6 ± 11.0 years) were male and 16,012 (mean age, 50.5 ± 10.1 years) were female, authors found strong associations between the studied obesity-related indices and incidence of DM, and sex differences (Chung et al.). Hence, to better control DM, reducing body weight may be beneficial in addition to lifestyle modifications, diet control, and pharmacological interventions (Chung et al.).

Conversely, after a certain period of low-volume high-intensity interval training (LVHIIT), glycemic control, insulin resistance, body weight, lipid profile, and cardiorespiratory outcomes were significantly improved in T2DM patients (Peng, Y. et al.). This concept has been applied also to a cohort of 4,196 German company employees and divided into three risk groups based on their European Society of Cardiology–Systematic Coronary Risk Evaluation score (ESC-SCORE), (Brinkmann et al.). In these subjects, authors found that the ESC-SCORE changes from baseline differed significantly between the groups, with the intervention group achieving more favorable results in all follow-up visits 6, 12, 24, and 36 months later (at each time point: ITT: $p < 0.001$; PP: $p \leq 0.010$) (Brinkmann et al.). Thus, they found the feasibility of attracting employees with pre-DM/DM at high cardiovascular (CVD) mortality risk to participate in a multimodal lifestyle program following a free CVD mortality risk screening at their workplace (Brinkmann et al.). The lifestyle intervention used in the PreFord study shows high potential for improving the health of company employees with pre-DM/DM in the long term (Brinkmann et al.). In the lifestyle intervention, we could report the laughter yoga and its effects on glycemic control among individuals with T2DM (Hirosaki et al.). The proposed study intervention consisted of a 12-week laughter yoga program that resulted in (in the laughter yoga group) a significant improvement in HbA1c levels and an increase in sleep duration (Hirosaki et al.). However, having fun could be a self-care intervention to ameliorate DM status (Hirosaki et al.).

According to the published articles on the current Research Topic, we could report that lifestyle factors could negatively influence and condition the management and clinical outcomes of DM. The more robust control of lifestyle factors could, on one side, result in the amelioration of glucose homeostasis and insulin resistance in DM patients. This could be seen as a glucose-dependent effect. On the other side, stronger control of lifestyle factors could reduce inflammatory/oxidative stress in DM patients (Wu et al.). This could be evidenced by the glucose-independent effect of lifestyle factor control. Finally, the control of lifestyle factors could reduce CVD disease and mortality via glucose-dependent and -independent effects. Furthermore, we might promote the control of lifestyle factors as a relevant therapeutic strategy for managing and treating patients living with DM.

Author contributions

CS: Conceptualization, Investigation, Writing – original draft, Writing – review & editing. GS: Writing – review & editing, Investigation. ND'O: Investigation, Writing – review & editing.

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.

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References

1. Khan MAB, Hashim MJ, King JK, Govender RD, Mustafa H, Al Kaabi J. Epidemiology of type 2 diabetes - global burden of disease and forecasted trends. *J Epidemiol Glob Health* (2020) 10(1):107–11. doi: 10.2991/jegh.k.191028.001

2. Sardu C, Trotta MC, Sasso FC, Sacra C, Carpinella G, Mauro C, et al. SGLT2-inhibitors effects on the coronary fibrous cap thickness and MACEs in diabetic patients with inducible myocardial ischemia and multi vessels non-obstructive coronary artery stenosis. *Cardiovasc Diabetol* (2023) 22(1):80. doi: 10.1186/s12933-023-01814-7

3. Sardu C, Paolisso G, Marfella R. Inflammatory related cardiovascular diseases: from molecular mechanisms to therapeutic targets. *Curr Pharm Des* (2020) 26 (22):2565–73. doi: 10.2174/1381612826666200213123029

4. Paolisso P, Bergamaschi L, Gragnano F, Gallinoro E, Cesaro A, Sardu C, et al. Outcomes in diabetic patients treated with SGLT2-Inhibitors with acute myocardial infarction undergoing PCI: The SGLT2-I AMI PROTECT Registry. *Pharmacol Res* (2023) 187:106597. doi: 10.1016/j.phrs.2022.106597

5. Marfella R, Sardu C, D'Onofrio N, Fumagalli C, Scisciola L, Sasso FC, et al. SGLT-2 inhibitors and in-stent restenosis-related events after acute myocardial infarction: an observational study in patients with type 2 diabetes. *BMC Med* (2023) 21(1):71. doi: 10.1186/s12916-023-02781-2

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Prevalence and potential risk factors of self-reported diabetes among elderly people in China: A national cross-sectional study of 224,142 adults

Xing Hu[1,2], Lingbing Meng[3], Zhimin Wei[1], Hongxuan Xu[3], Jianyi Li[3], Yingying Li[3], Na Jia[3], Hui Li[3], Xin Qi[3], Xuezhai Zeng[3], Qiuxia Zhang[4], Juan Li[5] and Deping Liu[3]*

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Aim: To evaluated the prevalence and potential risk factors of self-reported diabetes among the elderly in China, by demographic data, socioeconomic factors, and psychological factors.

Methods: Descriptive analysis and Chi-square analysis were used to assess the prevalence and variation between self-reported diabetes and non-diabetes by demographic data, living habits, socioeconomic factors and comorbidities. Univariate and multivariate logistic regression were used to describe the odds ratios (OR) of diabetes prevalence in different groups, while stratification analysis was performed to describe prevalence based on gender, age, and urban/rural areas.

Results: 215,041 elderly adults (102,692 males and 112,349 females) were eventually included in the analysis. The prevalence of self-reported diabetes among the elderly in China is about 8.7%, with the highest prevalence in Beijing (20.8%) and the lowest prevalence in Xizang (0.9%). Logistic regression analysis showed that urban area ($P < 0.001$), older age (65–84 years old, $P < 0.001$), female ($P < 0.001$), higher income($P < 0.001$), poor sleep quality ($P = 0.01$) and some other factors were potential risk factors for diabetes.

Conclusions: This study illustrates the prevalence and potential risk factors of diabetes among the elderly in China Meanwhile, these results provide information to assist the government in controlling non-communicable diseases in the elderly.

KEYWORDS

prevalence, self-reported, diabetes, elderly, national cross-sectional study

Introduction

A growing aging population in China is one of the key challenges facing public healthcare in the country. In fact, China has already become an aging society. In 2019, there were 164.5 million citizens aged 65 or older, including 26 million aged 80 years or more (1). It is predicted that the total population will reach 1.40–1.44 billion by 2030 and 1.29–1.40 billion by 2050 according to a study by the Chinese Center for Disease Control (CDC). The proportion of elderly individuals aged 65 years or more was continuously increasing, from 6.96% in 2000 to 8.87% in 2010, and 13.50% in 2020; this age group will comprise 20% of the total population by 2033 and 30% by 2050 from the Fifth Population Census to the Seventh Population Census (2).

Meanwhile, age-related diseases such as diabetes and complications will impose a significant burden on family and public healthcare systems (3). In 2019, it was estimated that 19.3% of people aged 65–99 years (135.6 million, 95% confidence interval (CI): 107.6–170.6 million) live with diabetes globally. Over the next two decades, the number of people with diabetes will grow from 195.2 million to 276.2 million worldwide (4). In 2015, diabetes cost US$ 1.31 trillion (95% CI 1.28–1.36), or 1.8% (95% CI 1.8–1.9) of the global economy (5). China has experienced a dramatic increase in diabetes prevalence, (6), from 2.5% in 1994 to 9.7% in 2008, and to 11.6% in 2010 (7–9). As of 2019, there are 116.4 million adults in China with diabetes, representing approximately 12.8% of its adult population (10, 11). Approximately USD 165 billion will be spent on diabetes-related health care in 2021, USD 185 billion in 2035, and USD 193 billion in 2045 (11).

The biopsychosocial model was proposed by Engel (12). Cultural, social, and psychological factors are linked with people's health (12). It is known that living conditions and lifestyle are important variables influencing the onset and progress of diabetes. However, previous studies about the prevalence of diabetes had several limitations. First, Most of these studies are regional studies carried out in east China, North China or South China (13–16). Second, the prevalence of diabetes in the general population has been studied more than in the elderly (17). Thirdly, most of the studies have only focused on the biological model of patients (18, 19). And there is little detailed information about socioeconomic factors and psychological factors. For governmental precision medical policies, it is extremely important to find out the comprehensive situation of diabetes patients.

In recent studies, some socioeconomic, lifestyle, and metabolic factors have been identified as risk factors for diabetes (20–22). From an another perspective, this study comprehensively evaluated the prevalence of diabetes among elderly Chinese patients by biomedical factors (age, gender, smoking, alcohol consumption, sleep quality, exercise, comorbidities) and social-psychological factors (education level, marital status, living alone, medical insurance, gainful employment, economic status, and spiritual and cultural life) to find out the awareness and potential risk factors among elderly diabetes patients.

Methods

Study design and participants

Government-affiliated China Research Center on Aging is one of China's leading aging research institutions. China Research Centre on Aging initiated the Sample Survey of the Aged Population in Urban and Rural China (SSAPUR) project in 2000. During the survey, the socioeconomic and health characteristics of elderly people over the age of 60 were investigated. This major national condition survey of the elderly in China was followed by longitudinal surveys in 2006 and 2010, the sample size was expanded and resampled in 2015 by Office of the China National Committee on Aging. All four surveys used a similar research design. The 2000 survey included 18,987 observations, the 2006 survey included 18,458 observations, and the 2010 survey included 18,689 observations.

The present study is based on data from the fourth SSAPUR, which was an extensive and large investigation survey of elderly people in China (comprising individuals aged ≥60 years, who were permanent residents and nationally representative; the survey was carried out for 1 month, from 1–31 August 2015. This investigation adopted a stratified multistage and probability proportional to size (PPS) sampling method, with regional sample sizes selected according to the area's proportion of people aged 60 years or more. The first time such a large number has been collected, including all provinces, autonomous regions, municipalities, and Xinjiang Production and Construction Corps across the country, covering 466 counties (districts), 1864 townships (sub-districts), and 7,456 village (residential) committees. The SSAPUR is China's largest elderly population database.

The fourth SSAPUR questionnaire was a large-scale epidemiological survey, which had been used in the Global Burden of Disease study and the World Health Survey. The survey covered nine aspects, including basic demographic information, family status, health, care and nursing services, economic status, social participation, rights protection, livable environment, and spiritual and cultural life (including psychology). Details of the fourth SSAPUR study design and the sampling method are provided in the Supplemental material (Supplements 1, 2).

The research protocol has been approved by the Ethical Review Committee of Beijing Hospital (No.2021BJYYEC-294-01) and approved by National Bureau of Statistics (No [2014] 87). All participants have provided written informed consent.

Date collection

All data were collected by trained study personnel in accordance with standardized protocols. On the cover of each questionnaire, there was a unique number, start and end time, and the signatures of the surveyors. Due to the huge amount of information gathered, we removed unnecessary information, such as commuting mode and children's work, according to the research purpose, and retained the demographic characteristics, health, social participation, family lives, and psychological information.

At baseline, demographic characteristics included gender, age, education level, household registration, and marital status. The "age" field was filled in, either according to an individual's ID card in the first instance or based on interviews with the senior citizen or his/her family members if he/she did not have an ID card. "Household registration" refers to agricultural household registration or non-agricultural household registration, either written in the household register or determined by the investigators. Education level was classified as follows: uneducated (never received school education at any level or of any type provided by the state or other institutions running schools); primary education (highest level of education received was primary school, whether in school, graduated, or dropped out); junior high-school education (highest level of education received was junior high-school, whether in school, graduated, or dropped out); high-school education (highest level of education received by a person, whether in school, graduated, or dropped out, including general high school, vocational high school, or secondary professional school); junior college (highest level of education received was at junior college); bachelor's degree or above (highest level of education received was a bachelor's degree or above).

Smoking was categorized as never smoked and other situations (including former and current smokers). Alcohol consumption was categorized as never or occasionally, 1–2 times a week, at least three times a week, or often drunk. Sleep quality divided by sleep time cannot describe sleep quality well. In this study, Sleep quality was categorized as very good, relatively good, average, relatively poor, very poor which sorted by elderly themselves. Exercise refers to all types of physical activities that are carried out consciously for the purpose of fitness, but does not include housework or farming. Medical insurance refers to the components of China's medical insurance system (basic medical insurance for urban workers, basic medical insurance for urban residents, and new rural cooperative medical care) and any other medical insurance. Gainful employment refers to the interviewed elderly people who were actually engaged in various production, management, or service activities to earn wages before the survey. Poverty was defined as having an annual household income of <6,000 yuan (US$ 963) in the previous year (2014). Economic status was selected according to the self-rating criteria of the interviewed elderly individuals. Public benefit activities cover safeguarding community public order, helping to mediate neighborhood disputes, safeguarding the community health environment, helping neighbors, caring about educating the next generation (not including educating your own grandchildren), and participating in cultural and scientific promotion activities. Spiritual and cultural life includes watching TV/listening to the radio, reading books/ newspapers, going to the cinema or the theater, Tai Chi/health exercises, playing gateball/table tennis/badminton, or playing mahjong/cards/chess. Chronic diseases, including malignant tumor, cataract/glaucoma, hypertension, cardiac–cerebral vascular disease (CCVD), osteoarthrosis, and chronic obstructive pulmonary disease (COPD), and were self-reported by the interviewed elderly individuals.

Definition

As a cross-sectional study, the prevalence of diabetes among elderly in this study include those diagnosed by health professionals in the past regardless of subtype (Type 1, Type 2 or any other subtypes), either on diet control, oral hypoglycemic drugs and/or injective insulin. Other chronic diseases which were used as independent variable factors, including malignant tumor, cataract/glaucoma, hypertension and so on, refer to self-reported diseases that has been definitively diagnosed by health professionals before.

Statistical analysis

From a total of 224,142 cases, we excluded those with missing data, including 9,084 cases whose diabetes status was not clear and 17 individuals who had more than 10 missing independent variables.

The prevalence of diabetes was described by province, in descending order from largest to smallest. Baseline characteristics and other factors were summarized as numbers with proportions. The statistical significance of differences was assessed using Chi-square analysis for categorical variables and a *post hoc* two-tailed Newman–Keuls test when two or more groups were compared.

In univariate logistic regression analysis, demographic data (household registration, age, gender and education level), living habits (smoking, alcohol consumption, sleep quality and exercise), socioeconomic factors (medical insurance, gainful employment, poverty, economic status, public benefit activities, spiritual cultural life), and comorbidities (malignant tumor, cataract/glaucoma, hypertension, CCVD, COPD) were analyzed as independent variables. A P-value <0.05 was considered statistically significant. After that, statistically significant independent variables in the univariate logistic regression analysis were included in the multivariate regression

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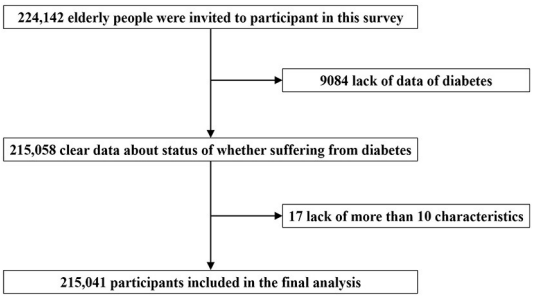

FIGURE 1
Flowchart of participants on self-reported diabetes of the fourth SSAPUR study.

analysis. Stratification analysis was also performed, based on gender, age, and residing in urban or rural areas.The prevalence of diabetes after stratification was tested by chi-square test. A *P*-value <0.05 was considered statistically significant. Using the methods of Robert Newcombe, the lower and upper limits of the 95% confidence intervals for the proportion of diabetics were calculated. All statistical analyses were performed using SPSS 24.0 (IBM Corp., Armonk, NY, USA).

Results

Finally, a total of 215,041 participants (102,692 male and 112,349 female) were included in this analysis (Figure 1). Table 1 shows the number of participants and prevalence of diabetes among the elderly in 31 provinces in China. Hong Kong, Macao and Taiwan were not included in this survey. The prevalence of diabetes is 8.7% among the elderly in China, with the highest prevalence in Beijing (20.8%), Tianjin (17.2%), and Shanghai (16%), and the lowest prevalence in Xizang (0.9%), Guangxi (3.4%), and Hainan (3.9%). Figure 2 shows the distribution of the prevalence of diabetes.

The prevalence distribution of diabetes with related factors is shown in Table 2. In urban areas, diabetes prevalence was significantly higher than in rural areas (11.4 vs. 5.8%, $P = 0.001$). The relationship between age and diabetes exhibited a "spoon-shaped" relationship. The prevalence of diabetes was highest in the 70–74 years age group (9.7%), followed by 7.7% and 7.0% in the groups aged 60–64 and ≥85 years, respectively. Females had a higher prevalence of diabetes than males (9.8 vs. 7.6%, $P < 0.001$). The prevalence of diabetes varied among the elderly with different education levels, from uneducated to bachelor's degree or above, with a gradually increasing trend from 7.4 to 16.2% ($P < 0.001$). The prevalence of diabetes also differed by smoking, alcohol consumption, sleep quality, and exercise. There was a difference between the prevalence of diabetes by self-reported economic status ($P < 0.001$), with the "very generous" having the highest prevalence of diabetes (10.1%) and the "relatively difficult" group having the lowest prevalence (8.2%). Diabetes prevalence did not differ significantly between patients with and without cardio-cerebral vascular disease, osteoarthritis, or COPD. The prevalence of diabetes in the hypertensive group (36.9%) was higher than that in the non-hypertensive group (14.6 vs. 5.2%, $P < 0.001$). Patients with cataract/glaucoma (16%) had a higher combined diabetes rate than those without cataract/glaucoma (12.9 vs. 7.9%, $P < 0.001$). Malignant tumor

TABLE 1 The prevalence of diabetes in the elderly of China.

*Province	Participants (proportion)	Diabetes	
		No (proportion)	Yes (proportion)
Total	215,041(100%)	196,319(91.3%)	18,722(8.7%)
Beijing	3,359 (1.6%)	2,660(79.2%)	699(20.8%)
Tianjin	1,920 (0.9%)	1,589(82.8%)	331(17.2%)
Shanghai	4,296 (2.0%)	3,607(84.0%)	689(16.0%)
Sinkiang	2,378 (1.1%)	2,068(87.0%)	310(13.0%)
Qinghai	957 (0.4%)	838(87.6%)	119(12.4)
Inner Mongolia	3,343 (1.6%)	2,930(87.6%)	413(12.4%)
Fujian	5,247 (2.4%)	4,667(88.9%)	580(11.1%)
Zhejiang	9,595 (4.5%)	8,574(89.4%)	1,021(10.6%)
Liaoning	8,573 (4.0%)	7,711(89.9%)	862(10.1%)
Shaanxi	5,754 (2.7%)	5,195(90.3%)	559(9.7%)
Hebei	10,701 (5.0%)	9,683(90.5%)	1,018(9.5%)
Jiangsu	15,629 (7.3%)	14,150(90.5%)	1,479(9.5%)
Jilin	4,222 (2.0%)	3,825(90.6%)	397(9.4%)
Shandong	17,718 (8.2%)	16,134(91.1%)	1,584(8.9%)
Sichuan	16,150 (7.5%)	14,713(91.1%)	1 437(8.9%)
Shanxi	5,250 (2.4%)	4,786(91.2%)	464(8.8%)
Chongqing	6,225 (2.9%)	5,684(91.3%)	541(8.7%)
Anhui	11,240 (5.2%)	10,307(91.7%)	933(8.3%)
Heilongjiang	5,610 (2.6%)	5,147(91.7%)	463(8.3%)
Ningxia	956 (0.4%)	878(91.8%)	78(8.2%)
Hubei	3,551 (1.7%)	3,272(92.1%)	279(7.9%)
Hunan	11,911 (5.5%)	10,982(92.2%)	929(7.8%)
Henan	14,682 (6.8%)	13,553(92.3%)	1,129(7.7%)
Guangdong	13,350 (6.2%)	12,504(93.7%)	846(6.3%)
Jiangxi	6,214 (2.9%)	5,837(93.9%)	377(6.1%)
Gansu	3,344 (1.6%)	3,145(94.0%)	199(6.0%)
Guizhou	5,705 (2.7%)	5,407(94.8%)	298(5.2%)
Yunnan	6,670 (3.1%)	6,322(94.8%)	348(5.2%)
Hainan	1,432 (0.7%)	1,376(96.1%)	56(3.9%)
Guangxi	8,134 (3.8%)	7,858(96.6%)	276(3.4%)
Tibet	925 (0.4%)	917(99.1%)	8(0.9%)

*Hong Kong, Macao and Taiwan were not included in this survey.

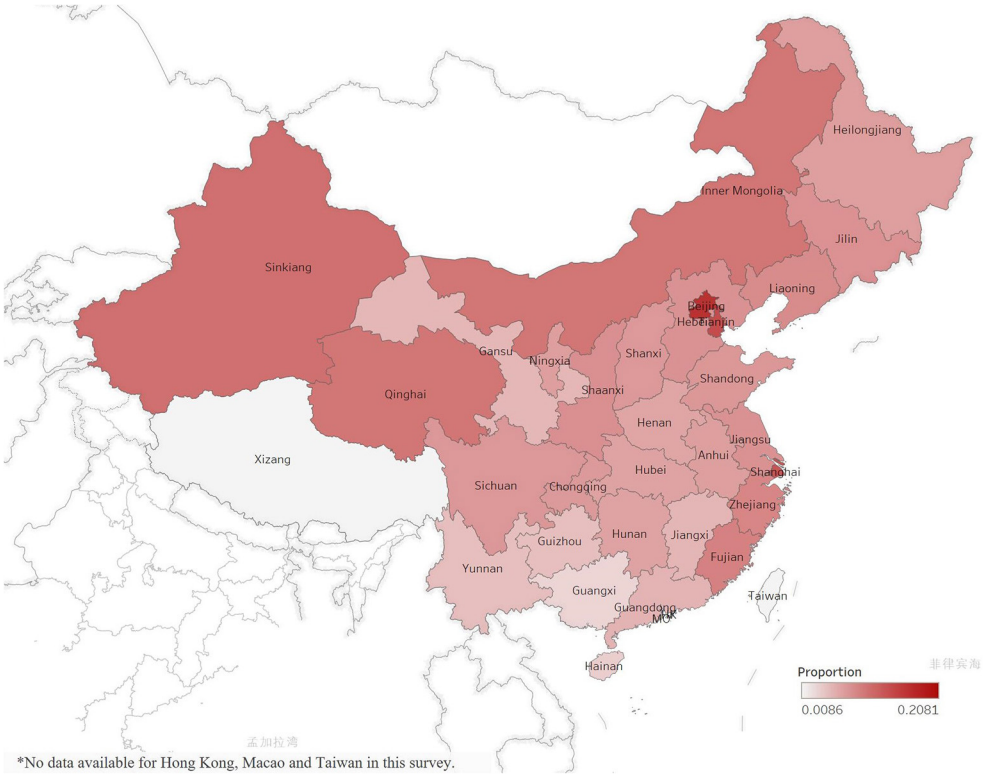

FIGURE 2
Geographic distribution of diabetes prevalence in 31 selected provinces/municipalities in the fourth SSAPUR study. Hong Kong, Macao and Taiwan were not included in this survey.

patients also had a higher prevalence of diabetes (1.1%) (11.2 vs. 8.7%, $P < 0.001$).

Table 3 presents the results of univariate and multivariate logistic regression analysis. Univariate logistic regression showed that the odds ratio (OR) for diabetes in rural areas was 0.48 (95% confidence interval (CI): 0.47–0.50, $P < 0.001$) compared with urban areas. The ORs of diabetes in all age groups increased and then decreased, with the OR of diabetes in the 75–79 years group being 1.23 (95%CI: 1.17–1.29, $P < 0.001$)compared with the 60–64 years group, and the OR of diabetes in the group aged ≥85 years being 0.89 (95%CI: 0.82–0.96, $P < 0.001$). The OR for diabetes in men was 0.76 (95%CI: 0.73–0.78, $P < 0.001$) based on women. The risk of diabetes increased with higher education level. Compared with non-smokers, smokers had a lower prevalence of diabetes.The OR for "very poor" sleep quality was 1.79 (95%CI: 1.41–2.28, $P < 0.001$) compared with "very good" sleep quality. The odds of diabetes among the elderly from poor families with an annual household income <6,000 yuan (US$ 963) was 0.62 times that of non-poor families (95%CI: 0.59–0.65, $P < 0.001$). Those with malignant tumors (OR: 1.32, 95%CI: 1.17–1.50, $P < 0.001$), cataract/glaucoma (OR: 1.73, 95%CI: 1.66–1.79, $P < 0.001$), and high blood pressure (OR: 3.10, 95%CI: 3.01–3.20, $P < 0.001$) showed a higher prevalence of diabetes than people without these diseases. In this study, the proportion of diabetes patients with CCVD or osteoarthrosis was not statistically significant compared with the proportion of people without such diseases (Figure 3). In multivariate logistic regression analysis, the statistically significant factors were included. The results are presented in Table 3 and they were all statistically significant (Figure 4).

By stratifying by gender, we explored the prevalence and differences between parameters and diabetes (Table 4). A total of 10,964 female individuals with diabetes were identified among 112,349 female participants. Diabetes prevalence varied between urban and rural areas among females (12.0 vs. 7.2%, $P < 0.001$) and among age groups ($P < 0.001$). In addition, a total of 7,758 male individuals with diabetes were identified among 102,692 male participants. Among males, the prevalence of diabetes varied throughout urban and rural areas (10.7 vs. 4.3%, $P < 0.001$) and among age groups ($P < 0.001$). Regardless of gender, the prevalence of diabetes differed by comorbidities such as cataract glaucoma ($P < 0.001$) and hypertension ($P < 0.001$). However, The prevalence of diabetes in women with or without COPD was significantly different (12.8 vs. 9.7%, $P < 0.001$), but this was not seen in men (7.5 vs. 7.6%, $P = 0.728$). In males

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TABLE 2 The related factors of proportion with (or without) diagnosed diabetes.

Factor	Total (proportion)	Diabetes(proportion)		*P*-value
		No	Yes	
Household registration				<0.001
Urban area	111,940(52.1%)	99,210(88.6%)	12,730(11.4%)	
Rural area	103,101(47.9%)	97,109(94.2%)	5,992(5.8%)	
Age(years)				<0.001
60–64	70,913(33.0%)	65,419(92.3%)	5,494(7.7%)	
65–69	50,723(23.6%)	45,999(90.7%)	4,724(9.3%)	
70–74	35,760(16.6%)	32,300(90.3%)	3,460(9.7%)	
75–79	28,131(13.1%)	25,502(90.7%)	2,629(9.3%)	
80–84	18,426(8.6%)	16,783(91.1%)	1,643(8.9%)	
≥85	11,088(5.2%)	10,316(93.0%)	772(7.0%)	
Gender				<0.001
Female	112,349(52.2%)	101,385(90.2%)	10,964(9.8%)	
Male	102,692(47.8%)	94,934(92.4%)	7,758(7.6%)	
Education level				<0.001
Uneducated	63,102(29.4%)	58,439(92.6%)	4,663(7.4%)	
Primary education	89,059(41.5%)	82,186(92.3%)	6,873(7.7%)	
Junior high school	40,508(18.9%)	36,245(89.5%)	4,263(10.5%)	
High school	15,087(7.0%)	13,250(87.8%)	1,837(12.2%)	
Junior College	4,269(2.0%)	3,609(84.5%)	660(15.5%)	
Bachelor degree or above	2,322(1.1%)	1,946(83.8%)	376(16.2%)	
Marital status				<0.001
Married	155,973(72.5%)	142,133(91.1%)	13,840(8.9%)	
Widowed	54,142(25.2%)	49,539(91.5%)	4,603(8.5%)	
Divorce	1,795(0.8%)	1,630(90.8%)	165(9.2%)	
Never married	3,131(1.5%)	3,017(96.4%)	114(3.6%)	
Live alone				<0.001
No	186,118(86.6%)	169,755(91.2%)	16,363(8.8%)	
Yes	28,923(13.4%)	26,564(91.8%)	2,359(8.2%)	
Smoking				<0.001
No	14,533(6.8%)	13,110(90.2%)	1,423(9.8%)	
Yes	200,508(93.2%)	183,209(91.4%)	17,299(8.6%)	
Alcohol consumption				<0.001
Never or occasionally	211,936(98.6%)	193,394(91.3%)	18,542(8.7%)	
1-2 times a week	849(0.4%)	794(93.5%)	55(6.5%)	
At least 3 times a week	1,970(0.9%)	1,855(94.2%)	115(5.8%)	
Often drunk	286(0.1%)	276(96.5%)	10(3.5%)	
Sleep quality				<0.001
Very good	3,145(1.5%)	2,890(91.9%)	255(8.1%)	
Relatively good	6,531(3.0%)	6,006(92.0%)	525(8.0%)	
Average	200,588(93.3%)	183,141(91.3%)	17,447(8.7%)	
Relatively poor	3,993(1.9%)	3,605(90.3%)	388(9.7%)	
Very poor	784(0.4%)	677(86.4%)	107(13.6%)	
Exercise (per week)				<0.001
Never exercise	105,213(48.9%)	97,919(93.1%)	7,294(6.9%)	
Less than once	9,453(4.4%)	8,623(91.2%)	830(8.8%)	

(Continued)

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TABLE 2 (Continued)

Factor	Total (proportion)	Diabetes(proportion)		*P*-value
		No	Yes	
Once or twice	27,582(12.8%)	25,157(91.2%)	2,425(8.8%)	
Three to five times	26,339(12.2%)	23,702(90.0%)	2,637(10.0%)	
Six times and above	46,454(21.6%)	40,918(88.1%)	5,536(11.9%)	
Medical insurance				<0.001
No	1,960(0.9%)	1,852(94.5%)	108(5.5%)	
Yes	213,081(99.1%)	194,467(91.3%)	18,614(8.7%)	
Gainful employment				<0.001
No	193,528(90.0%)	175,946(90.9%)	17,582(9.1%)	
Yes	21513(10.0%)	20,373(94.7%)	1,140(5.3%)	
Poverty				<0.001
No	185,872(86.4%)	168,859(90.8%)	17,013(9.2%)	
Yes	29,169(13.6%)	27,460(94.1%)	1,709(5.9%)	
Economic status				<0.001
Very generous	2,738(1.3%)	2,462(89.9%)	276(10.1%)	
Relatively ample	31,721(14.8%)	28,815(90.8%)	2,906(9.2%)	
Basically enough	126,650(58.9%)	115,657(91.3%)	10,993(8.7%)	
Tougher	45,135(21.0%)	41,429(91.8%)	3,706(8.2%)	
Very difficult	8,797(4.1%)	7,956(90.4%)	841(9.6%)	
Public benefit activities				<0.001
No	117,267(54.5%)	106,276(90.6%)	10,991(9.4%)	
Yes	97,774(45.5%)	90,043(92.1%)	7,731(7.9%)	
Spiritual cultural life				<0.001
No	16,892(7.9%)	15,773(93.4%)	11,191(6.6%)	
Yes	198,149(92.1%)	180,546(91.1%)	17,603(8.9%)	
Malignant tumor				<0.001
No	212,578(98.9%)	194,131(91.3%)	18,447(8.7%)	
Yes	2,463(1.1%)	2,188(88.8%)	275(11.2%)	
Cataract/glaucoma				<0.001
No	180,626(84.0%)	166,343(92.1%)	14,283(7.9%)	
Yes	34,415(16.0%)	29,976(87.1%)	4,439(12.9%)	
Hypertension				<0.001
No	135,768(63.1%)	128,653(94.8%)	7,115(5.2%)	
Yes	79,273(36.9%)	67,666(85.4%)	11,607(14.6%)	
Cardiac-cerebral vascular disease				0.945
No	159,128(74.0%)	145,270(91.3%)	13,858(8.7%)	
Yes	55,913(26.0%)	51,049(91.3%)	4,864(8.7%)	
Osteoarthrosis				0.515
No	121,140(56.3%)	110,551(91.3%)	10,589(8.7%)	
Yes	93,901(43.7%)	85,768(91.3%)	8,133(8.7%)	
COPD				
No	193,092(89.8%)	176,336(91.3%)	16,756(8.7%)	0.164
Yes	21,949(10.2%)	19,983(91.0%)	1,966(9.0%)	

with and without osteoarthrosis, prevalence of diabetes varied significantly (7.1 vs. 7.8%, $P < 0.001$), but not in females (9.8 vs. 9.8%, $P = 0.971$).

"Urban and rural" stratification was used to analyze the prevalence and differences between parameters and diabetes (Table 4). Within the 111,940 urban participants, 12,730

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TABLE 3 Univariate and multivariate logistic analysis for diabetes.

Factor	Univariate		Multivariate	
	OR(95%CI)	*P*-value	OR(95%CI)	*P*-value
Household registration		<0.001	0.62(0.60–0.65)	<0.001
Urban area	1		1	
Rural area	0.48(0.47–0.50)	<0.001		
Age(years)		<0.001	0.97(0.96–0.98)	<0.001
60-64	1		1	
65-69	1.22 (1.17–1.27)	<0.001		
70-74	1.28(1.22–1.33)	<0.001		
75-79	1.23(1.17–1.29)	<0.001		
80-84	1.17(1.10–1.24)	<0.001		
≥85	0.89(0.82–0.96)	0.004		
Gender		<0.001	0.78(0.75–0.80)	<0.001
Female	1		1	
Male	0.76(0.73–0.78)	<0.001		
Education level		<0.001	1.14 (1.12–1.16)	<0.001
Uneducated	1		1	
Primary education	1.05 (1.01–1.09)	0.017		
Junior high school	1.47(1.41–1.54)	<0.001		
High school	1.74(1.64–1.84)	<0.001		
Junior College	2.29(2.10-2.50)	<0.001		
Bachelor degree or above	2.42(2.16-2.72)	<0.001		
Smoking		<0.001	0.93(0.88–0.99)	0.025
No	1		1	
Yes	0.87(0.82–0.92)	<0.001		
Alcohol consumption		<0.001	0.90(0.83–0.98)	0.014
Never or occasionally	1		1	
1–2 times a week	0.72(0.55–0.95)	0.02		
At least 3 times a week	0.65(0.54–0.78)	<0.001		
Often drunk	0.38(0.20–0.71)	0.003		
Sleep quality		<0.001	1.06(1.02–1.11)	0.010
Very good	1		1	
Relatively good	0.99(0.85–1.16)	0.906		
Average	1.08(0.95–1.23)	0.244		
Relatively poor	1.22(1.03–1.44)	0.019		
Very poor	1.79(1.41-2.28)	<0.001		
Exercise(per week)		<0.001	1.07 (1.06–1.09)	<0.001
Never exercise	1		1	
Less than once	1.29(1.20–1.39)	<0.001		
Once or twice	1.29(1.23–1.36)	<0.001		
Three to five times	1.49(1.43–1.57)	<0.001		
Six times and above	1.82 (1.75–1.88)	<0.001		
Medical insurance			0.69 (0.57–0.85)	<0.001
No	1		1	
Yes	0.61(0.50–0.74)	<0.001		
Gainful employment			0.69(0.65–0.74)	<0.001
No	1		1	
Yes	0.56(0.53–0.60)	<0.001		

(Continued)

TABLE 3 (Continued)

Factor	Univariate		Multivariate	
	OR(95%CI)	*P*-value	OR(95%CI)	*P*-value
Poverty			0.77(0.73–0.81)	<0.001
No	1		1	
Yes	0.62(0.59–0.65)	<0.001		
Economic status			1.10 (1.07–1.12)	<0.001
Very generous	1		1	
Relatively ample	0.90(0.79–1.03)	0.111		
Basically enough	0.85(0.75–0.96)	0.010		
Tougher	0.80 (0.70–0.91)	0.001		
Very difficult	0.94(0.82–1.09)	0.422		
Public benefit activities			0.85 (0.82–0.88)	<0.001
No	1		1	
Yes	0.83(0.81–0.86)	<0.001		
Spiritual cultural life			1.10 (1.03–1.8)	0.005
No	1		1	
Yes	1.37 (1.29–1.46)	<0.001		
Malignant tumor			1.16(1.02–1.32)	0.025
No	1		1	
Yes	1.32(1.17–1.50)	<0.001		
Cataract/glaucoma			1.52(1.46–1.58)	<0.001
No	1		1	
Yes	1.73(1.67–1.79)	<0.001		
Hypertension			2.86(2.77-2.95)	<0.001
No	1		1	
Yes	3.10(3.01-3.20)	<0.001		
Cardiac-cerebral vascular disease				
No	1			
Yes	1.00(0.97–1.03)	0.945		
Osteoarthrosis				
No	1			
Yes	1.00(0.96–1.02)	0.515		
COPD				
No	1			
Yes	1.04(0.99–1.09)	0.164		

individuals had diabetes. The prevalence of diabetes in urban areas varied between females and males (12 vs. 10.7%, $P < 0.001$) and between age groups ($P < 0.001$). Out of 103,101 participants living in urban areas, 5,992 were diagnosed with diabetes. The prevalence of diabetes in rural areas varied between females and males (7.2 vs. 4.3%, $P < 0.001$).

An age-specific analysis of the prevalence and differences between parameters and diabetes was further conducted (Table 4). We divided the continuous variable "age" into three groups, spaced at 10 years. A total of 10,218 individuals with diabetes were identified among 121,636 participants aged between 60 and 69 years. The prevalence of diabetes in this age group varied between urban and rural areas (10.7 vs. 6.0%, $P < 0.001$) and between females and males (9.6 vs. 7.1%, $P < 0.001$). The resulting bar chart is shown in Figure 5.

Discussion

This national cross-sectional study of older adults aged more than 60 years describes the national distribution of self-reported diabetes by demographic data, living habits, socioeconomic factors and comorbidities. A stratified sampling method was used to investigate the elderly people of all provinces in China. The questionnaire has high reliability and validity, and the

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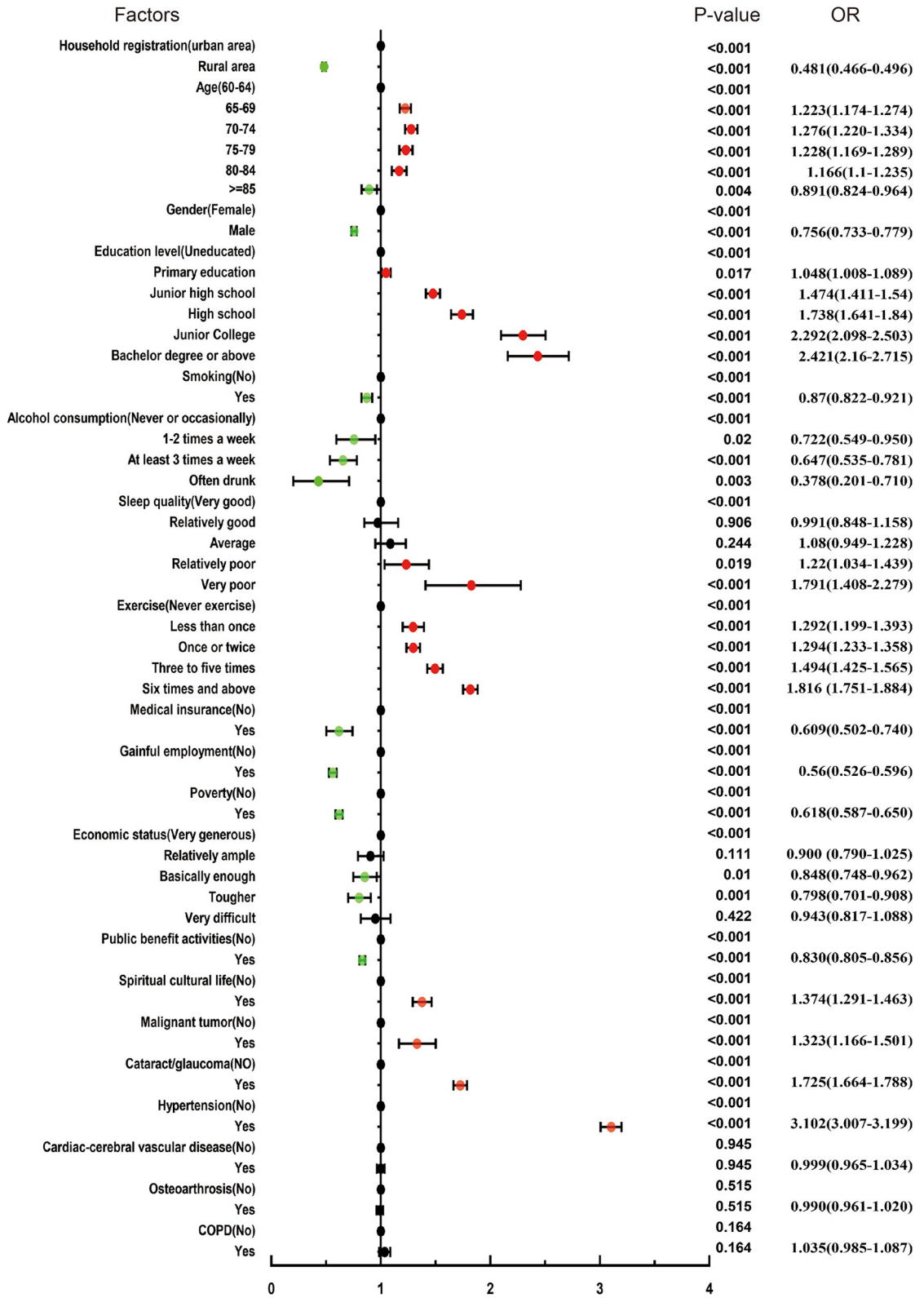

FIGURE 3
Association between related factors and diabetes by univariable logistic regression analysis.

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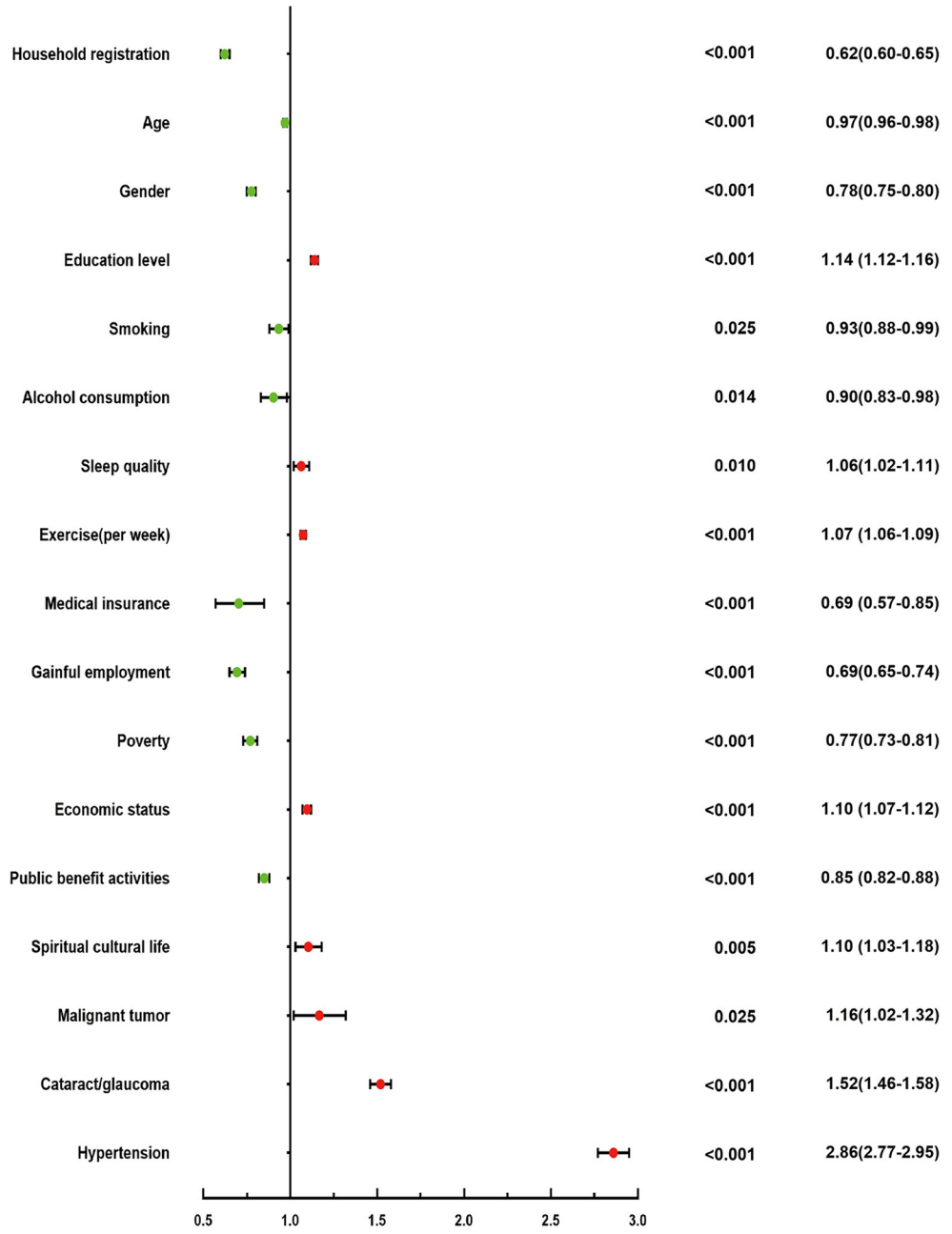

FIGURE 4
Association between related factors and diabetes by multivariate logistic regression analysis.

amount of data is large and reliable. Meanwhile, socioeconomic factors, living habits and comorbidities were included in the analysis as potential risk factors. This further expands our understanding of the causes on diabetes. Further stratified analyses were conducted to illustrate the differences of diabetes and other risk factors by gender, age, urban and rural areas. It can help medical workers to provide targeted prevention and treatment measures.

In this study, the prevalence of self-reported diabetes in China among people aged more than 60 years is 8.7%. Weiqing Wang et al. analyzed data from the China Cardiometabolic Disease and Cancer Cohort Study, which included 93,781

TABLE 4 The prevalence of diabetes by gender/urban and rural/age stratification.

Factors	Gender		Age			Urban and Rural area	
	Female	Male	60–69	70–79	≥80	Urban area	Rural area
Gender							
Female			5,947(9.6%)	3,626(10.8%)	1,391(8.2%)	7,160(12%)	3,804(7.2%)
Male			4,271(7.1%)	2,463(8.1%)	1,024(8.2%)	5,570(10.7%)	2,188(4.3%)
P-value			<0.001	<0.001	0.943	<0.001	<0.001
Age(years)							
60-64	3,187(8.8%)	2,307(6.6%)				3,502(9.7%)	1,992(5.7%)
65-69	2,760(10.7%)	1,964(7.9%)				3,148(12.2%)	1,576(6.3%)
70-74	2,016(10.9%)	1,444(8.4%)				2,392(12.9%)	1,068(6.2%)
75-79	1,610(10.8%)	1,019(7.7%)				1,878(12.6%)	751(5.7%)
80-84	965(9.4%)	678(8.4%)				1,237(11.9%)	406(5.0%)
≥85	426(6.4%)	346(7.9%)				573(9.4%)	199(4.0%)
P-value	<0.001	<0.001				<0.001	<0.001
Household registration							
Urban area	7,160(12.0%)	5,570(10.7%)	6,650(10.7%)	4,270(12.8%)	1,810(11.0%)		
Rural area	3,804(7.2%)	2,188(4.3%)	3,568(6.0%)	1,819(6.0%)	605(4.6%)		
P-value	<0.001	<0.001	<0.001	<0.001	<0.001		
Education level							
Uneducated	3,987(8.3%)	676(4.6%)	1,971(7.6%)	1,655(7.8%)	1,037(6.5%)	2,350(9.6%)	2,313(6%)
Primary education	4,120(9.8%)	2,753(5.8%)	3,992(7.3%)	2,183(8.5%)	698(7.9%)	4,185(10.2%)	2,688(5.6%)
Junior high school	1,828(12.7%)	2,435(9.3%)	2,732(9.6%)	1,208(12.2%)	323(14.1%)	3,450(12.9%)	813(5.9%)
High school	749(13.5%)	1,088(11.4%)	999(10.8%)	651(14.6%)	187(13.9%)	1,686(13.3%)	151(6.3%)
Junior College	174(14.1%)	486(18%)	360(14.5%)	207(16.3%)	93(18.1%)	651(15.6%)	9(10.7%)
Bachelor degree or above	81(12.9%)	295(17.4%)	137(16.1%)	168(16.5%)	71(15.8%)	375(16.2%)	1(12.5%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.036
Marital status							
Married	7,200(10.0%)	6,640(7.9%)	8,717(8.5%)	4,089(9.8%)	1,034(9.2%)	9,485(11.5%)	4,355(5.9%)
Widowed	3,675(9.3%)	928(6.3%)	1,295(8.4%)	1,936(9.3%)	1,372(7.7%)	3,067(11.3%)	1,536(5.7%)
Divorce	83(11.8%)	82(7.5%)	128(9.2%)	34(10.7%)	3(3.4%)	135(10.4%)	30(6%)
Never married	6(3.4%)	108(3.7%)	78(4%)	30(3.1%)	6(2.8%)	43(4.5%)	71(3.2%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Live alone							
No	9,297(9.8%)	7,066(7.8%)	9,394(8.5%)	5,051(9.6%)	1,918(8.6%)	11,241(11.5%)	5,122(5.8%)
Yes	1,667(9.6%)	692(6%)	824(7.8%)	1,038(9.2%)	497(7%)	1,489(10.8%)	870(5.8%)
P-value	0.482	<0.001	0.019	0.213	<0.001	0.021	0.741
Smoking							
No	1,056(9.9%)	367(9.5%)	762(9.5%)	457(10.9%)	204(8.8%)	978(12.4%)	445(6.7%)
Yes	9,908(9.7%)	7,391(7.5%)	9,456(8.3%)	5,632(9.4%)	2,211(8.1%)	11,752(11.3%)	5,547(5.8%)
P-value	0.649	<0.001	<0.001	0.001	0.239	0.002	0.002
Alcohol consumption							
Never or occasionally	10,946(9.8%)	7,596(7.6%)	10,104(8.4%)	6,039(9.6%)	2,399(8.2%)	12,615(11.4%)	5,927(5.8%)
1–2 times a week	10(8.1%)	45(6.2%)	40(6.9%)	12(5.8%)	3(4.6%)	36(8.9%)	19(4.3%)
At least 3 times a week	8(4.3%)	107(6%)	67(5.3%)	36(7%)	12(6.2%)	73(7.9%)	42(4%)
Often drunk	0(0%)	10(3.8%)	7(3.6%)	2(3.2%)	1(3.1%)	6(4.9%)	4(2.4%)
P-value	0.03	0.003	<0.001	0.016	0.355	<0.001	0.009

(Continued)

TABLE 4 (Continued)

Factors	Gender		Age			Urban and Rural area	
	Female	Male	60–69	70–79	≥80	Urban area	Rural area
Sleep quality							
Very good	108(8.7%)	147(7.7%)	141(7.6%)	80(9.1%)	34(8.1%)	179(9.8%)	76(5.8%)
Relatively good	276(9.2%)	249(7.0%)	300(7.8%)	157(8.9%)	68(7.3%)	396(11%)	129(4.4%)
Average	10,230(9.7%)	7,217(7.6%)	9,516(8.4%)	5,682(9.5%)	2,249(8.2%)	11,851(11.4%)	5,596(5.8%)
Relatively poor	270(10.6%)	118(8.1%)	205(9.8%)	134(10.7%)	49(7.7%)	245(13.6%)	143(6.5%)
Very poor	80(14.5%)	27(11.5%)	56(13.5%)	36(14.5%)	15(12.5%)	59(15.3%)	48(12%)
P-value	0.001	0.111	<0.001	0.038	0.392	0.001	<0.001
Exercise							
Never exercise	4,774(8.3%)	2,520(5.3%)	3,641(6.5%)	2,370(7.6%)	1,283(7.3%)	3,760(9.6%)	3,534(5.4%)
Less than once	533(10.3%)	297(7.0%)	397(7.9%)	293(10.3%)	140(8.8%)	615(11.7%)	215(5.1%)
Once or twice	1,437(9.9%)	988(7.6%)	1,362(8.3%)	788(9.9%)	275(8.6%)	1,711(10.9%)	714(6%)
3–5five times	1,490(11.1%)	1,147(8.9%)	1,597(9.9%)	801(10.5%)	239(9.5%)	1,985(11.7%)	652(6.9%)
Six times and above	2,730(12.6%)	2,806(11.3%)	3,221(11.6%)	1,837(13%)	478(10.5%)	4,659(13.4%)	877(7.6%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Medical insurance							
No	10,886(9.8%)	7,728(7.6%)	10,155(8.4%)	6,057(9.6%)	2,402(8.2%)	12,654(11.4%)	5,960(5.8%)
Yes	78(7.2%)	30(3.4%)	63(5.9%)	32(6.1%)	13(3.6%)	76(7.6%)	32(3.3%)
P-value	0.004	<0.001	0.003	0.008	0.002	<0.001	0.001
Gainful employment							
No	10,556(10.0%)	7,026(8%)	9,250(9%)	5,936(9.7%)	2,396(8.2%)	11,998(11.9%)	5,584(6%)
Yes	408(6.4%)	732(4.8%)	968(5.2%)	153(5.5%)	19(7.1%)	732(6.8%)	408(3.8%)
P-value	<0.001	<0.001	<0.001	<0.001	0.512	<0.001	<0.001
Poverty							
No	9,801(10.2%)	7,212(8.0%)	9,400(8.6%)	5,423(10.3%)	2,190(9.1%)	12,186(11.7%)	4,827(5.9%)
Yes	1,163(7.2%)	546(4.2%)	818(6.5%)	666(6.0%)	225(4.2%)	544(6.8%)	1,165(5.5%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.039
Economic status							
Very generous	132(10.4%)	144(9.8%)	147(9.5%)	84(10.6%)	45(11%)	229(12.1%)	47(5.6%)
Relatively ample	1,467(9.5%)	1,439(8.9%)	1,516(8.1%)	957(11%)	433(10.3%)	2,313(11.2%)	593(5.3%)
Basically enough	6,422(9.7%)	4,571(7.6%)	6,054(8.3%)	3,554(9.6%)	1,385(8.2%)	7,899(11.5%)	3,094(5.4%)
Tougher	2,416(9.9%)	1,290(6.2%)	2,040(8.5%)	1,212(8.3%)	454(6.9%)	1,883(10.9%)	1,823(6.5%)
Very difficult	527(11.2%)	314(7.7%)	461(10.4%)	282(9.6%)	98(6.8%)	406(12.5%)	435(7.9%)
P-value	0.006	<0.001	<0.001	<0.001	<0.001	0.056	<0.001
Public benefit activities							
No	6,643(10.3%)	4,348(8.3%)	5,333(9%)	3,786(10.4%)	1,872(8.7%)	7,709(12.1%)	3,282(6.1%)
Yes	4,321(9%)	3,410(6.8%)	4,885(7.8%)	2,303(8.4%)	543(6.9%)	5,021(10.4%)	2,710(5.5%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Spiritual cultural life							
No	773(7.1%)	346(5.7%)	392(6.7%)	408(7.3%)	319(5.9%)	543(9.3%)	576(5.2%)
Yes	10,191(10.0%)	7,412(7.7%)	9,826(8.5%)	5,681(9.7%)	2,096(8.7%)	12,187(11.5%)	5,416(5.9%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.004
Malignant tumor							
No	10,804(9.7%)	7,643(7.5%)	10,080(8.4%)	5,997(9.5%)	2,370(8.1%)	12,507(11.3%)	5,940(5.8%)
Yes	160(12.8%)	115(9.5%)	138(9.7%)	92(12.1%)	45(16.1%)	223(14.3%)	52(5.7%)
P-value	<0.001	0.011	0.08	0.014	<0.001	<0.001	0.913

(Continued)

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TABLE 4 (Continued)

Factors	Gender		Age			Urban and Rural area	
	Female	Male	60–69	70–79	≥80	Urban area	Rural area
Cataract/glaucoma							
No	8,066(8.9%)	6,217(6.9%)	8,409(7.8%)	4,346(8.5%)	1,528(7.1%)	9,492(10.3%)	4,791(5.4%)
Yes	2,898(13.5%)	1,541(11.9%)	1,809(13.3%)	1,743(13.6%)	887(11.2%)	3,238(16.4%)	1,201(8.2%)
P-value	<0.001	<0.001	0.03	<0.001	<0.001	<0.001	<0.001
Hypertension							
No	3,929(5.8%)	3,186(4.7%)	4,278(5.3%)	2,132(5.7%)	705(4%)	4,779(7.1%)	2,336(3.4%)
Yes	7,035(15.8%)	4,572(13.1%)	5,940(14.6%)	3,957(14.9%)	1,710(14.2%)	7,951(17.8%)	3,656(10.6%)
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Cardiac-cerebral vascular disease							
No	7,944(9.8%)	5,914(7.6%)	7,519(8.3%)	4,501(9.5%)	1,838(8.5%)	9,539(11.3%)	4,319(5.8%)
Yes	3,020(9.7%)	1,844(7.4%)	2,699(8.6%)	1,588(9.6%)	577(7.4%)	3,191(11.4%)	1,673(6%)
P-value	0.943	0.275	0.191	0.875	0.002	0.640	0.196
Osteoarthrosis							
No	5,567(9.8%)	5,022(7.8%)	5,929(8.4%)	3,376(9.7%)	1,284(7.9%)	7,602(11.3%)	2,987(5.6%)
Yes	5,397(9.8%)	2,736(7.1%)	4,289(8.3%)	2,713(9.3%)	1,131(8.5%)	5,128(11.5%)	3,005(6.1%)
P-value	0.971	<0.001	0.504	0.073	0.069	0.203	<0.001
COPD							
No	9,900(9.7%)	6,856(7.6%)	9,297(8.4%)	5,390(9.6%)	2,069(8.1%)	11,522(11.3%)	5,234(5.7%)
Yes	1,064(10.8%)	902(7.5%)	921(8.9%)	699(9.1%)	346(9%)	1208(12.1%)	758(6.4%)
P-value	<0.001	0.728	0.068	0.136	0.057	0.024	0.007

subjects with a mean age of 55.7 years, of whom 67% were women. During a mean follow-up period of about 3 years, 6,171 new cases of diabetes were identified. And the incidence of diabetes was 6.58% based on blood glucose testing (20). In a study of the China Kadoorie Biobank, 8,784 out of 461,211 (prevalence: 19.0%) adults aged 30–79 years were diagnosed with type 2 diabetes during a median follow-up of about 7 years (23). Wang et al. proposed that the self-reported prevalence of diabetes is 8.4% in middle-aged and elderly Chinese (24). In another study of about 10, 000 participants conducted in 2011–2012, the prevalence of self-reported diabetes and screening-detected prevalence was 6.0 and 9.8% among people over 44 years of age (25). Our study was a nationwide cross-sectional study in 2015. Self-reported prevalence of diabetes was slightly higher than the two studies above, probably because of increased aging, physical examination, and adequate nutrition of the elderly. Of those surveyed, 58.7% to 69.9% were unaware of their diagnosis (8, 9, 26). According to the available data, undiagnosed diabetes still accounts for a large proportion of cases, and prediabetes may represent an even larger proportion (27).

Women are more likely than men to suffer from diabetes according to our study. Gender differences in diabetes have varied in previously reported studies (28). However, research has shown that some important risk factors, such as obesity, sex hormones, and psychological stress problems, are more common in women, supporting our finding that older women develop diabetes more frequently than men (29). Our study showed that age is an independent risk factor for diabetes. However, the prevalence of diabetes does not completely increase with age. The prevalence of diabetes is highest in people aged 70–74 years and lowest in people aged more than 85 years. A meta-analysis revealed a prevalence of 11.0% (95% CI 9.0–13.0%) among 55–64-year-olds, 14.1% (95% CI 12.3–16%) among 65–74-year-olds, and 11.0% (95% CI 9.0–13.0%) over 75-year-olds (30). This is similar to the results of our study. These results were obtained because self-reported diagnosis of diabetes in the old may be biased due to cognitive decline and shortened life expectancy in the elderly with diabetic macroangiopathy.

According to our study, diabetes prevalence is higher in economically developed provinces than in less developed ones. Meanwhile, compared to rural areas, urban areas had a higher prevalence of diabetes, according to our survey. As in some earlier geographical studies, the prevalence of diabetes in economically developed provinces and northern provinces was higher than that in economically underdeveloped and central or southern regions (30, 31). A study of 512,869 participants in China indicated that 4.1% of diabetes patients live in rural areas, compared with 8.1% in urban areas (18). It is well known that

diabetes is highly related to nutritional status and obesity, and diet structure and lifestyle can affect the incidence of diabetes. At the same time, the medical conditions in the developed areas are better, and people pay more attention to health, so the early detection of diabetes is more likely. The prevalence of diabetes in participants with higher level of education was higher, which is similar to many previously published studies (31–33).

In our study, older adults with diabetes smoked and drank less but exercised often. Smoking and drinking are recognized as unhealthy lifestyles that can increase the risk of diabetes, while exercise is recognized as a healthy lifestyle, so this finding could be interpreted as reflecting good health education and lifestyle interventions of people with diabetes in China. In our study, Poor sleep quality is an independent risk factor for diabetes. And the prevalence of diabetes was significantly higher among relatively poor and very poor sleepers, which could be related to chronic stress stimulation and increased body mass index., The study by Wang et al. (34) shows that obstructive sleep apnea has been linked to abnormal glucose metabolism in laboratory-based experiments. Sleep apnea is highly correlated with poor sleep quality, which may partly explain the relationship between sleep and diabetes. Diabetes patients' sleep duration is also associated with glycemic control.

Interestingly, the never-married group had significantly lower rates of diabetes than the married group. The specific cause of this is unclear and needs further study. We found that 99.1% of Chinese citizens had medical insurance, and the prevalence of diabetes among this group was also higher, which may be related to increased rates of outpatient visits and subsequent diagnoses. Our study also suggests that diabetic patients are more actively involved in spiritual and cultural life than non-diabetic patients. This may benefit from the widespread awareness of lifestyle intervention for diabetes.

People with hypertension were significantly more likely to develop diabetes than those with normal blood pressure. Diabetes and hypertension share numerous pathophysiological mechanisms and genetic factors. Consequently, both clinical entities contribute synergistically to micro- and macro-vasculopathy and cardiovascular death (35). In a study of 318,664 individuals, it was found that T2DM is associated with hypertension, but that the causal relationship is unlikely (36). The prevalence of diabetes in tumor patients and cataract/glaucoma patients was also higher than that in patients without these comorbidities. Diabetes patients may be at a higher risk of cancer due to risk factors such as age, obesity, inactivity, and smoking. Several types of cancer are also affected by diabetes, including hepatocellular cancer, hepatobiliary cancer, pancreatic cancer, ovarian cancer, breast cancer, endometrial cancer, and gastrointestinal cancer. Hyperglycemia, increased bioactivity of insulin-like growth factor 1, hyperinsulinemia, dysregulation of sex hormones, oxidative stress, and chronic inflammation are some of the biological mechanisms linking diabetes and cancer (37). It is recognized that T2DM is a risk factor for cataract development (38). Glaucoma and diabetes share some risk factors and pathophysiologic features, but their pathophysiology is not completely understood. The presence of diabetes and elevated fasting glucose levels is also related to elevated intraocular pressure, which is one of the key risk factors for glaucomatous optic neuropathy (39). Diabetes prevalence was not statistically significant in the elderly with or without COPD, but not among women. A neglected relationship is that of the diabetes–lung association, which is epidemiologically and clinically well-established, including asthma and COPD; however, the underlying mechanism and pathophysiology are not fully understood (40). In our study, there were no statistically significant differences of prevalence of diabetes in the elderly with or without osteoarthritis, but it was not seen among men. The reason for this gender difference is unclear. A meta-analysis of 49 studies found a significant association between osteoarthritis and type 2 diabetes (41). There are two major pathways involved in the pathogenesis of T2DM leading to osteoarthritis: oxidative stress and low-grade chronic inflammation caused by chronic hyperglycemia and insulin resistance (42).

Limitations

The limitations of this study may include the following aspects. Firstly, this is a cross-sectional study. And potential risk factor analysis is correlation analysis, not causality analysis. Secondly, we do not test blood sugar to distinguish the hidden diabetes and pre-diabetes, nor do we gather information of the control and treatment of diabetes among the elderly. There are biases from the older persons due to recall bias or cognitive impairment. Thirdly, some variables were not evaluated. The dimensions and number of variables were large, and there was a lack of assessment on presence and change of these modifiable lifestyles, before or after the diagnosis of diabetes. Nonetheless, to our knowledge, the present study demonstrated the awareness of diabetes diagnosis, lifestyle and economic status of the Chinese elderly. The study also provided insights into socioeconomic, lifestyle, comorbidities and other potential risk factors for diabetes. These results could be helpful for further research and comprehensive understanding of diabetes.

Conclusions

With the advent of the aging society in China, the prevalence of diabetes as a disease of aging is increasing. Besides genetic and metabolic factors, socioeconomic factors, living habits, and comorbidities are also potential independent risk factors for diabetes. This study makes us realize that diabetes has a complex pathogenesis involving both environmental and individual factors. Further studies can be conducted based on

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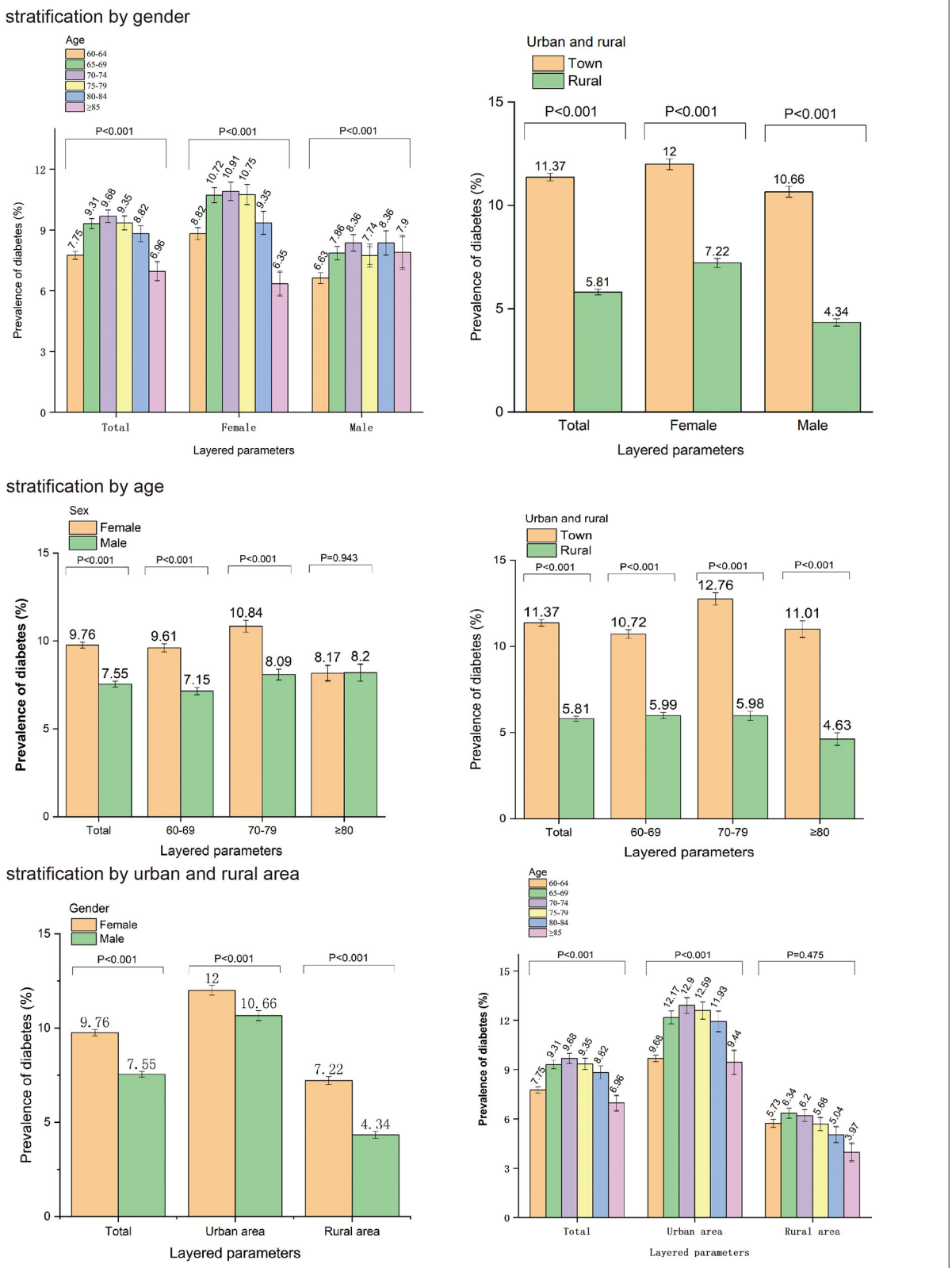

FIGURE 5
The relationship between parameters and diabetes was further analyzed by gender, age, and urban and rural residence stratification.

the results drawn from this study. Diabetes, as one of the diseases with increasing prevalence, and its serious complications have a great impact on the physical and mental health of patients, which needs more attention and financial investment from the government. This study also provides more relevant references for medical administrative departments on diabetes.

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 research protocol has been approved by the Ethical Review Committee of Beijing Hospital (No. 2021BJYYEC-294-01) and approved by National Bureau of Statistics (No. [2014] 87). All participants have obtained written informed consent. The patients/participants provided their written informed consent to participate in this study.

Author contributions

DL and JuL: conceived and designed the study. XH and LM: supervised the study. XH, HX, JiL, YL, and NJ: performed statistical analysis. XH, ZW, HL, XQ, XZ, and QZ: drafted the report. All authors revised the report and approved the final version before submission. All authors contributed to data collection, analysis, and interpretation.

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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/fpubh.2022.1051445/full#supplementary-material

SUPPLEMENT 1
Sampling plan of the fourth Sample Survey of the Aged Population in Urban and Rural China (SSAPUR).

SUPPLEMENT 2
Personal questionnaire of the fourth SSAPUR.

References

1. Fang EF, Xie C, Schenkel JA, Wu C, Long Q, Cui H, et al. A research agenda for ageing in China in the 21st century (2nd edition): Focusing on basic and translational research, long-term care, policy and social networks. *Ageing Res Rev.* (2020) 64:101174. doi: 10.1016/j.arr.2020.101174

2. Luo Y, Su B, Zheng X. Trends and challenges for population and health during population aging—China, 2015-2050. *China CDC Wkly.* (2021) 3:593–8. doi: 10.46234/ccdcw2021.158

3. Sinclair AJ, Abdelhafiz AH, Forbes A, Munshi M. Evidence-based diabetes care for older people with Type 2 diabetes: a critical review. *Diabet Med.* (2019) 36:399–413. doi: 10.1111/dme.13859

4. Sinclair A, Saeedi P, Kaundal A, Karuranga S, Malanda B, Williams R. Diabetes and global ageing among 65-99-year-old adults: Findings from the International Diabetes Federation Diabetes Atlas, 9(th) edition. *Diabetes Res Clin Pract.* (2020) 162:108078. doi: 10.1016/j.diabres.2020.108078

5. Bommer C, Heesemann E, Sagalova V, Manne-Goehler J, Atun R, Bärnighausen T, et al. The global economic burden of diabetes in adults aged 20-79 years: a cost-of-illness study. *Lancet Diabetes Endocrinol.* (2017) 5:423–30. doi: 10.1016/S2213-8587(17)30097-9

6. Ma R. Epidemiology of diabetes and diabetic complications in China. *Diabetologia.* (2018) 61:1249–60. doi: 10.1007/s00125-018-4557-7

7. Pan XR, Yang WY Li GW, Liu J. Prevalence of diabetes and its risk factors in China, 1994. National diabetes prevention and control cooperative group. *Diabet Care.* (1997) 20:1664–9. doi: 10.2337/diacare.20.11.1664

8. Yang SH, Dou KF, Song WJ. Prevalence of diabetes among men and women in China. *N Engl J Med.* (2010) 362:2425–6. doi: 10.1056/NEJMoa0908292

9. Xu Y, Wang L, He J, Bi Y, Li M, Wang T, et al. Prevalence and control of diabetes in Chinese adults. *JAMA.* (2013) 310:948–59. doi: 10.1001/jama.2013.168118

10. Bommer C, Sagalova V, Heesemann E, Manne-Goehler J, Atun R, Bärnighausen T, et al. Global economic burden of diabetes in adults: projections from 2015 to 2030. *Diabetes Care.* (2018) 41:963–70. doi: 10.2337/dc17-1962

11. International Diabetes Federation. *China diabetes report 2000–2045, 10th edn.* (2021). Available online at: https://diabetesatlas.org/data/en/country/42/cn.html (accessed December 6, 2021).

12. Engel GL. The need for a new medical model: a challenge for biomedicine. *Science.* (1977) 196:129–36. doi: 10.1126/science.847460

13. Liu C, Li X, Lin M, Zheng L, Chen X. A cross-sectional study on diabetes epidemiology among people aged 40 years and above in Shenyang, China. *Sci Rep.* (2020) 10:17742. doi: 10.1038/s41598-020-74889-x

14. Yan X, Xia H, Li H, Deng X, Yang L, Zhao S, et al. Diabetes in Shenzhen, China: epidemiological investigation and health care challenges. *J Glob Health.* (2017) 7:011102. doi: 10.7189/jogh.07.011102

15. Wang R, Zhang P, Li Z, Lv X, Cai H, Gao C, et al. The prevalence of pre-diabetes and diabetes and their associated factors in Northeast China: a cross-sectional study. *Sci Rep.* (2019) 9:2513. doi: 10.1038/s41598-019-39221-2

16. Huo L, Ji L, Deng W, Shaw JE, Zhang P, Zhao F, et al. Age distribution and metabolic disorders in people with Type 1 diabetes in Beijing and Shantou, China: a cross-sectional study. *Diabet Med.* (2018) 35:721–8. doi: 10.1111/dme.13616

17. Ruan Y, Yan QH, Xu JY, Yang QD, Yao HH Li R, et al. Epidemiology of diabetes in adults aged 35 and older from Shanghai, China. *Biomed Environ Sci.* (2016) 29:408–16. doi: 10.3967/bes2016.053

18. Bragg F, Holmes MV, Iona A, Guo Y, Du H, Chen Y, et al. Association between diabetes and cause-specific mortality in rural and urban areas of China. *JAMA.* (2017) 317:280–9. doi: 10.1001/jama.2016.19720

19. Wang H, Yao J, Yin X, Guo X, Yin J, Qu H, et al. Organisational and individual characteristics associated with glycaemic control among patients with type 2 diabetes: cross-sectional study in China. *BMJ Open.* (2020) 10:e036331. doi: 10.1136/bmjopen-2019-036331

20. Tiange W, Zhiyun Z, Guixia W, Qiang L, Yu X, Mian L, et al. Age-related disparities in diabetes risk attributable to modifiable risk factor profiles in Chinese adults: a nationwide, population-based, cohort study. *Lancet Healthy Longevity.* (2021) 2:e618–28. doi: 10.1016/S2666-7568(21)00177-X

21. Zhang Y, Pan XF, Chen J, Xia L, Cao A, Zhang Y, et al. Combined lifestyle factors and risk of incident type 2 diabetes and prognosis among individuals with type 2 diabetes: a systematic review and meta-analysis of prospective cohort studies. *Diabetologia.* (2020) 63:21–33. doi: 10.1007/s00125-019-04985-9

22. Stringhini S, Tabak AG, Akbaraly TN, Sabia S, Shipley MJ, Marmot MG, et al. Contribution of modifiable risk factors to social inequalities in type 2 diabetes: prospective Whitehall II cohort study. *BMJ.* (2012) 345:e5452. doi: 10.1136/bmj.e5452

23. Lv J, Yu C, Guo Y, Bian Z, Yang L, Chen Y, et al. Adherence to a healthy lifestyle and the risk of type 2 diabetes in Chinese adults. *Int J Epidemiol.* (2017) 46:1410–20. doi: 10.1093/ije/dyx074

24. Wang Z, Li X, Chen M. Socioeconomic factors and inequality in the prevalence and treatment of diabetes among middle-aged and elderly adults in China. *J Diabetes Res.* (2018) 2018:1471808. doi: 10.1155/2018/1471808

25. Wu H, Jackson CA, Wild SH, Jian W, Dong J, Gasevic D. Socioeconomic status and self-reported, screen-detected and total diabetes prevalence in Chinese men and women in 2011-2012: a nationwide cross-sectional study. *J Glob Health.* (2018) 8:020501. doi: 10.7189/jogh.08.020501

26. Wang L, Gao P, Zhang M, Huang Z, Zhang D, Deng Q, et al. Prevalence and ethnic pattern of diabetes and prediabetes in China in 2013. *JAMA.* (2017) 317:2515–23. doi: 10.1001/jama.2017.7596

27. Li Y, Teng D, Shi X, Qin G, Qin Y, Quan H, et al. Prevalence of diabetes recorded in mainland China using 2018 diagnostic criteria from the American Diabetes Association: national cross sectional study. *BMJ.* (2020) 369:m997. doi: 10.1136/bmj.m997

28. Wu H, Bragg F, Yang L, Du H, Guo Y, Jackson CA. et al. Sex differences in the association between socioeconomic status and diabetes prevalence and incidence in China: cross-sectional and prospective studies of 05 million adults. *Diabetologia.* (2019) 62:1420–9. doi: 10.1007/s00125-019-4896-z

29. Kautzky-Willer A, Harreiter J, Pacini G. Sex and gender differences in risk, pathophysiology and complications of type 2 diabetes mellitus. *Endocr Rev.* (2016) 37:278–316. doi: 10.1210/er.2015-1137

30. Yang L, Shao J, Bian Y, Wu H, Shi L, Zeng L, et al. Prevalence of type 2 diabetes mellitus among inland residents in China (2000-2014): a meta-analysis. *J Diabetes Investig.* (2016) 7:845–52. doi: 10.1111/jdi.12514

31. Zhou M, Astell-Burt T, Bi Y, Feng X, Jiang Y, Li Y, et al. Geographical variation in diabetes prevalence and detection in china: multilevel spatial analysis of 98,058 adults. *Diabetes Care.* (2015) 38:72–81. doi: 10.2337/dc14-1100

32. Mendenhall E, Kohrt BA, Norris SA, Ndetei D, Prabhakaran D. Non-communicable disease syndemics: poverty, depression, and diabetes among low-income populations. *Lancet.* (2017) 389:951–63. doi: 10.1016/S0140-6736(17)30402-6

33. Cao G, Cui Z, Ma Q, Wang C, Xu Y, Sun H, et al. Changes in health inequalities for patients with diabetes among middle-aged and elderly in China from 2011 to 2015. *BMC Health Serv Res.* (2020) 20:719. doi: 10.1186/s12913-020-05609-4

34. Wang T, Lu J, Wang W, Mu Y, Zhao J, Liu C, et al. Sleep duration and snoring associate with hypertension and glycaemic control in patients with diabetes. *Diabet Med.* (2015) 32:1001–7. doi: 10.1111/dme.12809

35. Yildiz M, Esenboga K, Oktay AA. Hypertension and diabetes mellitus: highlights of a complex relationship. *Curr Opin Cardiol.* (2020) 35:397–404. doi: 10.1097/HCO.0000000000000748

36. Sun D, Zhou T, Heianza Y, Li X, Fan M, Fonseca VA, et al. Type 2 diabetes and hypertension. *Circ Res.* (2019) 124:930–7. doi: 10.1161/CIRCRESAHA.118.314487

37. Suh S, Kim KW. Diabetes and cancer: cancer should be screened in routine diabetes assessment. *Diabetes Metab J.* (2019) 43:733–43. doi: 10.4093/dmj.2019.0177

38. Song BJ, Aiello LP, Pasquale LR. Presence and risk factors for glaucoma in patients with diabetes. *Curr Diab Rep.* (2016) 16:124. doi: 10.1007/s11892-016-0815-6

39. Khateeb J, Fuchs E, Khamaisi M. Diabetes and lung disease: a neglected relationship. *Rev Diabet Stud.* (2019) 15:1–15. doi: 10.1900/RDS.2019.15.1

40. Louati K, Vidal C, Berenbaum F, Sellam J. Association between diabetes mellitus and osteoarthritis: systematic literature review and meta-analysis. *RMD Open.* (2015) 1:e000077. doi: 10.1136/rmdopen-2015-000077

41. Veronese N, Cooper C, Reginster JY, Hochberg M, Branco J, Bruyère O, et al. Type 2 diabetes mellitus and osteoarthritis. *Semin Arthritis Rheum.* (2019) 49:9–19. doi: 10.1016/j.semarthrit.2019.01.005

42. Drinkwater JJ, Davis WA, Davis T. A systematic review of risk factors for cataract in type 2 diabetes. *Diabetes Metab Res Rev.* (2019) 35:e3073. doi: 10.1002/dmrr.3073

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The effect of low volume high-intensity interval training on metabolic and cardiorespiratory outcomes in patients with type 2 diabetes mellitus: A systematic review and meta-analysis

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Aims: The present systematic review and meta-analysis of randomized controlled trials (RCTs) was conducted to investigate the effect of low volume high-intensity interval training (LVHIIT) on the metabolic and cardiorespiratory outcomes in patients with type 2 diabetes mellitus (T2DM).

Methods: Relevant articles were sourced from PubMed, EBSCO, Web of Science, Embase, and the Cochrane Library from inception to October 2022. The study search strategy and all other processes were implemented in accordance with the PRISMA statement.

Results: Five randomized controlled trials that satisfied the inclusion criteria were included in this meta-analysis. The LVHIIT group had significantly lower fasting blood glucose levels (RR= -1.21; 95% CI= -2.02— -0.40, p = 0.0032) and HbA1c levels (RR= -0.65; 95% CI= -1.06— -0.23, p = 0.002) and higher levels of insulin resistance indicator HOMA-IR (RR= -1.34; 95% CI = -2.59— -0.10, p = 0.03) than the control group. Moreover, our results show that LVHIIT can reduce body mass (RR = -0.94, 95% CI = -1.37— -0.51, p<0.0001) and body mass index (RR = -0.31, 95% CI = -0.47— -0.16, p<0.0001). LVHIIT had a better therapeutic effect on blood lipid metabolism, such as total cholesterol, high-density lipoprotein, low-density lipoprotein and triglycerides. However, the change in fasting insulin levels was not statistically significant (RR= -1.43; 95% CI = -3.46— 0.60, p =0.17). Furthermore, LVHIIT reduced the systolic blood pressure (RR =-4.01, 95% CI = -4.82 – -3.21, p<0.0001) and improved peak oxygen uptake (VO_{2peak}) compared to the control group (RR= 5.45; 95% CI = 1.38 – 9.52, p =0.009).

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Conclusion: After a certain period of LVHIIT, glycaemic control, insulin resistance, body weight, lipid profile and cardiorespiratory outcomes were significantly improved in T2DM patients.

KEYWORDS

low volume high-intensity interval training, meta-analysis, metabolism outcome, type 2 diabetes mellitus, cardiorespiratory outcomes

Introduction

Type 2 diabetes mellitus (T2DM) is a metabolic disease characterized by increased blood glucose concentrations. T2DM affects more than 400 million people worldwide, a figure that is expected to exceed 642 million people by 2040. In addition to severe suffering for the actual patient, the economic cost for disability and treatment places a heavy burden on society (1). Although many pharmacological treatments have emerged in recent years, these medications not only fail to prevent the progression of diabetes and its complications but also cause many side effects.

Cardiometabolic risk factors including central obesity, hypertension, dyslipidemia, and insulin resistance are strongly associated with the development and progression of T2DM (2). Lifestyle interventions appear as the efficient strategy to minimize cardiometabolic risk factors and improve T2D, which has gained increasing attention and acceptance among patients due to their simplicity and repeatability (3–5). Lifestyle interventions can be used as primary or supplementary treatments for T2DM patients according to the current ADA (American Diabetes Association) guidelines (6). Recent clinical trials have demonstrated that intensive lifestyle interventions can reduce the incidence of diabetes by 58% compared to those without lifestyle interventions (7). Among these lifestyle interventions, physical exercise results in improved insulin sensitivity and glucose homeostasis, which has long been recommended as one of the key therapeutic interventions for T2DM (8). High-intensity interval training (HIIT) consists of alternating repetitions of short periods of high-intensity exercise interspersed with less active or passive recovery periods. HIIT should be performed at 80–100% of the max heart rate interval, with a lower heart rate during the rest period. Compared to widely used moderate-intensity continuous training (MICT), HIIT has been proposed as a lower total energy expended exercise intervention that may bring about similar positive effects (9, 10). Collective evidence suggests that HIIT contributes to greater improvements in cardiorespiratory fitness compared to MICT (11), where cardiorespiratory fitness is inverse associated with the incidence of T2DM (12).

Nevertheless, normal HIIT has a much higher exercise intensity with a higher risk of injury in T2DM patients (13). Therefore, a milder HIIT exercise protocol is needed to reduce the risk of HIIT while improving metabolic and cardiorespiratory outcomes in individuals with T2DM. Additionally, lack of time is one of the common obstacles to physical activities. Low volume high-intensity interval training (LVHIIT) is a type of HIIT with the reduced total training volume (14). It has been suggested that LVHIIT could improve cardiorespiratory fitness as effective as high-volume HIIT, suggesting that LVHIIT may serve as a potent and time-efficient physical activity intervention strategy (15–17). The beneficial roles of LVHIIT on body composition have also been demonstrated (16, 18). However, the effects of LVHIIT on metabolic and cardiorespiratory outcomes in patients with T2DM remain unclear. In addition, new studies with more detailed data and high evidence levels have been published. Thus, we performed the current systematic review and meta-analysis of randomized controlled trials (RCTs) to investigate the effects of LVHIIT on T2DM patients. The results of this investigation may guide future decision-making regarding the use of lifestyle interventions among patients with T2DM.

Materials and methods

This systematic review and meta-analysis followed the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) statement and the Cochrane Handbook for Systematic Reviews of Interventions (19). Ethical approval and patient consent were not required because all analyses were based on previously published studies.

Literature search and selection criteria

LVHIT as intervention treatment vs no exercise or shame exercise (exercise at very low density). We systematically searched several databases including PubMed, EBSCO, Web of science, EMbase, and the Cochrane Library from inception to

October 2022. The structured search strategies used the combination of RCTs and LVHIIT with diabetes patients: ["Low volume" OR "high-intensity interval training" AND (exercise OR physical activity) AND ("randomized controlled trials") AND (diabetes) NOT (review) NOT (meta) NOT (animal experiment)]. The reference lists of retrieved studies and relevant reviews were hand-searched, and the process mentioned above was performed repeatedly to ensure the inclusion of all eligible studies. Inclusion criteria were as follows (1): randomized control trials, (2) T2DM diagnosis before participating in the experiment, (3)as there is no universal standard definition of HIIT, thus we used the HIIT standard proposed by previous meta-analysis (20, 21), (4) LVHIT as intervention treatment vs no exercise or shame exercise, (5) sufficient data for extraction, (6) full text only, and studies with all languages were included.

Data extraction and outcome measures

Baseline information that was extracted from the original studies included the following: first author, published year, number of patients, patient age and gender distributions, the evaluation of the evidence level, detailed intervention method and time of period. Data were independently extracted by two investigators. Discrepancies were resolved by consensus.

The primary outcomes were fasting blood glucose (FBG), HOMA-IR and HbA1c. Secondary outcomes were fasting insulin, body mass index (BMI), body mass, plasma lipid metabolism (TC, HDL, LDL and triglyceride) and the cardiorespiratory fitness parameters including systolic blood pressure (SBP), diastolic blood pressure (DBP) and relative $VO_{2\ peak}$.

Quality assessment of individual studies

The methodological quality of each RCT was assessed by the Jadad Scale which consists of three evaluation elements: randomization (0-2 points), blinding (0-2 points), dropouts and withdrawals (0-1 points) (22). One point was be allocated to each element if it had been conducted and mentioned appropriately in the original article. The total score of the Jadad Scale ranges from 0 to 5 points. An article with a total Jadad score that is less than or equal to 2 is considered to be of low quality. Concurrently, a study is thought to be of high quality if its total Jadad score greater or equal to 3 (23).

Statistical analysis

Risk Ratio (RR) with 95% confidence intervals (CIs) was calculated for dichotomous outcomes. Heterogeneity was evaluated using the I^2 statistic, with $I^2 > 50\%$ taken to indicate significant heterogeneity (24). Sensitivity analysis was performed to evaluate the influence of a single study on the overall estimate by omitting one study in turn or performing subgroup analysis. The random-effects model was used for meta-analysis. Owing to the limited number of included studies (<10), publication bias was not assessed. Statistical significance was accepted at $P < 0.05$. All the data are presented as mean ± SD. All statistical analyses were performed using Review Manager Software Version 5.3 (The Cochrane Collaboration, Software Update, Oxford, UK).

Results

Literature searches, study characteristics, and quality assessment

In total, 18085 articles including 64 in PubMed, 2141 in EBSCO, 15863 in Web of science, 5 in Embase and 12 in central Cochrane were initially identified from the databases. After removing duplicates, 5537 articles were retained. A total of 5326 studies were excluded from our study due to unrelated abstracts and titles. We also excluded 3 studies that were not RCTs, 2 studies that presented insufficient data, and 1 study that reported an improper methodology. Ultimately, five RCTs satisfied the inclusion criteria and were included in this meta-analysis (10, 25–28). The article selection process was performed in accordance with the PRISMA statement and the flow chart is shown as Figure 1. The baseline characteristics of the 5 included studies are shown in Table 1. Only Afousi et al. reported the age range of the patients (45-60 years old) (26). Four studies compared LVHIIT to no exercise, and one compared LVHIIT to a sham-exercise placebo. Four groups (10, 25–27) used cycling, and 1 group used jogging/running (28). There were no statistically significant differences in the patient baseline characteristics. All studies reported the exercise duration: 3 studies reported 12 weeks, 1 study for 1 weeks and 1 for 16 weeks. All studies included in our meta-analysis were published between 2016 and 2022, and the total sample size was 119. The detailed information on medication intake was shown in Table 2. The mean Jadad score ranged from 3 to 5. The main limitation of the included studies was the blinding methods. The Jadad scores for each study are also presented in Table 1.

Primary outcomes

Fasting blood glucose

Four studies examined pre- and post-LVHIIT FBG levels (10, 25, 27, 28). The results showed a significant difference in FBG in the LVHIIT group compared with the control group (RR= -1.21; 95% CI -2.02— -0.40, p = 0.0032), and there was significant heterogeneity (I^2 = 73%, P = 0.01; Figure 2A). After

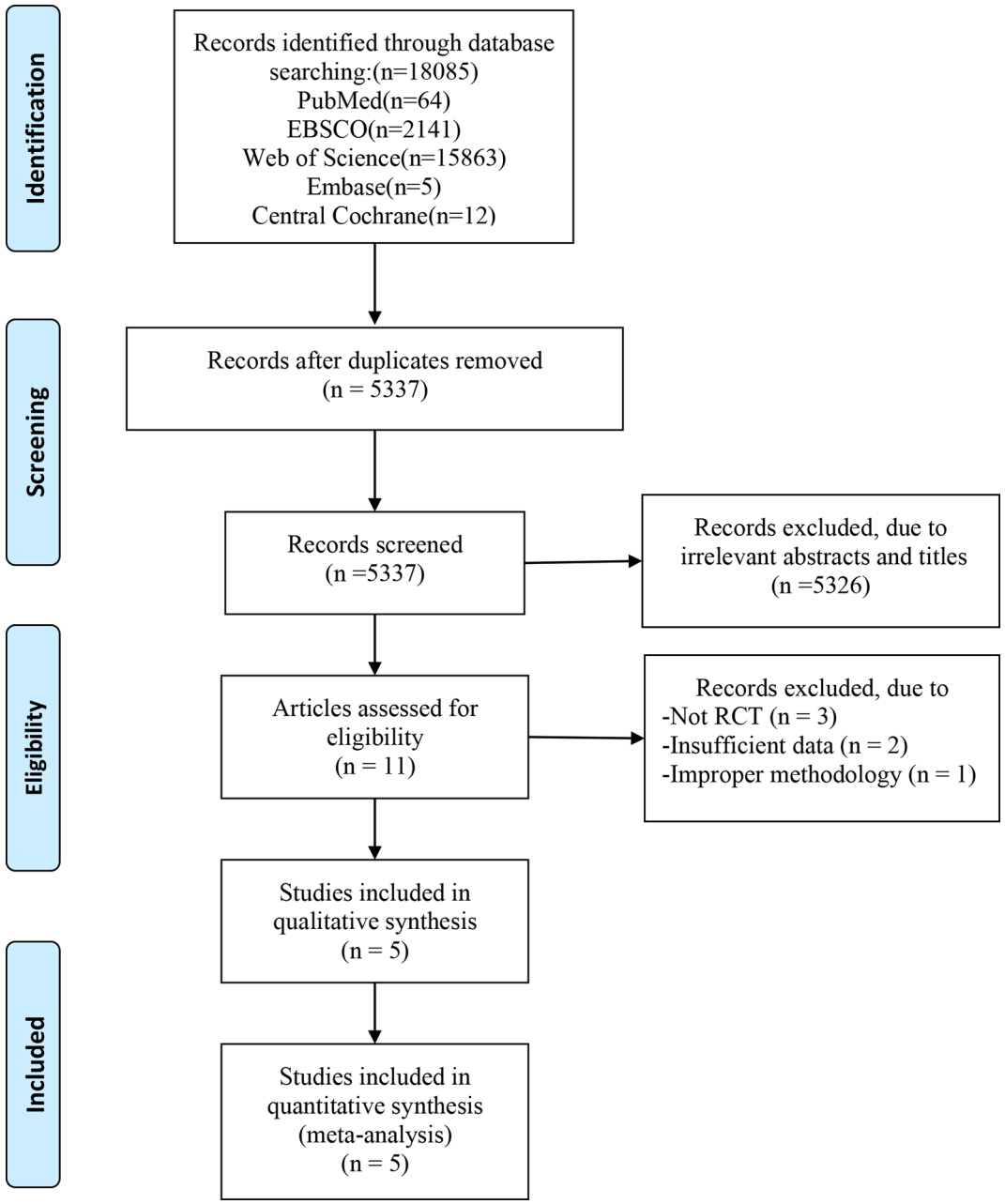

FIGURE 1
PRISMA flow chart.

removing the Winding et al. study (10), the heterogeneity became nonsignificant (I^2 = 0%, P = 0.78), and the overall effect of exercise remained significant (RR= -0.82; CI -1.22 – -0.42, p = 0.0032; Figure 2B).

HbA1c

Three studies examined changes in HbA1c levels (10, 25, 27). Our meta-analysis indicated that LVHIIT can significantly reduce the HbA1c levels (RR= -0.65; 95% CI= -1.06 – -0.23, p = 0.002; Figure 3), and there was nonsignificant heterogeneity (I^2 = 21%, p = 0.28).

HOMA-IR

Only two studies examined HOMA-IR levels (10, 28). As shown in Figure 3, HOMA-IR levels were significantly lower in the LVHIIT group than in the control group (RR= -1.34; 95% CI

TABLE 1 Characteristics of included studies.

No.	Author	Year	Low volume HIIT group					Control group					Jadad Score
			Number	Age (Years)	Male	Duration (Weeks)	Intervention	Number	Age (Years)	Male	Duration (Weeks)	Intervention	
1	Alvarez et al.	2016	13	46.0 ± 3.0	0	16	Jogging/running; 3 times/week; Progressive exercise program intervals interspersed with recovery periods of low-intensity walking; Reach 90–100% and less than 70% heart rate of their age predicted.	10	43.0 ± 2.4	0	16	Non-exercising	3
2	Winding et al.	2017	13	54.0 ± 6.0	7	11	Cycling; 3 times/week;20 min consisting of 1 minute at 95% Workload peak and 1 min 20% Work load peak recovery.	13	57 ± 7	5	11	Non-exercising	4
3	Afousi et al.	2018	18	54.78 ± 6.19	9	12	Cycling; 3 times/week; 1.5 min at 85–90% HRmax separated by 2 min at 55–60% HRmax.	17	54.24 ± 5.61	9	12	Non-exercising	3
4	Way et al.	2020	12	56.9 ± 2.1	7	12	Cycling; 3 times/week; 4 min high-intensity at 90% $\dot{V}_{O2}$peak and a 5 min cool-down at 50% $\dot{V}$O2peak	11	51.9 ± 1.4	7	12	Sham-exercise placebo	4
5	Li et al.	2022	13	38.0 ± 6.0	13	12	Cycling; 5 times/week; 8 minutes 80%–95% HRmax and recovery at 25% intensity.	12	40 ± 7	12	12	Non-exercising	5

-2.59— -0.10, p = 0.03; Figure 4), and there was significant heterogeneity (I^2 = 54%, P = 0.14).

Secondary outcomes

Fasting insulin

Four studies examined pre- and post-LVHIIT fasting insulin levels (10, 25, 27, 28). Our results revealed that LVHIIT did not significantly change the fasting insulin level compared to the control group (RR= -1.43; 95% CI = -3.46 – 0.60, p =0.17; Figure 5A), and there was significant heterogeneity (I^2 = 92%, p <0.00001). After removing the study by Li et al. (25), the overall effect of LVHIIT remained nonsignificant (RR= -3.52; 95% CI = -10.95 – 3.90, p =0.35; Figure 5B), and the level of heterogeneity was lower (I^2 = 31%, p =0.24).

BMI and body mass

All five included studies examined BMI and body mass (10, 25–28). Our results revealed that LVHIIT reduces BMI (RR = -0.31, 95% CI = -0.47 – -0.16, p<0.0001; Figure 6A) and body mass (RR = -0.94, 95% CI = -1.37 – -0.51, p<0.0001; Figure 6B), and there was nonsignificant heterogeneity (I^2 = 0%, P =0.82 and I^2 = 0%, P =0.92, respectively).

Blood lipid metabolism

Four studies (10, 26–28) examined data relating to blood lipid indicators. For total cholesterol, our meta-analysis indicated that there was no difference after a period of LVHIIT (RR = -1.24, 95% CI = -3.09—0.61, p=0.19; Figure 7A), and there was a significant level of heterogeneity (I^2 = 99%, P <0.00001). However, after excluding one study, LVHIIT was found to significantly reduce total cholesterol (RR = -0.24, 95% CI = -0.35 to -0.13, p<0.0001; Figure 7B), and the heterogeneity became nonsignificant (I^2 = 0%, P =0.68). For high-density lipoprotein, the meta-analysis showed that there was a significant difference between the LVHIIT group and the control group (RR = 0.21, 95% CI = 0.08 – 0.33, p=0.001; Figure 7E), and there was a significant level of heterogeneity (I^2 = 62%, P=0.05). After removing the study by Way et al., the results still showed that LVHIIT increased plasma HDL levels (RR = 0.28, 95% CI =0.24—0.32, p<0.00001), but the heterogeneity was nonsignificant (I^2 = 0%, P =0.58; Figure 7F). Overall, LVHIIT can reduce TC, LDL and triglyceride levels and increase HDL levels.

Cardiorespiratory fitness parameters

All five studies included the SBP and DBP (10, 25–28). Our results indicated that LVHIIT can reduce the SBP (RR =-4.01, 95% CI = -4.82 – -3.21, p<0.0001; Figure 8A) but not DPB (RR =-1.52, 95% CI = -3.31 – 0.26, p=0.09; Figure 8B) with nonsignificant

TABLE 2 Medication intake of patients.

Study	Medication intake (Control: LVHIIT), n
Alvarez et al.	Metformin 10:13; Glibenclamide 8:12; ACE inhibitor 3:3; Levothyroxine 1:1.
Winding et al.	Metformin 6:1; DPP-4 inhibitor 0:3; Sulfonylureas 1:3; GLP-1 analogues 1:2.
Afousi et al.	Diuretic 8:9; ACE inhibitors 5:4; Angiotensin blockers 4:3; Metformin10:9; Sulfonylureas 8:8; DPP-4 inhibitors 4:6; Statins 7:8.
Way et al.	Anti-Hyperglycemic 12:11; Anti-Hypertensive 8:6; Lipid Lowering 5:7.
Li et al.	Metformin 7:6; Sulfonylureas 3:3; DPP-4 inhibitors 2:3; Alpha-glucosidase inhibitor0:1.

heterogeneity (I^2 = 0%, P =0.77 and I^2 = 37%, P =0.18, respectively). Three studies reported the VO_{2peak} and our result showed that LVHIIT significantly improved VO_{2peak} compared to the control group (RR= 5.45; 95% CI = 1.38 – 9.52, p =0.009; Figure 8C) and there was significant heterogeneity (I^2 = 70%, p =0.04) (10, 26, 27). After removing the study by Way et al. (27), the overall effect of LVHIIT remained significant (RR= 7.66; 95% CI = 5.21 – 10.11, p<0.00001; Figure 8D), and the level of heterogeneity was lower (I^2 = 0%, p =0.70).

Discussion

Increased evidence has shown that physical exercise is an essential component of all effective interventions for the treatment and prevention of T2DM. As different types of exercise bring different benefits to patients, a series of clinical trials and meta-analyses have been performed to determine the positive function of each type of exercise. Earlier studies have shown that aerobic exercise, resistance training and HIIT independently have beneficial effects on preventing T2DM (5). As T2DM patients often report a "lack of time" as one barrier to regular exercise (29), LVHIIT may be a more time-effective strategy. LVHIIT has already been proven to improve cardiovascular health in T2DM patients. However, the effect of LVHIIT on diabetes-related indicators such as glycaemic control, insulin level, and HbA1c remain unclear. To evaluate this type of exercise and obtain higher-level evidence, we performed this meta-analysis.

Hyperglycaemia is the key characteristic of diabetes mellitus and is the main cause of complications in the heart, vasculature, eyes, kidneys and nerve system (30). Almost all types of exercise can reduce hyperglycaemia by improving insulin resistance in peripheral organs, such as skeletal muscle, liver and adipocytes (31–34), which will enhance blood glucose uptake and transport. Jelleyman et al. reported in their meta-analysis that regular HIIT can significantly reduce fasting glucose in metabolic syndrome or T2DM but not in healthy people compared to no exercise patients (35). Our present research found a reduction in fasting glucose levels among T2DM patients after LVHIIT intervention. Little et al. reported that LVHIIT can reduce hyperglycaemia by enhancing insulin signaling, the insulin-stimulated glucose disposal rate, glucose transporter protein (GLUT4) levels, and mitochondrial capacity in muscle, which further confirms our

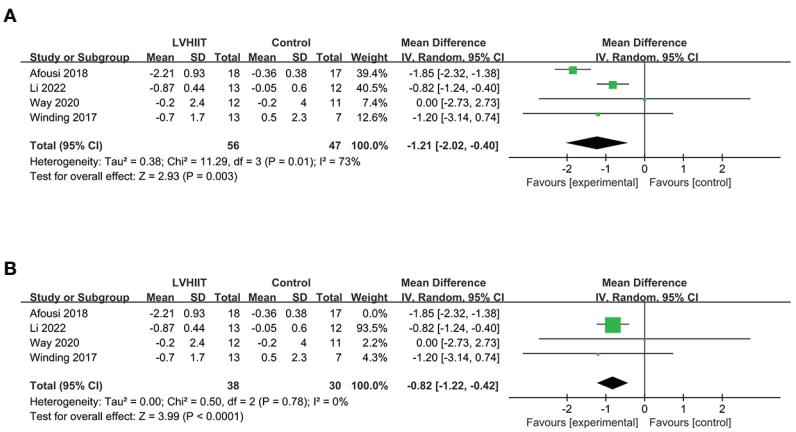

FIGURE 2
(A) Forest plot for the meta-analysis of **(A)** fasting blood glucose and **(B)** after sensitivity analysis.

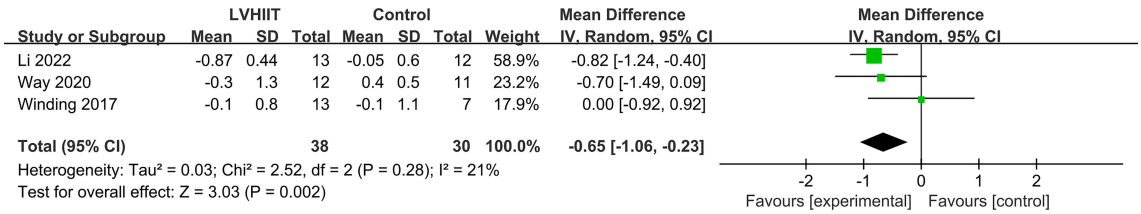

FIGURE 3
Forest plot for the meta-analysis of percentage of HbA1c.

results (34). HOMA-IR, a model for estimating insulin sensitivity through glucose concentrations and fasting insulin, was also improved in the LVHIIT group (36). The level of heterogeneity was higher for this outcome, which may be due to different blood sampling times and the specific calculation model they used. Thus, we can assume that LVHIIT can significantly improve hyperglycaemia and insulin resistance in T2DM patients. HbA1c is another indicator for blood glucose concentration and is a very important predictor for the incidence of complications and death related to diabetes. A previous study reported that each 1% increase in HbA1c is associated with a 37% increase in diabetic microvascular complications and a 21% increase in the risk of mortality. Thus, HbA1c is also a crucial marker for evaluating the therapeutic method of diabetes (37, 38). First, the formation of HbA1c is related to the average blood glucose concentration at three months. Second, a previous study showed that extent to which HbA1c levels decrease depends on the type and volume of exercise (39). Our results showed that medium- or long-term (11-16 weeks) LVHIIT can significantly reduce HbA1c and benefit T2DM patients. Notably, these glycaemic-related indicators were also improved in a short-term (2 weeks) experiment. Unfortunately, the study had a small sample size, and the evidence level of the study design was not high (34). Due to the improvement in insulin resistance, fasting insulin should be lower. However, our result shows a lower tendency of insulin without statistical significance. This result may partly explain why exercise improved insulin signaling in peripheral tissue rather than enhancing the insulin secretion function of β-cells (40). Ishiguro et al. assumed that insulin improvement may be restricted in patients with impaired basal insulin secretion with severe insulin resistance or impaired basal insulin secretion (41).

A previous meta-analysis that only included aerobic training and resistance training showed that BMI and body mass had nonsignificant reductions (42). Jelleyman et al. reported that HIIT can reduce body mass and BMI compared to the control group. However, the included study had relatively inconsistent baselines, as the included studies had different types of patients, such as healthy, overweight, T2DM and other chronic diseases (35). Our results further confirmed that LVHIIT can help reduce BMI and body weight in T2DM patients. However, body composition, such as body fat percentage, waist circumference and waist-hip ratio, should be further investigated. Our study also showed that LVHIIT significantly improved the blood lipid profile. The study by Way et al. contributed to the overall heterogeneity of this outcome because of the baseline characteristics of their patients (some patients were taking lipid-lowering medication). The main shortcoming for this outcome is the lack of the consistent dietary interventions across studies. Thus, future studies should provide a more consistent energy intake to determine the real efficiency of LVHIIT and plasma lipid metabolism. Corres et al. have reported that compared to high-volume moderate intensity continuous training, LVHIIT contributes to better improvements in cardiopulmonary function which is verified by our meta-analysis. Furthermore, they report that LVHIIT has a lowest withdraw rate compared to other type of exercises (43). Afousi et al. report that LVHIIT can decrease the oscillatory

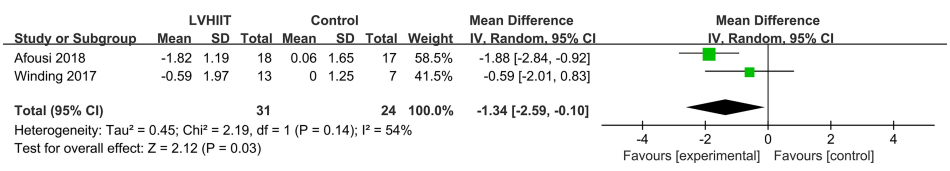

FIGURE 4
Forest plot for the meta-analysis of HOMA-IR.

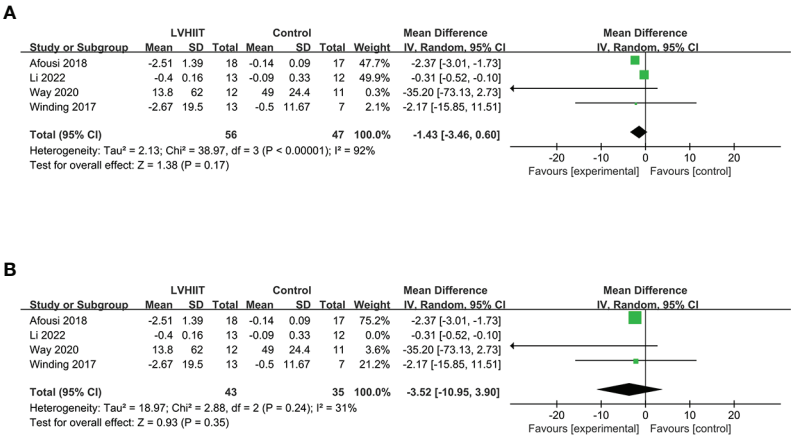

FIGURE 5
Forest plot for the meta-analysis of **(A)** fasting insulin and **(B)** after sensitivity analysis.

shear-induced improvement inflow-mediated dilatation and outward artery remodeling in T2DM patients compared to MICT (26).

Admittedly, there were some limitations in this meta-analysis. First, the number of participants in the studies was relatively small. Second, the LVHIIT period is approximately 11 to 16 weeks; therefore, it is difficult to determine the effects of shorter or longer LVHITT interventions. Third, there was always a certain amount of heterogeneity because there is no fully standardized LVHIIT protocol for T2DM patients. Fourth, liver dysfunction is tightly associated with T2DM (44), while the parameters of liver function were lacked in present studies. Lastly, the missing negative and unpublished data in the original studies may have led to publication bias and skewed our conclusions. Thus, we suggest that robust RCTs with large sample sizes and a standard protocol with more outcome parameters be performed in future studies to obtain more accurate data and verify our results.

Conclusion

In conclusion, this systematic review demonstrates that LVHIIT is an effective intervention for improving the metabolism of T2DM patients. Our results indicate that LVHIIT can reduce fasting blood glucose, HbA1c, insulin

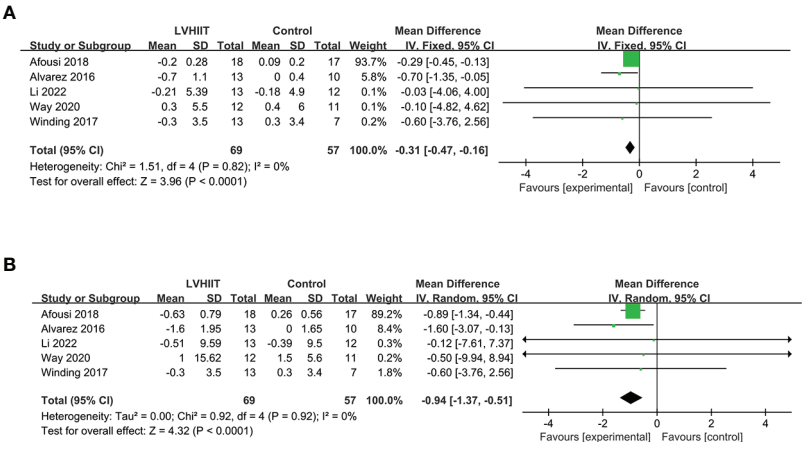

FIGURE 6
Forest plot for the meta-analysis of **(A)** body mass index and **(B)** body weight.

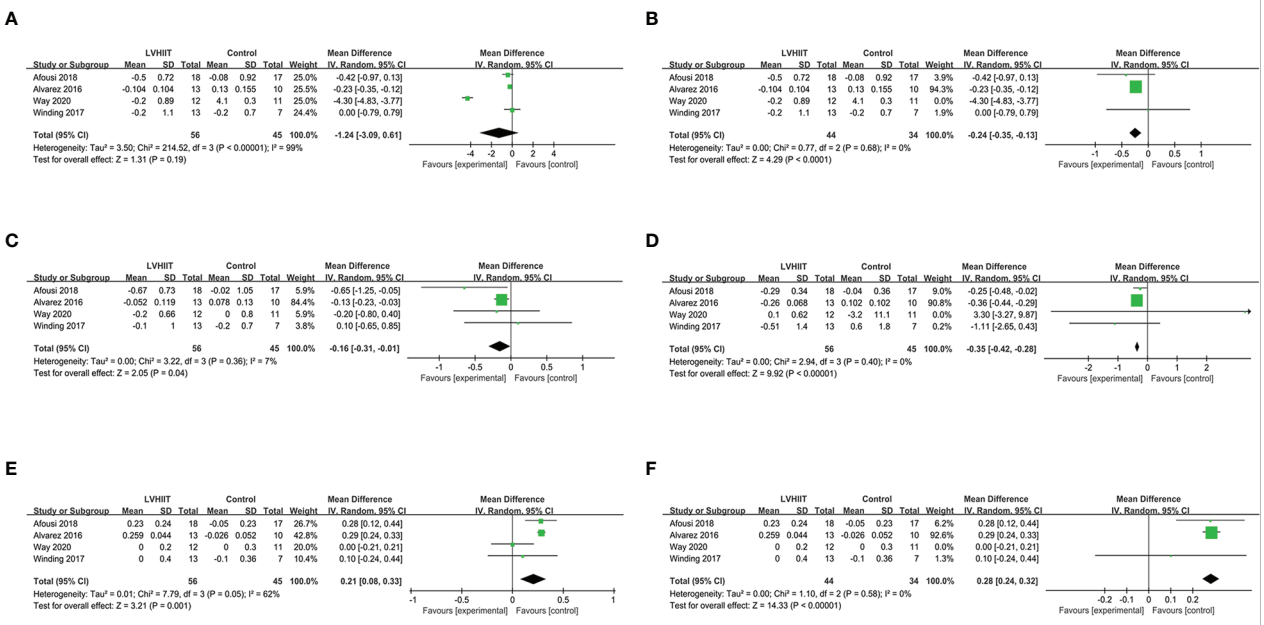

FIGURE 7
Forest plot for the meta-analysis of lipid profile **(A)** total cholesterol and **(B)** after sensitivity analysis; **(C)** low density lipoprotein; **(D)** triglyceride; **(E)** high density lipoprotein and **(F)** after sensitivity analysis.

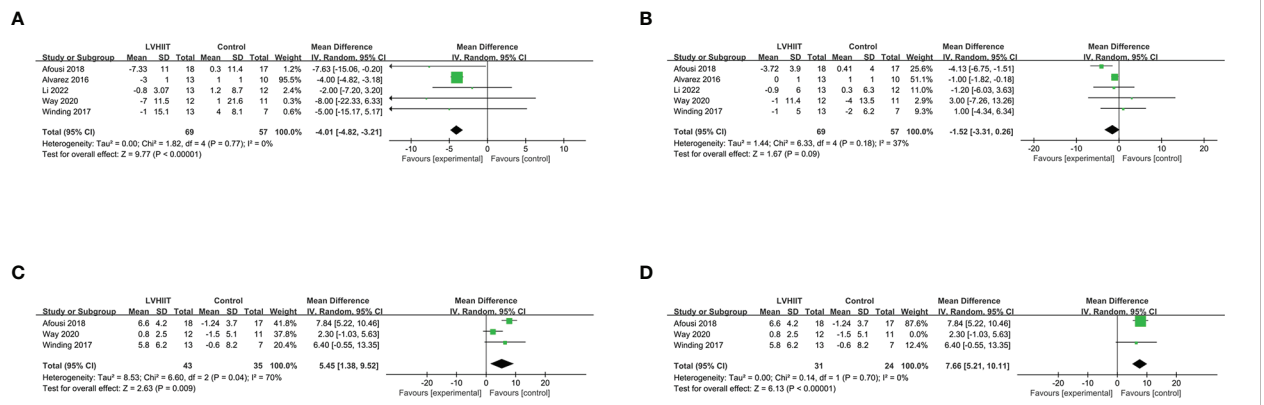

FIGURE 8
Forest plot for the meta-analysis of cardiorespiratory fitness **(A)** systolic blood pressure and **(B)** diastolic blood pressure; **(C)** relative VO2 peak; **(D)** after sensitivity analysis.

resistance, and body mass. Moreover, LVHIIT can improve the blood lipid profile, SBP and relative VO_{2peak}. Nevertheless. Because of the current limitations of the included studies, multicenter, large-scale, prospective RCTs with more stable baselines should be performed to validate the present results.

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.

Author contributions

ZW and XZ participated in the design of this study. YP and ZW drafted the manuscript, YP, YO, and KW collected and analysis the data, XZ critically revised the manuscript. All authors have read and approved the final manuscript.

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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.

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References

1. Ogurtsova K, da Rocha Fernandes JD, Huang Y, Linnenkamp U, Guariguata L, Cho NH, et al. Idf diabetes atlas: Global estimates for the prevalence of diabetes for 2015 and 2040. *Diabetes Res Clin Pract* (2017) 128:40–50. doi: 10.1016/j.diabres.2017.03.024

2. Wilson PW, Meigs JB. Cardiometabolic risk: A framingham perspective. *Int J Obes (Lond)* (2008) 32 Suppl 2:S17–20. doi: 10.1038/ijo.2008.30

3. Vetter ML, Wadden TA, Chittams J, Diewald LK, Panigrahi E, Volger S, et al. Effect of lifestyle intervention on cardiometabolic risk factors: Results of the power-up trial. *Int J Obes (Lond)* (2013) 37 Suppl 1(0 1):S19–24. doi: 10.1038/ijo.2013.92

4. Sagarra R, Costa B, Cabré JJ, Solà-Morales O, Barrio F. Lifestyle interventions for diabetes mellitus type 2 prevention. *Rev Clin Esp (Barc)* (2014) 214(2):59–68. doi: 10.1016/j.rce.2013.10.005

5. Zheng Y, Ley SH, Hu FB. Global aetiology and epidemiology of type 2 diabetes mellitus and its complications. *Nat Rev Endocrinol* (2018) 14(2):88–98. doi: 10.1038/nrendo.2017.151

6. Colberg SR, Albright AL, Blissmer BJ, Braun B, Chasan-Taber L, Fernhall B, et al. Exercise and type 2 diabetes: American college of sports medicine and the American diabetes association: Joint position statement. exercise and type 2 diabetes. *Med Sci Sports Exerc* (2010) 42(12):2282–303. doi: 10.1249/MSS.0b013e3181eeb61c

7. Knowler WC, Barrett-Connor E, Fowler SE, Hamman RF, Lachin JM, Walker EA, et al. Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. *N Engl J Med* (2002) 346(6):393–403. doi: 10.1056/NEJMoa012512

8. Magkos F, Hjorth MF, Astrup A. Diet and exercise in the prevention and treatment of type 2 diabetes mellitus. *Nat Rev Endocrinol* (2020) 16(10):545–55. doi: 10.1038/s41574-020-0381-5

9. Colberg SR, Sigal RJ, Yardley JE, Riddell MC, Dunstan DW, Dempsey PC, et al. Physical Activity/Exercise and diabetes: A position statement of the American diabetes association. *Diabetes Care* (2016) 39(11):2065–79. doi: 10.2337/dc16-1728

10. Winding KM, Munch GW, Iepsen UW, Van Hall G, Pedersen BK, Mortensen SP. The effect on glycaemic control of low-volume high-intensity interval training versus endurance training in individuals with type 2 diabetes. *Diabetes Obes Metab* (2018) 20(5):1131–9. doi: 10.1111/dom.13198

11. Taylor JL, Bonikowske AR, Olson TP. Optimizing outcomes in cardiac rehabilitation: The importance of exercise intensity. *Front Cardiovasc Med* (2021) 8:734278. doi: 10.3389/fcvm.2021.734278

12. Qiu S, Cai X, Yang B, Du Z, Cai M, Sun Z, et al. Association between cardiorespiratory fitness and risk of type 2 diabetes: A meta-analysis. *Obes (Silver Spring)* (2019) 27(2):315–24. doi: 10.1002/oby.22368

13. Mendes R, Sousa N, Reis VM, Themudo-Barata JL. Prevention of exercise-related injuries and adverse events in patients with type 2 diabetes. *Postgrad Med J* (2013) 89(1058):715–21. doi: 10.1136/postgradmedj-2013-132222

14. Gibala MJ, Little JP, Macdonald MJ, Hawley JA. Physiological adaptations to low-volume, high-intensity interval training in health and disease. *J Physiol* (2012) 590(5):1077–84. doi: 10.1113/jphysiol.2011.224725

15. Jayo-Montoya JA, Maldonado-Martín S, Aispuru GR, Gorostegi-Anduaga I, Gallardo-Lobo R, Matajira-Chia T, et al. Low-volume high-intensity aerobic interval training is an efficient method to improve cardiorespiratory fitness after myocardial infarction: Pilot study from the interfarct project. *J Cardiopulm Rehabil Prev* (2020) 40(1):48–54. doi: 10.1097/hcr.0000000000000453

16. Gorostegi-Anduaga I, Corres P, MartinezAguirre-Betolaza A, Pérez-Asenjo J, Aispuru GR, Fryer SM, et al. Effects of different aerobic exercise programmes with nutritional intervention in sedentary adults with Overweight/Obesity and hypertension: Exerdiet-hta study. *Eur J Prev Cardiol* (2018) 25(4):343–53. doi: 10.1177/2047487317749956

17. Martin-Smith R, Cox A, Buchan DS, Baker JS, Grace F, Sculthorpe N. High intensity interval training (Hiit) improves cardiorespiratory fitness (Crf) in healthy, overweight and obese adolescents: A systematic review and meta-analysis of controlled studies. *Int J Environ Res Public Health* (2020) 17(8):2955. doi: 10.3390/ijerph17082955

18. Sultana RN, Sabag A, Keating SE, Johnson NA. The effect of low-volume high-intensity interval training on body composition and cardiorespiratory fitness: A systematic review and meta-analysis. *Sports Med* (2019) 49(11):1687–721. doi: 10.1007/s40279-019-01167-w

19. Liberati A, Altman DG, Tetzlaff J, Mulrow C, Gøtzsche PC, Ioannidis JP, et al. The prisma statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: Explanation and elaboration. *PloS Med* (2009) 6(7):e1000100. doi: 10.1371/journal.pmed.1000100

20. García-Hermoso A, Cerrillo-Urbina AJ, Herrera-Valenzuela T, Cristi-Montero C, Saavedra JM, Martínez-Vizcaíno V. Is high-intensity interval training more effective on improving cardiometabolic risk and aerobic capacity than other forms of exercise in overweight and obese youth? a meta-analysis. *Obes Rev* (2016) 17(6):531–40. doi: 10.1111/obr.12395

21. Lora-Pozo I, Lucena-Anton D, Salazar A, Galán-Mercant A, Moral-Munoz JA. Anthropometric, cardiopulmonary and metabolic benefits of the high-intensity interval training versus moderate, low-intensity or control for type 2 diabetes: Systematic review and meta-analysis. *Int J Environ Res Public Health* (2019) 16 (22):4524. doi: 10.3390/ijerph16224524

22. Jadad AR, Moore RA, Carroll D, Jenkinson C, Reynolds DJ, Gavaghan DJ, et al. Assessing the quality of reports of randomized clinical trials: Is blinding necessary? *Control Clin Trials* (1996) 17(1):1–12. doi: 10.1016/0197-2456(95)00134-4

23. Kjaergard LL, Villumsen J, Gluud C. Reported methodologic quality and discrepancies between Large and small randomized trials in meta-analyses. *Ann Intern Med* (2001) 135(11):982–9. doi: 10.7326/0003-4819-135-11-200112040-00010

24. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. *Stat Med* (2002) 21(11):1539–58. doi: 10.1002/sim.1186

25. Li J, Cheng W, Ma H. A comparative study of health efficacy indicators in subjects with T2dm applying power cycling to 12 weeks of low-volume high-intensity interval training and moderate-intensity continuous training. *J Diabetes Res* (2022) 2022:9273830. doi: 10.1155/2022/9273830

26. Ghardashi Afousi A, Izadi MR, Rakhshan K, Mafi F, Biglari S, Gandomkar Bagheri H. Improved brachial artery shear patterns and increased flow-mediated dilatation after low-volume high-intensity interval training in type 2 diabetes. *Exp Physiol* (2018) 103(9):1264–76. doi: 10.1113/ep087005

27. Way KL, Sabag A, Sultana RN, Baker MK, Keating SE, Lanting S, et al. The effect of low-volume high-intensity interval training on cardiovascular health outcomes in type 2 diabetes: A randomised controlled trial. *Int J Cardiol* (2020) 320:148–54. doi: 10.1016/j.ijcard.2020.06.019

28. Alvarez C, Ramirez-Campillo R, Martinez-Salazar C, Mancilla R, Flores-Opazo M, Cano-Montoya J, et al. Low-volume high-intensity interval training as a therapy for type 2 diabetes. *Int J Sports Med* (2016) 37(9):723–9. doi: 10.1055/s-0042-104935

29. Thomas N, Alder E, Leese GP. Barriers to physical activity in patients with diabetes. *Postgrad Med J* (2004) 80(943):287–91. doi: 10.1136/pgmj.2003.010553

30. Stumvoll M, Goldstein BJ, van Haeften TW. Type 2 diabetes: Principles of pathogenesis and therapy. *Lancet* (2005) 365(9467):1333–46. doi: 10.1016/s0140-6736(05)61032-x

31. Dunstan DW, Daly RM, Owen N, Jolley D, De Courten M, Shaw J, et al. High-intensity resistance training improves glycemic control in older patients with type 2 diabetes. *Diabetes Care* (2002) 25(10):1729–36. doi: 10.2337/diacare.25.10.1729

32. Thompson D, Karpe F, Lafontan M, Frayn K. Physical activity and exercise in the regulation of human adipose tissue physiology. *Physiol Rev* (2012) 92 (1):157–91. doi: 10.1152/physrev.00012.2011

33. Diniz TA, de Lima Junior EA, Teixeira AA, Biondo LA, da Rocha LAF, Valadão IC, et al. Aerobic training improves nafld markers and insulin resistance through ampk-Ppar-α signaling in obese mice. *Life Sci* (2021) 266:118868. doi: 10.1016/j.lfs.2020.118868

34. Little JP, Gillen JB, Percival ME, Safdar A, Tarnopolsky MA, Punthakee Z, et al. Low-volume high-intensity interval training reduces hyperglycemia and increases muscle mitochondrial capacity in patients with type 2 diabetes. *J Appl Physiol (1985)* (2011) 111(6):1554–60. doi: 10.1152/japplphysiol.00921.2011

35. Jelleyman C, Yates T, O'Donovan G, Gray LJ, King JA, Khunti K, et al. The Effects of high-intensity interval training on glucose regulation and insulin resistance: A meta-analysis. *Obes Rev* (2015) 16(11):942–61. doi: 10.1111/obr.12317

36. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: Insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia* (1985) 28 (7):412–9. doi: 10.1007/bf00280883

37. Woerle HJ, Neumann C, Zschau S, Tenner S, Irsigler A, Schirra J, et al. Impact of fasting and postprandial glycemia on overall glycemic control in type 2 diabetes importance of postprandial glycemia to achieve target Hba1c levels. *Diabetes Res Clin Pract* (2007) 77(2):280–5. doi: 10.1016/j.diabres.2006.11.011

38. Sung KC, Seo MH, Rhee EJ, Wilson AM. Elevated fasting insulin predicts the future incidence of metabolic syndrome: A 5-year follow-up study. *Cardiovasc Diabetol* (2011) 10:108. doi: 10.1186/1475-2840-10-108

39. Umpierre D, Ribeiro PA, Schaan BD, Ribeiro JP. Volume of supervised exercise training impacts glycaemic control in patients with type 2 diabetes: A systematic review with meta-regression analysis. *Diabetologia* (2013) 56(2):242–51. doi: 10.1007/s00125-012-2774-z

40. Holten MK, Zacho M, Gaster M, Juel C, Wojtaszewski JF, Dela F. Strength training increases insulin-mediated glucose uptake, Glut4 content, and insulin signaling in skeletal muscle in patients with type 2 diabetes. *Diabetes* (2004) 53 (2):294–305. doi: 10.2337/diabetes.53.2.294

41. Ishiguro H, Kodama S, Horikawa C, Fujihara K, Hirose AS, Hirasawa R, et al. In search of the ideal resistance training program to improve glycemic control and its indication for patients with type 2 diabetes mellitus: A systematic review and meta-analysis. *Sports Med* (2016) 46(1):67–77. doi: 10.1007/s40279-015-0379-7

42. Boulé NG, Haddad E, Kenny GP, Wells GA, Sigal RJ. Effects of exercise on glycemic control and body mass in type 2 diabetes mellitus: A meta-analysis of controlled clinical trials. *Jama* (2001) 286(10):1218–27. doi: 10.1001/jama.286.10.1218

43. Corres P, MartinezAguirre-Betolaza A, Fryer SM, Gorostegi-Anduaga I, Arratibel-Imaz I, Aispuru GR, et al. Long-term effects in the exerdiet-hta study: Supervised exercise training vs. physical activity advice. *Res Q Exerc Sport* (2020) 91 (2):209–18. doi: 10.1080/02701367.2019.1656794

44. De Silva NMG, Borges MC, Hingorani AD, Engmann J, Shah T, Zhang X, et al. Liver function and risk of type 2 diabetes: Bidirectional mendelian randomization study. *Diabetes* (2019) 68(8):1681–91. doi: 10.2337/db18-1048

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Sex difference in the associations among obesity-related indices with incidence of diabetes mellitus in a large Taiwanese population follow-up study

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Background: Obesity is a major risk factor for diabetes mellitus (DM), which is in turn a major risk factor for cardiovascular diseases such as coronary artery disease and stroke. As few studies have investigated sex differences in the association between obesity and incidence of DM, the aim of this longitudinal study was to explore this issue in a large group of Taiwanese participants.

Methods: A total of 24,346 participants were enrolled in this study, of whom 8,334 (mean age, 50.6 ± 11.0 years) were male and 16,012 (mean age, 50.5 ± 10.1 years) were female. The following obesity-related indices were studied: body mass index, waist-to-height ratio, waist-to-hip ratio (WHR), body roundness index, conicity index (CI), body adiposity index, abdominal volume index, lipid accumulation product (LAP), and visceral adiposity index (VAI).

Results: The analysis showed significant associations between all of these indices with incidence of DM (all $p < 0.001$). In the male participants, the strongest predictors for incidence of DM were LAP (AUC = 0.692), WHtR (AUC = 0.684), and WHR (AUC = 0.683). In the female participants, the strongest predictors were LAP (AUC = 0.744), WHtR (AUC = 0.710) and VAI (AUC = 0.710), followed by BRI (AUC = 0.708).

Conclusion: Strong associations were found between the studied obesity-related indices and incidence of DM, and sex differences were found. Hence, to better control DM, reducing body weight may be beneficial in addition to lifestyle modifications, diet control, and pharmacological interventions.

KEYWORDS

obesity-related index, sex difference, incident diabetes mellitus, Taiwan biobank, follow-up

Introduction

Diabetes is one of the most prevalent metabolic disorders worldwide, and it is associated with severe complications and heavy financial and medical burdens. According to the 10th International Diabetes Federation Diabetes Atlas, the estimated global prevalence of diabetes among individuals aged 20 to 79 years in 2021 was 10.5% (536.6 million people) (1). According to the Taiwan Health Promotion Administration, the prevalence of diabetes mellitus (DM)

in Taiwan between 2017 and 2020 was 11.05%, which is higher than the global prevalence (10.5%) (2).

Type 2 DM is the most common type of diabetes, characterized by insulin resistance, decrease in the number of beta cells, and hyperglycemia (3). Type 2 DM complications include cardiovascular diseases and other microvascular diseases affecting the kidneys, retina, and neurological system, leading to poor clinical outcomes (4). DM-related complications may pose a considerable economic burden on society even while having negative effects on society for patients and their families. Moreover, according to the cause of death statistics of 2021 from Taiwan Ministry of Health and Welfare, diabetes is one of Taiwan's top ten causes of death, and the number of deaths is increasing year by year (5). Therefore, it is of great importance to identify the risk factors associated with the development type 2 DM.

Many anthropometric indicators are simple and convenient tools for evaluating central obesity and the risk of metabolic syndrome. Such indicators include waist-to-hip ratio (WHR), waist-to-height ratio (WHtR), lipid accumulation product (LAP), body roundness index (BRI), visceral adiposity index (VAI), abdominal volume index (AVI), conicity index (CI), and body adiposity index (BAI) (6, 7). All of these anthropometric indicators can be calculated using simple clinical measurements such as waist circumference (WC), hip circumference (HC), body mass index (BMI), body height (BH), body weight (BW), triglycerides (TG), and high-density lipoprotein cholesterol (HDL-C). These obesity-related indices can evaluate obesity, which is defined by an excess accumulation of adipose tissue. Previous research has suggested the relationship between obesity with insulin resistance and type 2 diabetes, which is that non-esterified fatty acids secreted from adipose tissue in obese people may lead to insulin resistance and β-cell dysfunction (8). Our recent research revealed that these obesity indices are associated with fatty liver (9), albuminuria and advanced kidney disease (10), lung function (11), osteoporosis (12), hypertension (13), peripheral artery disease (14), and dementia (15). Previous studies have also demonstrated a relationship between obesity-related indices and DM (16–20). Nevertheless, few studies have investigated sex differences in the relationships between obesity-related indices and incidence of DM.

In this population-based cohort study, we enrolled over 26,000 participants from the Taiwan Biobank (TWB) and examined sex differences in the associations between obesity-related indices and incidence of DM. In addition, we determined the cutoff value of each obesity index to predict incidence of DM in men and women.

Materials and methods

Taiwan biobank

The population in Taiwan is rapidly aging, and hence the Ministry of Health and Welfare created the TWB to promote health care and prevent chronic diseases. The participants in the TWB are aged from 30 to 70 years and none have a previous diagnosis of cancer. Data available in the TWB include medical, genomic and lifestyle factors (21, 22). Ethical approval for the TWB was given by the Ethics and Governance Council of the TWB and Institutional Review Board on Biomedical Science Research, Academia Sinica, Taiwan.

During enrollment into the TWB, all participants provided data about their age and personal medical history (i.e. hypertension and DM). They also underwent physical examinations to obtain information on WC, HC, BW, BH and BMI. Fasting serum samples were obtained from all of the patients, and laboratory tests were conducted using an auto-analyzer (Roche Diagnostics GmbH, D-68298 Mannheim COBAS Integra 400). Laboratory data were also recorded at baseline after an 8-hour fast including: fasting glucose, glycated hemoglobin A1c (HbA1c), hemoglobin, TGs, total cholesterol, HDL-C, low-density lipoprotein cholesterol (LDL-C), estimated glomerular filtration rate (eGFR) [calculated using the 4-variable Modification of Diet in Renal Disease study equation (23)], and uric acid. Serum levels of creatinine were calculated using the compensated Jaffé (kinetic alkaline picrate) method using a calibrator that could traced in isotope-dilution mass spectrometry (24).

Systolic blood pressure (BP) and diastolic BP measurements were also performed in each participant with an automated BP monitor by a trained staff member. All measurements were made in triplicate after abstaining from smoking, caffeine, and exercise for at least 30 min. We used average BP measurements for analysis. Regular exercise was defined as exercising at least three times a week for at least 30 min each time, which is based on the projected "Physical Fitness 333 Plan," promoted by the Ministry of Education in Taiwan in 1999 (25). Due to the widespread promotion, people in Taiwan still follow the "Physical Fitness 333 Plan" as a guideline for regular exercise. This study was conducted according to the Declaration of Helsinki, and approved by the Institutional Review Board of Kaohsiung Medical University Hospital [KMUHIRB-E(I)-20210058].

Study design

This study is an observational cohort study.

Sample population and sample size

A total of 27,033 participants (males: 9,555; females: 17,478) were screened, of whom had follow-up data for a median of 4 years and signed informed consent forms. Those with no data on WC ($n = 1$), HC ($n = 1$), BH ($n = 1$), and BW ($n = 4$), those with no follow-up data on DM, serum fasting glucose or HbA1c ($n = 43$), and those with baseline DM ($n = 2,637$) were excluded. The remaining 24,346 participants (males: 8,334; females: 16,012) were enrolled (Figure 1).

Definition of incidence of DM

Participants with an HbA1c level <6.5%, fasting glucose level <126 mg/dL, and no self-reported past history of DM were classified into the non-DM group. The participants in whom DM developed during follow-up, defined as an HbA1c level ≥6.5%, fasting glucose level ≥126 mg/dL (26), or self-reported DM, were classified into the incidence of DM group.

Calculation of obesity-related indices

1. BMI was calculated as:

$$BMI = BW(kg)/BH^2(m)$$

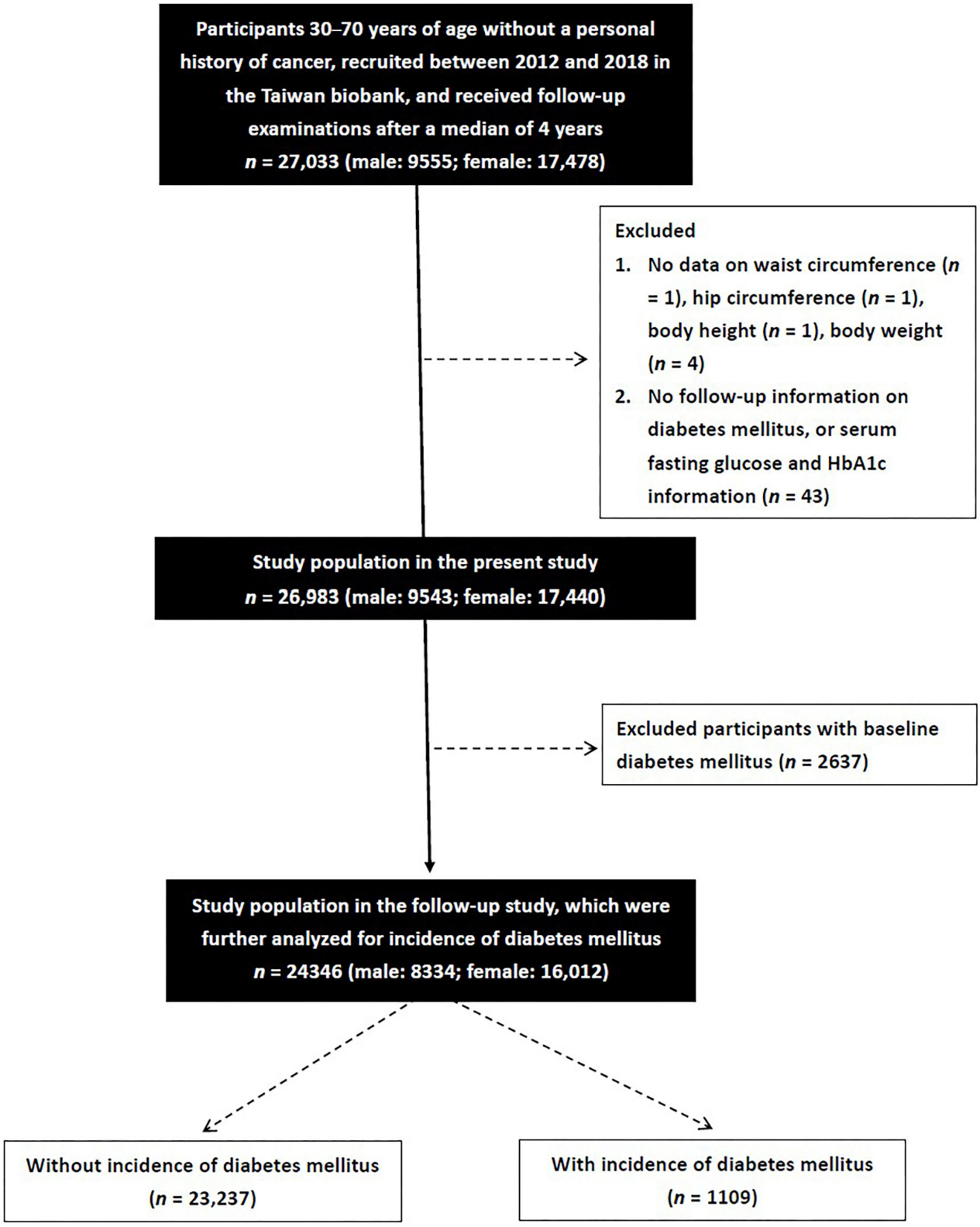

FIGURE 1
A total of 27,033 participants were screened. After exclusion those without waist circumference, hip circumference, body height, body weight, follow-up data on diabetes mellitus, serum fasting glucose or HbA1c, and those with baseline diabetes mellitus, the remaining 24,346 participants were enrolled.

2. WHR was calculated as:

$$\mathrm{WHR} = \mathrm{WC(cm)}/\mathrm{HC(cm)}$$

3. WHtR was calculated as:

$$\mathrm{WHtR} = \mathrm{WC(cm)}/\mathrm{BH(cm)}$$

4. BRI was calculated as:

$$\mathrm{BRI} = 364.2 - 365.5 \times \sqrt{1 - \left(\frac{\frac{\mathrm{WC_{(m)}}}{2\pi}}{0.5 \times \mathrm{BH_{(m)}}}\right)^2} \quad (27).$$

5. CI was calculated using the Valdez equation based on BW, BH and WC as:

$$\mathrm{CI} = \frac{\mathrm{WC_{(m)}}}{0.109 \times \sqrt{\frac{\mathrm{BW_{(kg)}}}{\mathrm{BH_{(m)}}}}} \quad (28).$$

6. BAI was calculated according to the method of Bergman and colleagues as:

$$\mathrm{BAI} = \frac{\mathrm{HC_{(cm)}}}{\mathrm{BH_{(m)}}^{3/2}} - 18 \quad (29).$$

7. AVI was calculated as

$$\mathrm{AVI} = \frac{2 \times (\mathrm{WC_{(cm)}})^2 + 0.7 \times (\mathrm{WC_{(cm)}} - \mathrm{HC_{(cm)}})^2}{1000} \quad (30).$$

8. LAP was calculated as:

$$\mathrm{LAP} = (\mathrm{WC_{(cm)}} - 65) \times \mathrm{TG_{(\frac{mmol}{L})}} \mathrm{inmalesandLAP}$$
$$= (\mathrm{WC_{(cm)}} - 58) \times \mathrm{TG_{(\frac{mmol}{L})}} \mathrm{infemales} \quad (31).$$

9. VAI score was calculated as described previously (32) using the following sex-specific equations (with TG levels in mmol/l and HDL-cholesterol levels in mmol/l):

$$\mathrm{VAI} = \left(\frac{\mathrm{WC_{(cm)}}}{39.68 + (1.88 \times \mathrm{BMI})}\right) \times \left(\frac{\mathrm{TG_{(\frac{mmol}{L})}}}{1.03}\right) \times \left(\frac{1.31}{\mathrm{HDL_{(\frac{mmol}{L})}}}\right) \mathrm{inmalesand}$$

$$\mathrm{VAI} = \left(\frac{\mathrm{WC_{(cm)}}}{36.58 + (1.89 \times \mathrm{BMI})}\right) \times \left(\frac{\mathrm{TG_{(\frac{mmol}{L})}}}{0.81}\right) \times \left(\frac{1.52}{\mathrm{HDL_{(\frac{mmol}{L})}}}\right) \mathrm{in\ females.}$$

TABLE 1 Clinical characteristics of the study participants classified by sex.

Characteristics	Male (n = 8,334)	Female (n = 16,012)	p
Age (year)	50.6 ± 11.0	50.5 ± 10.1	0.376
Hypertension (%)	15.1	8.8	<0.001
Smoking history (%)	58.2	7.5	<0.001
Alcohol history (%)	7.0	0.7	<0.001
Regular exercise habits (%)	48.3	47.4	0.188
Menstruation in female (%)	-	47.1	
Systolic BP (mmHg)	121.4 ± 16.1	113.9 ± 17.4	<0.001
Diastolic BP (mmHg)	76.9 ± 10.4	69.7 ± 10.2	<0.001
Body height (cm)	168.9 ± 6.2	157.1 ± 5.6	<0.001
Body weight (Kg)	71.1 ± 10.7	57.6 ± 9.0	<0.001
Waist circumference (cm)	86.7 ± 8.6	80.2 ± 9.2	<0.001
Hip circumference (cm)	96.9 ± 6.3	94.8 ± 6.7	<0.001
Laboratory parameters			
Fasting glucose (mg/dL)	94.4 ± 7.6	90.9 ± 7.4	<0.001
HbA1c (%)	5.59 ± 0.34	5.56 ± 0.34	<0.001
Hemoglobin (g/dL)	15.0 ± 1.1	13.0 ± 1.3	<0.001
Triglyceride (mg/dL)	130.4 ± 97.8	98.7 ± 61.3	<0.001
Total cholesterol (mg/dL)	192.7 ± 34.0	197.5 ± 35.3	<0.001
HDL-C (mg/dL)	48.5 ± 11.0	58.3 ± 13.0	<0.001
LDL-C (mg/dL)	123.2 ± 31.0	121.4 ± 31.4	<0.001
eGFR (mL/min/1.73 m^2)	99.1 ± 19.8	114.82 ± 25.6	<0.001
Uric acid (mg/dL)	6.5 ± 1.4	4.9 ± 1.1	<0.001
Obesity-related indices			
BMI (kg/m^2)	24.9 ± 3.3	23.3 ± 3.4	<0.001
WHR (%)	89.4 ± 5.4	84.5 ± 6.6	<0.001
WHtR (%)	51.4 ± 5.1	51.1 ± 6.1	<0.001
BRI	7.0 ± 1.6	6.4 ± 1.8	<0.001
CI	1.23 ± 0.06	1.22 ± 0.09	<0.001
BAI	26.2 ± 3.0	30.2 ± 3.8	<0.001
AVI	15.3 ± 3.0	13.2 ± 3.0	<0.001
LAP	34.4 ± 33.5	26.5 ± 23.3	<0.001
VAI	1.7 ± 1.7	1.6 ± 1.4	<0.001

DM, diabetes mellitus; BP, blood pressure; HbA1c, glycosylated hemoglobin A1c; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; BMI, body mass index; WHR, waist–hip ratio; WHtR, waist-to-height ratio; BRI, body roundness index; CI, conicity index; BAI, body adiposity index; AVI, abdominal volume index; LAP, lipid accumulation product; VAI, visceral adiposity index.

Statistical analysis

SPSS (version 19, IBM Inc., Armonk, NY) was used for all statistical analyses. All continuous variables are expressed as mean ± SD, categorical variables as percentages, or medians (25^{th}-75^{th} percentiles) were used to describe LAP and VAI. Normality tests were done to analyze the distribution of data collected for each group using the Kolmogorov-Smirnov. Homogeneity of variance was tested with the Levene test (Levene's test was used to assess the equality of variances, and an independent sample t-test). With respect to the comparison of data from different groups, the independent t-test was used for normally distributed variables, while the Mann–Whitney U test was used for non-normally distributed variables. Differences in categorical variables were examined by the chi-square test. Multivariable logistic regression analysis was performed to analyze associations between incidence of DM and the obesity-related indices, including all of the significant variables in univariate analysis. The natural logarithm was used for LAP and VAI. Receiver operating

characteristic (ROC) curve analysis and areas under the ROC curves (AUCs) were used to assess the performance and predictive ability of the obesity-related indices for incidence of DM, respectively. The Optimal Cutoff Value was found as the cutoff with highest Youden Index, or equivalently, the highest Sensitivity + Specificity. *P* values < 0.05 were considered to be statistically significant.

Results

The mean age of the 24,346 enrolled patients was 50.5 ± 10.4 years. The prevalence rate of incidence of DM was 5.7% in the males ($n = 8,334$) and 4.0% in the females ($n = 16,012$) ($p < 0.001$).

Comparisons of the clinical characteristics between the male and female participants

Compared to the male participants, the female participants had lower prevalence rates of hypertension, smoking and alcohol history, lower values of systolic and diastolic BP, BH, BW, WC, HC, fasting glucose, HbA1c, hemoglobin, TGs, LDL-C and uric acid, and higher total cholesterol, HDL-C and eGFR (Table 1). The values of eight of the nine studied obesity-related indices (BMI, WHR, WHtR, BRI, CI, AVI, LAP, and VAI but not BAI) were lower in the female participants than in the male participants.

Comparisons of the clinical characteristics between the male and female participants with and without incidence of DM

The male participants with incidence of DM were older and had higher rates of hypertension and a history of smoking and alcohol consumption than those without incidence of DM (Table 2). In addition, the male participants with incidence of hypertension had higher systolic and diastolic BPs, BH, BW, WC, HC, fasting glucose, HbA1c, TGs, total cholesterol, LDL-C and uric acid, and lower HDL-C and eGFR than those without incidence of DM. Moreover, the male participants with incidence of DM had higher values of all nine obesity-related indices than those without incidence of DM.

The female participants with incidence of DM were older, had a higher prevalence rate of hypertension, lower menstruation status, higher systolic and diastolic BPs, BW, WC, HC, fasting glucose, HbA1c, hemoglobin, TGs, total cholesterol, LDL-C and uric acid, and lower BH, HDL-C and eGFR than those without incidence of DM. Moreover, the female participants with incidence of DM had higher values of all studied obesity-related indices than those without incidence of DM.

Associations among obesity-related indices with incidence of DM by sex

The following multivariable logistic regression models were used to examine the associations between each obesity-related index with incidence of DM by sex:

1. For WHtR, WHR, CI, BRI, BMI, BAI, and AVI: adjustments for age, hypertension, smoking and alcohol history, systolic and diastolic BPs, hemoglobin, TGs, total cholesterol, HDL-C, LDL-C, eGFR and uric acid in males (significant variables in Table 2); age, hypertension, menstruation status, systolic and diastolic BPs, hemoglobin, TGs, total cholesterol, HDL-C, LDL-C, eGFR and uric acid in females (significant variables in Table 2).
2. For LAP: the same adjustments as in model 1 except for TGs.
3. For VAI: the same adjustments as in model 1 except for TGs and HDL-C.

The results showed that high BMI (per 1 kg/m^2; odds ratio [OR] = 1.140), WHR (per 0.01; OR = 1.082), WHtR (per 0.01; OR = 1.088), BRI (per 1; OR = 1.263), CI (per 0.1; OR = 1.425), BAI (per 1; OR = 1.082), AVI (per 1; OR = 1.124), LAP (log per 1; OR = 8.951) and VAI (log per 1; OR = 9.104) were significantly associated with incidence of DM in the male participants (all $p < 0.001$) (Table 3). Similarly, high BMI (per 1 kg/m^2; OR = 1.099), WHR (per 0.01; OR = 1.051), WHtR (per 0.01; OR = 1.056), BRI (per 1; OR = 1.171), CI (per 0.1; OR = 1.233), BAI (per 1; OR = 1.043), AVI (per 1; OR = 1.095), LAP (log per 1; OR = 8.687), and VAI (log per 1; OR = 6.629) were significantly associated with incidence of DM in the female participants (all $p < 0.001$).

Significant interactions were found between sex and WHR (β = −0.024; $p = 0.022$), CI (β = −0.232; $p = 0.008$), LAP (β = 0.008; $p < 0.001$), and VAI (β = 0.092; $p = 0.001$) on incident DM. However, no significant differences were found in the other indices. In the men, WHR and CI were more strongly correlated with incidence of DM than in the women, whereas LAP and VAI were more closely linked with incidence of DM in the women than in the men.

Performance and predictive ability of the obesity-related indices to identify incidence of DM in the male and female participants

The performance (ROC curves), predictive ability (AUCs) and J value of the obesity-related indices to identify incidence of DM in the male and female participants were analyzed (Figure 2). Table 4 showed that in the male participants, LAP had the highest AUC (0.692), followed by WHtR (0.684), WHR (0.683), BRI (0.677), VAI (0.671), AVI (0.661), BMI (0.660), CI (0.630), and BAI (0.626). In addition, Table 5 showed that in the female participants, LAP also had the highest AUC (0.744), followed by WHtR (0.710), VAI (0.710), BRI (0.708), BMI (0.700), AVI (0.698), WHR (0.693), BAI (0.642), and CI (0.630).

The AUC, cutoff, sensitivity, specificity, and Youden index values of the obesity-related indices to identify incidence of DM in the male and female participants are shown in Tables 4, 5, respectively. In male participants, LAP had the highest Youden index (0.282), followed by VAI (0.267) and WHR (0.256). In addition, in female participants, LAP had the highest Youden index (0.282), followed by WHtR (0.320), BMI (0.304), and VAI (0.256).

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TABLE 2 Clinical characteristics of the study participants classified by the presence of different sex and incidence of DM.

Characteristics	Male ($n = 8,334$)			Female ($n = 16,012$)		
	Non-diabetic ($n = 7,858$)	Diabetic ($n = 476$)	*p*	Non-diabetic ($n = 15,379$)	Diabetic ($n = 633$)	*p*
Age (year)	50.4 ± 11.0	53.9 ± 9.9	<0.001	50.3 ± 10.1	55.3 ± 8.3	<0.001
Hypertension (%)	14.3	29.0	<0.001	8.2	22.9	<0.001
Smoking history (%)	57.9	64.3	0.006	7.5	6.2	0.201
Alcohol history (%)	6.8	10.5	0.002	0.7	0.3	0.446
Regular exercise habits (%)	48.4	45.6	0.228	47.2	51.0	0.061
Menstruation in female (%)	-	-	-	47.9	27.8	<0.001
Systolic BP (mmHg)	121.1 ± 16.0	126.4 ± 16.6	<0.001	113.5 ± 17.2	124.4 ± 18.5	<0.001
Diastolic BP (mmHg)	76.8 ± 10.4	79.2 ± 10.3	<0.001	69.6 ± 10.2	74.1 ± 10.3	<0.001
Body height (cm)	169.0 ± 6.3	167.6 ± 6.1	<0.001	157.1 ± 5.6	155.9 ± 5.2	<0.001
Body weight (Kg)	70.8 ± 10.5	75.2 ± 12.4	<0.001	57.4 ± 8.9	62.4 ± 9.7	<0.001
Waist circumference (cm)	86.4 ± 8.5	91.6 ± 9.0	<0.001	80.0 ± 9.1	86.5 ± 9.2	<0.001
Hip circumference (cm)	96.8 ± 6.2	98.7 ± 7.2	<0.001	94.7 ± 6.7	97.2 ± 7.2	<0.001
Laboratory parameters						
Fasting glucose (mg/dL)	93.9 ± 7.2	102.8 ± 9.8	<0.001	90.5 ± 7.0	100.5 ± 10.0	<0.001
HbA1c (%)	5.57 ± 0.33	6.01 ± 0.29	<0.001	5.54 ± 0.33	6.04 ± 0.28	<0.001
Hemoglobin (g/dL)	15.0 ± 1.1	15.1 ± 1.3	0.118	13.0 ± 1.3	13.3 ± 1.2	<0.001
Triglyceride (mg/dL)	127.3 ± 90.8	181.9 ± 168.5	<0.001	96.9 ± 58.6	142.3 ± 95.4	<0.001
Total cholesterol (mg/dL)	192.4 ± 33.8	197.6 ± 38.1	0.001	197.1 ± 35.2	207.3 ± 36.0	<0.001
HDL-C (mg/dL)	48.8 ± 11.1	43.8 ± 8.9	<0.001	58.5 ± 13.0	52.4 ± 11.6	<0.001
LDL-C (mg/dL)	123.0 ± 30.8	126.3 ± 34.2	0.023	121.0 ± 31.3	130.9 ± 33.1	<0.001
eGFR (mL/min/1.73 m^2)	99.2 ± 19.8	96.9 ± 20.6	0.018	114.9 ± 25.7	112.9 ± 24.1	0.041
Uric acid (mg/dL)	6.5 ± 1.3	6.9 ± 1.5	<0.001	4.9 ± 1.1	5.5 ± 1.1	<0.001
Obesity-related indices						
BMI (kg/m^2)	24.8 ± 3.1	26.7 ± 3.6	<0.001	23.2 ± 3.4	25.7 ± 3.7	<0.001
WHR	0.89 ± 0.05	0.93 ± 0.05	<0.001	0.84 ± 0.07	0.89 ± 0.06	<0.001
WHtR	0.51 ± 0.05	0.55 ± 0.05	<0.001	0.51 ± 0.06	0.56 ± 0.06	< 0.001
BRI	7.0 ± 1.6	8.0 ± 1.8	<0.001	6.3 ± 1.8	7.7 ± 2.0	< 0.001
CI	1.23 ± 0.06	1.26 ± 0.06	<0.001	1.22 ± 0.08	1.26 ± 0.08	< 0.001
BAI	26.1 ± 3.0	27.5 ± 3.2	<0.001	30.2 ± 3.7	32.0 ± 3.9	< 0.001
AVI	15.2 ± 2.9	17.0 ± 3.4	<0.001	13.1 ± 2.9	15.2 ± 3.3	< 0.001
LAP	33.0 ± 31.4	56.4 ± 53.2	<0.001	25.7 ± 22.4	46.3 ± 34.0	< 0.001
VAI	1.6 ± 1.5	2.6 ± 3.2	<0.001	1.5 ± 1.3	2.6 ± 2.4	< 0.001

DM, diabetes mellitus; BP, blood pressure; HbA1c, glycosylated hemoglobin A1c; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; eGFR, estimated glomerular filtration rate; BMI, body mass index; WHR, waist–hip ratio; WHtR, waist-to-height ratio; BRI, body roundness index; CI, conicity index; BAI, body adiposity index; AVI, abdominal volume index; LAP, lipid accumulation product; VAI, visceral adiposity index.

Discussion

In this study, we investigated sex differences in the associations between nine obesity-related indices with incidence of DM after a median 4-year follow-up period. We found that all of the studied obesity indices were associated with incidence of DM in both the male and female participants. Furthermore, there were sex differences in the relationships between some of the obesity indices and incidence of DM. In the men, WHR and CI were more strongly correlated with incidence of DM than in the women, whereas LAP and VAI were more closely linked with incidence of DM in the women than in the men.

Our results demonstrate the predictive ability of obesity-related indices for incidence of DM. In the male participants, the strongest predictors for incidence of DM were LAP, WHtR, and WHR. In the female participants, the strongest predictors were LAP, WHtR, and

TABLE 3 Association of obesity-related indices with incidence of DM using multivariable logistic regression analysis.

Obesity-related indices	Male (n = 8,334)			Female (n = 16,012)		
	Multivariable			Multivariable		
	OR	95% confidence interval	p	OR	95% confidence interval	p
BMI (per 1 kg/m^2)[a]	1.140	1.105–1.175	<0.001	1.099	1.073–1.125	<0.001
WHR (per 0.01)[a]	1.082	1.061–1.104	<0.001	1.051	1.037–1.065	<0.001
WHtR (per 0.01)[a]	1.088	1.066–1.110	<0.001	1.056	1.041–1.070	<0.001
BRI (per 1)[a]	1.263	1.193–1.337	<0.001	1.171	1.123–1.221	<0.001
CI (per 0.1)[a]	1.425	1.208–1.682	<0.001	1.233	1.118–1.359	<0.001
BAI (per 1)[a]	1.082	1.048–1.117	<0.001	1.043	1.022–1.066	<0.001
AVI (per 1)[a]	1.124	1.089–1.159	<0.001	1.095	1.068–1.123	<0.001
LAP (log per 1)[b]	8.951	5.454–14.689	<0.001	8.687	5.782–13.052	<0.001
VAI (log per 1)[c]	9.104	6.141–13.497	<0.001	6.629	4.864–9.035	<0.001

Values expressed as odds ratio (OR) and 95% confidence interval. DM, diabetes mellitus; BMI, body mass index; WHR, waist–hip ratio; WHtR, waist-to-height ratio; BRI, body roundness index; CI, conicity index; BAI, body adiposity index; AVI, abdominal volume index; LAP, lipid accumulation product; VAI, visceral adiposity index. [a]Covariates in the multivariable model included age, hypertension, smoking and alcohol history, systolic and diastolic BPs, hemoglobin, triglyceride, total cholesterol, HDL-C, LDL-C, eGFR and uric acid in male (significant variables in Table 2); age, hypertension, menstruation status, systolic and diastolic BPs, hemoglobin, triglyceride, total cholesterol, HDL-C, LDL-C, eGFR and uric acid in female (significant variables in Table 2).
[b]Covariates as [a]Covariates, except for triglyceride.
[c]Covariates as [a]Covariates, except for triglyceride and HDL-C.

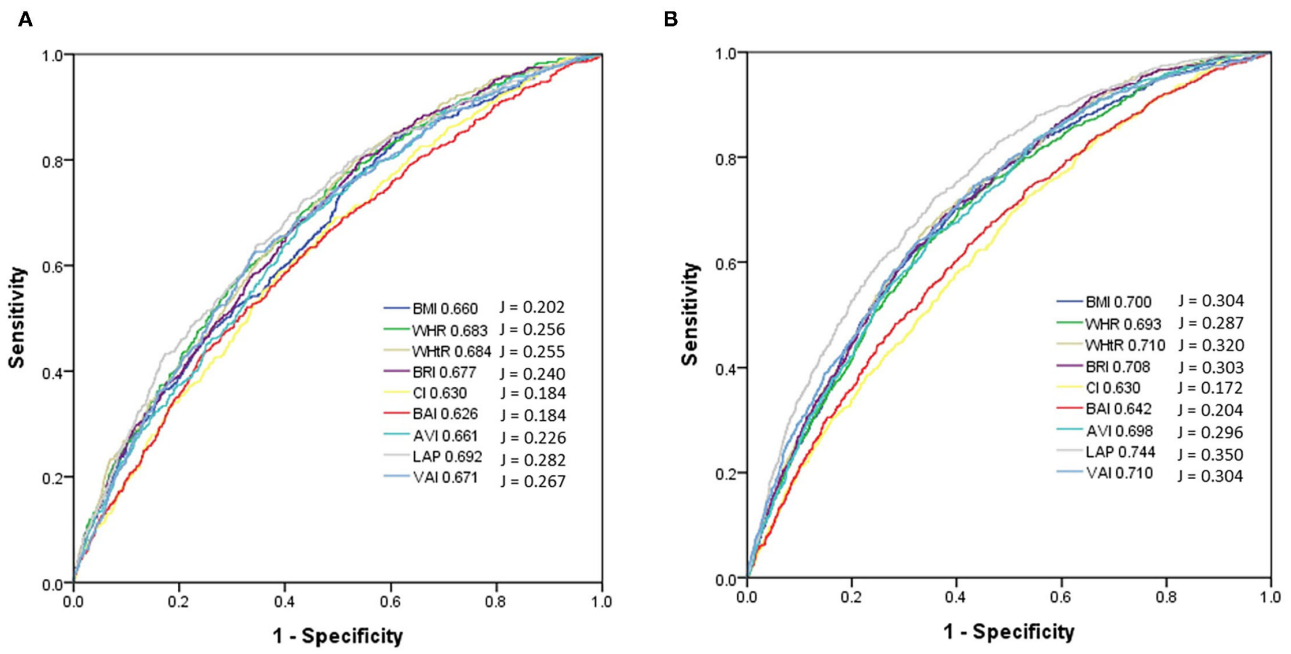

FIGURE 2
Comparison of the AUC and J values of 9 obesity-related indices for diagnosis of incidence of diabetes mellitus among **(A)** males and **(B)** females. In the male participants, LAP had the highest AUC (0.692) and J (0.282), followed by WHtR (AUC = 0.684, J = 0.255), WHR (AUC = 0.683, J = 0.256), BRI (AUC = 0.677, J = 0.240), VAI (AUC = 0.671, J = 0.267), AVI (AUC = 0.661, J = 0.226), BMI (AUC = 0.660, J = 0.202), CI (AUC = 0.630, J = 0.184), and BAI (AUC = 0.626, J = 0.184). In the female participants, LAP also had the highest AUC (0.744) and J (0.350), followed by WHtR (AUC = 0.710, J = 0.320), VAI (AUC = 0.710, J = 0.304), BRI (AUC = 0.708, J = 0.303), BMI (AUC = 0.700, J = 0.304), AVI (AUC = 0.698, J = 0.296), WHR (AUC = 0.693, J = 0.287), BAI (AUC = 0.642, J = 0.204), and CI (AUC = 0.630, J = 0.172).

VAI, followed by BRI. Previous epidemiologic studies have reported an association between central obesity and the incidence of DM (16–18, 33). Many obesity-related indices can be used to assess the degree of central obesity, of which LAP had the highest predictive ability for incidence of DM in both the male and female participants in the present study. In a Japanese study of 10,170 middle-aged individuals, a high LAP was associated with DM, with an OR of 19.09 (95% confidence interval: 6.57–55.50) in women and 7.40 (95% confidence interval: 5.10–10.75) in men (18). A 6-year cohort study conducted by Bozorgmanesh et al. with a total of 5,018 non-diabetic subjects also found that LAP was a strong predictor of incidence of DM in both sexes (34). LAP has two important components, namely WC and serum TG level, which reflect abdominal fat and lipid metabolism (35). Increased visceral fat and serum TG levels were linked with

TABLE 4 Area under curve (AUC), cutoff value, sensitivity, specificity, and Youden index of 9 obesity-related indices for incidence of DM in male participants.

Obesity-related indices	AUC (95% confidence interval)	Cutoff value	Sensitivity (%)	Specificity (%)	Youden index
BMI (kg/m^2)	0.660 (0.635–0.685)*	25.287	60.1	60.0	0.202
WHR	0.683 (0.659–0.707)*	0.908	62.8	62.8	0.256
WHtR	0.684 (0.661–0.708)*	0.526	62.8	62.7	0.255
BRI	0.677 (0.653–0.701)*	7.312	62.0	62.0	0.240
CI	0.630 (0.605–0.655)*	1.242	59.2	59.2	0.184
BAI	0.626 (0.600–0.652)*	26.676	59.2	59.2	0.184
AVI	0.661 (0.637–0.685)*	15.712	61.3	61.3	0.226
LAP	0.692 (0.668–0.716) *	32.649	64.1	64.1	0.282
VAI	0.671 (0.647–0.696)*	1.549	63.4	63.3	0.267

*$p < 0.001$. DM, diabetes mellitus; BMI, body mass index; WHR, waist–hip ratio; WHtR, waist-to-height ratio; BRI, body roundness index; CI, conicity index; BAI, body adiposity index; AVI, abdominal volume index; LAP, lipid accumulation product; VAI, visceral adiposity index.

TABLE 5 Area under curve (AUC), cutoff value, sensitivity, specificity, and Youden index of 9 obesity-related indices for incidence of DM in female participants.

Obesity-related indices	AUC (95% confidence interval)	Cutoff value	Sensitivity (%)	Specificity (%)	Youden index
BMI (kg/m^2)	0.700 (0.680–0.720)*	24.008	65.2	65.2	0.304
WHR	0.693 (0.673–0.713)*	0.864	64.3	64.4	0.287
WHtR	0.710 (0.691–0.729)*	0.529	66.0	66.0	0.320
BRI	0.708 (0.689–0.727)*	6.753	65.2	65.1	0.303
CI	0.630 (0.609–0.651)*	1.229	58.6	58.6	0.172
BAI	0.642 (0.620–0.663)*	30.689	60.2	60.2	0.204
AVI	0.698 (0.679–0.717)*	13.792	64.8	64.8	0.296
LAP	0.744 (0.725–0.762)*	27.819	67.5	67.5	0.350
VAI	0.710 (0.690–0.729)*	1.495	65.2	65.2	0.304

*$p < 0.001$. Abbreviations are the same as in Table 1. DM, diabetes mellitus; BMI, body mass index; WHR, waist–hip ratio; WHtR, waist-to-height ratio; BRI, body roundness index; CI, conicity index; BAI, body adiposity index; AVI, abdominal volume index; LAP, lipid accumulation product; VAI, visceral adiposity index.

insulin resistance in a Japanese study of metabolically obese subjects with normal weight and glucose tolerance (36). The association between visceral fat and DM could be because adipocytokines are released by visceral fat rather than subcutaneous adipose tissue, and their circulating levels are associated with the development of insulin resistance (36, 37). Growing evidence supports that WHtR has a significant relationship with new-onset DM (38–41), and a meta-analysis found that WHtR had a higher predictive power for the risk of DM than WC and BMI (38). This could be because BMI only measures total BW and cannot discriminate between visceral adipose and skeletal muscle mass (40), while WC does not take tall and short body shapes into account. Other research has revealed an important and independent association between WHR and DM (17, 42, 43). However, the use of WHR is controversial due to its uncertain biologic interpretation, lack of sensitivity to weight gain, and high variability across age, sex, and different populations (17). Many studies have shown that BRI can be used to predict visceral fat (27), metabolic syndrome (44) and DM (19, 45). In a prospective cohort study, BRI and WHtR were shown to have a similar predictive ability for DM in hypertensive patients, possibly because both BRI and WHtR are calculated based on WC and height (46). Another useful predictor of DM is VAI, which was first described by Amato et al. as visceral adipose dysfunction associated with cardiometabolic risk in a healthy population (47). Previous studies have reported that VAI may predict DM (20, 48), and that it is an indicator of adipose tissue dysfunction (49). In addition, other studies have reported a positive correlation between VAI and adipocytokines, and this may explain the pathogenesis of developing insulin resistance (50, 51).

We also found sex differences in the predictive ability of the obesity-related indices for incidence of DM. Previous studies have demonstrated sex differences in fat distribution, and the type of fat deposition has been shown to have a varied effect on cardiometabolic risk (52–54). Many mechanisms, including fat microenvironment, cell-intrinsic features, and sex hormones have been postulated to explain the sex dimorphism in adipose distribution (52). In men, fat deposits are more likely to be centrally distributed or apple-shaped, which is correlated with an increased risk of cardiometabolic disease. In women, fat deposits are prone to peripheral fat distribution or pear-shaped, which may protect against cardiometabolic disease (52, 54). Prior studies have reported sex differences in lipid profiles. Compared to men, premenopausal women have higher levels of serum HDL, and lower levels of LDL, very-low-density lipoprotein and TGs, which may lead to a protective effect against cardiovascular disease by estrogen (55). On the other hand, excess visceral fat has

been closely correlated with the impaired inhibition of free fatty acid release in response to insulin, along with hypertriglyceridemia and low HDL-C concentrations (56). Because women have a tendency to accumulate peripheral fat, elevated serum TGs along with reduced HDL-C, but not WC, may be better able to identify visceral adipocyte expansion in women. LAP is proportional to TG concentration and VAI is dependent on TG and HDL-C levels. Both can indicate the presence of extra lipid fuel and ectopic fat deposits, which increase the risk of metabolic disease. Our results suggest that abnormal lipid metabolism increases the risk of developing DM in women. In summary, this study suggested WHR and CI are more specific to men for predicting the development of type 2 DM, while LAP and VAI are more specific to women.

The strengths of this study include the large population-based investigation and follow-up of sex differences in the associations between nine obesity-related indices and incidence of DM. However, there were also some limitations. First, data on medications and other factors which could affect the findings of incidence of DM such as anti-diabetics, anti-hypertensives, lipid-lowering agents and proteinuria are not recorded in the TWB. This may have led to underestimation of the association between incidence of DM and the obesity-related indices. Second, this study was conducted in Taiwan with participants of Chinese ethnicity, and this may limit the generalizability of our findings. Third, as only approximately 25% of the enrollees in the TWB undergo follow-up evaluations, sample bias is possible.

In conclusion, we found that all nine studied obesity-related indices were significant predictors of incidence of DM, and we also found differences in the associations between the male and female participants. Hence, to better control DM, reducing body weight may be beneficial in addition to lifestyle modifications, diet control, and pharmacological interventions.

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 Institutional Review Board of Kaohsiung Medical University Hospital [KMUHIRB-E(I)-20210058]. The patients/participants provided their written informed consent to participate in this study.

Author contributions

Conceptualization, methodology, validation, formal analysis, writing—review and editing, supervision, software, investigation, resources, project administration, funding acquisition, and visualization: S-CC. Data curation: T-LC, Y-HL, P-YW, J-CH, and S-CC. Writing—original draft preparation: T-LC and S-CC. All authors have read and agreed to the published version of the manuscript.

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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.

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References

1. Sun H, Saeedi P, Karuranga S, Pinkepank M, Ogurtsova K, Duncan BB, et al. regional and country-level diabetes prevalence estimates for 2021 and projections for 2045. *Diabetes Res Clin Pract.* (2022) 183:109119. doi: 10.1016/j.diabres.2021.109119

2. Taiwan Health Promotion Administration. *Survey on the Prevalence of Hypertension, Hyperglycemia and Hyperlipidemia in Taiwan* (2021).

3. Stumvoll M, Goldstein BJ, van Haeften TW. Type 2 diabetes: principles of pathogenesis and therapy. *Lancet.* (2005) 365:1333–46. doi: 10.1016/S0140-6736(05)61032-X

4. Zheng Y, Ley SH, Hu FB. Global aetiology and epidemiology of type 2 diabetes mellitus and its complications. *Nat Rev Endocrinol.* (2018) 14:88–98. doi: 10.1038/nrendo.2017.151

5. Ministry of Health and Welfare. *Cause of Death Statistics, Taiwan* (2022).

6. Wu L, Zhu W, Qiao Q, Huang L, Li Y, Chen L. Novel and traditional anthropometric indices for identifying metabolic syndrome in non-overweight/obese adults. *Nutr Metab (Lond).* (2021) 18:3. doi: 10.1186/s12986-020-00536-x

7. Jao HF, Wung CH, Yu HC, Lee MY, Chen PC, Chen SC, et al. Sex difference in the associations among obesity-related indices with metabolic syndrome in patients with type 2 diabetes mellitus. *Int J Med Sci.* (2021) 18:3470–7. doi: 10.7150/ijms.63180

8. Kahn SE, Hull RL, Utzschneider KM. Mechanisms linking obesity to insulin resistance and type 2 diabetes. *Nature.* (2006) 444:840–6. doi: 10.1038/nature05482

9. Lin IT, Lee MY, Wang CW, Wu DW, Chen SC. Gender differences in the relationships among metabolic syndrome and various obesity-related indices with nonalcoholic fatty liver disease in a taiwanese population. *Int J Environ Res Public Health.* (2021) 18:857. doi: 10.3390/ijerph18030857

10. Ou YL, Lee MY, Lin IT, Wen WL, Hsu WH, Chen SC. Obesity-related indices are associated with albuminuria and advanced kidney disease in type 2 diabetes mellitus. *Ren Fail.* (2021) 43:1250–8. doi: 10.1080/0886022X.2021.1969247

11. Hsu YE, Chen SC, Geng JH, Wu DW, Wu PY, Huang JC. Obesity-related indices are associated with longitudinal changes in lung function: a large taiwanese population follow-up study. *Nutrients.* (2021) 13:4055. doi: 10.3390/nu13114055

12. Wung CH, Chung CY, Wu PY, Huang JC, Tsai YC, Chen SC, et al. Associations between metabolic syndrome and obesity-related indices and bone mineral density t-score in hemodialysis patients. *J Pers Med.* (2021) 11:775. doi: 10.3390/jpm11080775

13. Lee WC, Wu PY, Huang JC, Tsai YC, Chiu YW, Chen SC, et al. Sex difference in the associations among obesity-related indices with incident hypertension in a large taiwanese population follow-up study. *J Pers Med.* (2022) 12:972. doi: 10.3390/jpm12060972

14. Wung CH, Lee MY, Wu PY, Huang JC, Chen SC. Obesity-related indices are associated with peripheral artery occlusive disease in patients with type 2 diabetes mellitus. *J Pers Med.* (2021) 11:533. doi: 10.3390/jpm11060533

15. Huang SH, Chen SC, Geng JH, Wu DW, Li CH. Metabolic syndrome and high-obesity-related indices are associated with poor cognitive function in a large taiwanese population study older than 60 years. *Nutrients.* (2022) 14:1535. doi: 10.3390/nu14081535

16. Freemantle N, Holmes J, Hockey A, Kumar S. How strong is the association between abdominal obesity and the incidence of type 2 diabetes? *Int J Clin Pract.* (2008) 62:1391–6. doi: 10.1111/j.1742-1241.2008.01805.x

17. Vazquez G, Duval S, Jacobs DR, Silventoinen K. Comparison of body mass index, waist circumference, and waist/hip ratio in predicting incident diabetes: a meta-analysis. *Epidemiol Rev.* (2007) 29:115–28. doi: 10.1093/epirev/mxm008

18. Wakabayashi I, Daimon T. A strong association between lipid accumulation product and diabetes mellitus in japanese women and men. *J Atheroscler Thromb.* (2014) 21:282–8. doi: 10.5551/jat.20628

19. Chang Y, Guo X, Chen Y, Guo L, Li Z, Yu S, et al. A body shape index and body roundness index: two new body indices to identify diabetes mellitus among rural populations in northeast China. *BMC Public Health.* (2015) 15:794. doi: 10.1186/s12889-015-2150-2

20. Ahn N, Baumeister SE, Amann U, Rathmann W, Peters A, Huth C, et al. Visceral adiposity index (VAI), lipid accumulation product (LAP), and product of triglycerides and glucose (TyG) to discriminate prediabetes and diabetes. *Sci Rep.* (2019) 9:9693. doi: 10.1038/s41598-019-46187-8

21. Chen CH, Yang JH, Chiang CWK, Hsiung CN, Wu PE, Chang LC, et al. Population structure of Han Chinese in the modern Taiwanese population based on 10,000 participants in the Taiwan Biobank project. *Hum Mol Genet.* (2016) 25:5321–31. doi: 10.1093/hmg/ddw346

22. Fan CT, Hung TH, Yeh CK. Taiwan regulation of biobanks. *J Law Med Ethics.* (2015) 43:816–26. doi: 10.1111/jlme.12322

23. Levey AS, Bosch JP, Lewis JB, Greene T, Rogers N, Roth D. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. Modification of diet in renal disease study group. *Ann Internal Med.* (1999) 130:461–70. doi: 10.7326/0003-4819-130-6-199903160-00002

24. Vickery S, Stevens PE, Dalton RN, van Lente F, Lamb EJ. Does the ID-MS traceable MDRD equation work and is it suitable for use with compensated Jaffe and enzymatic creatinine assays? *Nephrol Dial Transplant.* (2006) 21:2439–45. doi: 10.1093/ndt/gfl249

25. Ministry of Education. *Physical Fitness 333 Plan, Taiwan* (1999).

26. American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care.* (2010) 33:S62–9. doi: 10.2337/dc10-S062

27. Thomas DM, Bredlau C, Bosy-Westphal A, Mueller M, Shen W, Gallagher D, et al. Relationships between body roundness with body fat and visceral adipose tissue emerging from a new geometrical model. *Obesity (Silver Spring).* (2013) 21:2264–71. doi: 10.1002/oby.20408

28. Valdez R. A simple model-based index of abdominal adiposity. *J Clin Epidemiol.* (1991) 44:955–6. doi: 10.1016/0895-4356(91)90059-I

29. Bergman RN, Stefanovski D, Buchanan TA, Sumner AE, Reynolds JC, Sebring NG, et al. A better index of body adiposity. *Obesity (Silver Spring).* (2011) 19:1083–9. doi: 10.1038/oby.2011.38

30. Guerrero-Romero F, Rodriguez-Morán M. Abdominal volume index. An anthropometry-based index for estimation of obesity is strongly related to impaired glucose tolerance and type 2 diabetes mellitus. *Arch Med Res.* (2003) 34:428–32. doi: 10.1016/S0188-4409(03)00073-0

31. H.S. Kahn. The "lipid accumulation product" performs better than the body mass index for recognizing cardiovascular risk: a population-based comparison. *BMC Cardiovasc Disord.* (2005) 5:26. doi: 10.1186/1471-2261-5-26

32. Amato MC, Giordano C, Galia M, Criscimanna A, Vitabile S, Midiri M, et al. Visceral Adiposity Index: a reliable indicator of visceral fat function associated with cardiometabolic risk. *Diabetes Care.* (2010) 33:920–2. doi: 10.2337/dc09-1825

33. Cao C, Hu H, Zheng X, Zhang X, Wang Y, He Y. Association between central obesity and incident diabetes mellitus among Japanese: a retrospective cohort study using propensity score matching. *Sci Rep.* (2022) 12:13445. doi: 10.1038/s41598-022-17837-1

34. Bozorgmanesh M, Hadaegh F, Azizi F. Diabetes prediction, lipid accumulation product, and adiposity measures; 6-year follow-up: Tehran lipid and glucose study. *Lipids Health Dis.* (2010) 9:45. doi: 10.1186/1476-511X-9-45

35. Mirmiran P, Bahadoran Z, Azizi F. Lipid accumulation product is associated with insulin resistance, lipid peroxidation, and systemic inflammation in type 2 diabetic patients. *Endocrinol Metab (Seoul).* (2014) 29:443–9. doi: 10.3803/EnM.2014.29.4.443

36. Katsuki A, Sumida Y, Urakawa H, Gabazza EC, Murashima S, Maruyama N, et al. Increased visceral fat and serum levels of triglyceride are associated with insulin resistance in Japanese metabolically obese, normal weight subjects with normal glucose tolerance. *Diabetes Care.* (2003) 26:2341–4. doi: 10.2337/diacare.26.8.2341

37. Lebovitz HE, Banerji MA. Point: visceral adiposity is causally related to insulin resistance. *Diab Care.* (2005) 28:2322–5. doi: 10.2337/diacare.28.9.2322

38. Kodama S, Horikawa C, Fujihara K, Heianza Y, Hirasawa R, Yachi Y, et al. Comparisons of the strength of associations with future type 2 diabetes risk among anthropometric obesity indicators, including waist-to-height ratio: a meta-analysis. *Am J Epidemiol.* (2012) 176:959–69. doi: 10.1093/aje/kws172

39. Jayawardana R, Ranasinghe P, Sheriff MH, Matthews DR, Katulanda P. Waist to height ratio: a better anthropometric marker of diabetes and cardio-metabolic risks in South Asian adults. *Diab Res Clin Pract.* (2013) 99:292–9. doi: 10.1016/j.diabres.2012.12.013

40. Mirzaei M, Khajeh M. Comparison of anthropometric indices (body mass index, waist circumference, waist to hip ratio and waist to height ratio) in predicting risk of type II diabetes in the population of Yazd, Iran. *Diabetes Metab Syndr.* (2018) 12:677–82. doi: 10.1016/j.dsx.2018.04.026

41. Xu Z, Qi X, Dahl AK, Xu W. Waist-to-height ratio is the best indicator for undiagnosed type 2 diabetes. *Diabet Med.* (2013) 30:e201–7. doi: 10.1111/dme.12168

42. Schmidt MI, Duncan BB, Canani LH, Karohl C, Chambless L. Association of waist-hip ratio with diabetes mellitus. Strength and possible modifiers. *Diab Care.* (1992) 15:912–4. doi: 10.2337/diacare.15.7.912

43. Qiao Q, Nyamdorj R. Is the association of type II diabetes with waist circumference or waist-to-hip ratio stronger than that with body mass index? *Eur J Clin Nutr.* (2010) 64:30–4. doi: 10.1038/ejcn.2009.93

44. Liu B, Liu B, Wu G, Yin F. Relationship between body-roundness index and metabolic syndrome in type 2 diabetes. *Diabetes Metab Syndr Obes.* (2019) 12:931–5. doi: 10.2147/DMSO.S209964

45. Zhao Q, Zhang K, Li Y, Zhen Q, Shi J, Yu Y, et al. Capacity of a body shape index and body roundness index to identify diabetes mellitus in Han Chinese people in Northeast China: a cross-sectional study. *Diabet Med.* (2018) 35:1580–7. doi: 10.1111/dme.13787

46. Liu Y, Liu X, Guan H, Zhang S, Zhu Q, Fu X, et al. Body roundness index is a superior obesity index in predicting diabetes risk among hypertensive patients: a prospective cohort study in China. *Front Cardiovasc Med.* (2021) 8:736073. doi: 10.3389/fcvm.2021.736073

47. Amato MC, Giordano C, Pitrone M, Galluzzo A. Cut-off points of the visceral adiposity index (VAI) identifying a visceral adipose dysfunction associated with cardiometabolic risk in a Caucasian Sicilian population. *Lipids Health Dis.* (2011) 10:183. doi: 10.1186/1476-511X-10-183

48. Bozorgmanesh M, Hadaegh F, Azizi F. Predictive performance of the visceral adiposity index for a visceral adiposity-related risk: type 2 diabetes. *Lipids Health Dis.* (2011) 10:88. doi: 10.1186/1476-511X-10-88

49. Amato MC, Giordano C. Visceral adiposity index: an indicator of adipose tissue dysfunction. *Int J Endocrinol.* (2014) 2014:730827. doi: 10.1155/2014/730827

50. Ciresi A, Amato MC, Pizzolanti G, Giordano Galluzzo C. Visceral adiposity index is associated with insulin sensitivity and adipocytokine levels in newly diagnosed acromegalic patients. *J Clin Endocrinol Metab.* (2012) 97:2907–15. doi: 10.1210/jc.2012-1518

51. Al-Daghri NM, Al-Attas OS, Alokail MS, Alkharfy KM, Charalampidis P, Livadas S, et al. Visceral adiposity index is highly associated with adiponectin values and glycaemic disturbances. *Eur J Clin Invest.* (2013) 43:183–9. doi: 10.1111/eci.12030

52. Lumish HS, O'Reilly M, Reilly MP. Sex differences in genomic drivers of adipose distribution and related cardiometabolic disorders: opportunities for precision medicine. *Arterioscler Thromb Vasc Biol.* (2020) 40:45–60. doi: 10.1161/ATVBAHA.119.313154

53. Chang E, Varghese M, Singer K. Gender and sex differences in adipose tissue. *Curr Diab Rep.* (2018) 18:69. doi: 10.1007/s11892-018-1031-3

54. Schorr M, Dichtel LE, Gerweck AV, Valera RD, Torriani M, Miller KK, et al. Sex differences in body composition and association with cardiometabolic risk. *Biol Sex Differ.* (2018) 9:28. doi: 10.1186/s13293-018-0189-3

55. Wang X, Magkos F, Mittendorfer B. Sex differences in lipid and lipoprotein metabolism: it's not just about sex hormones. *J Clin Endocrinol Metab.* (2011) 96:885–93. doi: 10.1210/jc.2010-2061

56. Ebbert JO, Jensen MD. Fat depots, free fatty acids, and dyslipidemia. *Nutrients.* (2013) 5:498–508. doi: 10.3390/nu5020498

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Sociodemographic and behavioural risk factors associated with low awareness of diabetes mellitus medication in Indonesia: Findings from the Indonesian Family Life Survey (IFLS-5)

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Introduction: Low awareness of the necessity of taking medication is common among patients with diabetes mellitus (DM) due to their lack of understanding of the disease. Therefore, it is essential to determine the underlying risks influencing low awareness to design effective intervention strategies. This study aims to evaluate the association of sociodemographic and behavioural factors with low awareness to take medication among patients with DM in Indonesia.

Method: Retrospective data were obtained from the Indonesian Family Life Survey (IFLS-5), a national cross-sectional population-based survey among respondents with DM aged ≥15 years. DM status was confirmed by HbA1c testing, while sociodemographic and other health-related information was obtained from self-reported data. Gender, age, educational level, marital status, economic status, comorbidity, religiosity, residence and health insurance status were considered sociodemographic, whereas blood glucose monitoring status, sleeping problems, depression status, having a general medical check-up, satisfaction with healthcare needs and happiness status were considered behavioural risk factors. Awareness of DM medication was determined by self-reported responses to the question asked by the surveyor. Logistic regression analysis was used to evaluate the association between sociodemographic and behavioural factors and low awareness of DM medication. Odds ratios (ORs) with 95% confidence intervals (CIs) were reported.

Result: Most of the 706 respondents were female (58.8%) and aged 55–65 years (28.8%). Most of them showed low awareness of diabetes medication (87.7%). Irregular blood glucose monitoring (OR: 23.61, 95% CI 11.46–48.65; $p < 0.001$), without any comorbidity (OR: 2.03, 95% CI 1.05–3.90; $p = 0.034$), never had any general medical check-up (OR: 2.52, 95% CI 1.12–5.36; $p = 0.016$), 26–35 years of age (OR: 4.96, 95% CI 1.06–23.19; $p = 0.042$), 36–45 years of age (OR: 5.04, 95% CI 1.17–21.69; $p = 0.030$) and having no health insurance coverage (OR: 2.08, 95% CI 1.12–3.87; $p = 0.021$) were significantly associated with low awareness of diabetes medication.

Conclusion: Healthcare professionals should regularly evaluate blood glucose level, perform routine medical check-ups, prioritise patient satisfaction by providing appropriate care, involve patients in decision-making by determining their needs and then tailor an intervention to meet the need for, and improve their awareness of, DM medication.

KEYWORDS

awareness medication, diabetes mellitus, determinants, IFLS, Indonesia

1. Introduction

Diabetes mellitus (DM) has increased drastically in the past three decades; around 422 million people worldwide have diabetes, with deaths totalling 1.5 million (1). The number of patients with DM in Southeast Asia is predicted to increase by 113 million by 2030 (2). DM ranks second among the most common non-communicable diseases in Indonesia, with a prevalence of 10.9% (3). In addition, more than half of the individuals with DM in Indonesia (73.7%) were unaware of their condition (4). Therefore, if DM is left unmanaged and untreated, it could lead to either microvascular (retinopathy, nephropathy and neuropathy) or macrovascular (stroke, cardiovascular disease and peripheral artery disease) complications (5). Unfortunately, no complete cure for diabetes has been found; thus, long-term treatment to prevent or delay complications and maintain the patient's quality of life is needed (6).

Healthcare professionals have a crucial role in developing strategies to facilitate medication adherence so that patients can optimise diabetes treatment and limit the progression of diabetes (6). This emphasises the significance of patient education and awareness of DM medication after having acquired awareness of DM (7). Thus, the first approach for healthcare practitioners could be to increase DM medication awareness. Awareness of medication was defined as a patient's common knowledge or understanding about his/her medication without direct instruction or as a sort of medication self-consciousness (8). Awareness is one of the five primary types of related concerns with the potential to improve therapy since they are obstacles to medication adherence from the patient's perspective (9). It is somewhat distinct from medication adherence, defined as the process whereby patients take their medication as prescribed (10). Inadequate medication adherence among DM patients remains a major problem leading to disease progression (11). At least 45% of treatment failures among DM patients are caused by low adherence to anti-diabetic medical treatments (12). This could lead to increases in health expenses yearly at both patient and societal levels (13).

Patients with good comprehension and high awareness of diabetic medication exhibited an improvement in their glucose control (14) and medication adherence (15). Therefore, identifying factors associated with medication awareness is an important first step to improving medication adherence. Although studies about medication awareness in patients with diabetes are limited, some studies have explored a positive correlation between the frequency of healthcare professional counselling and the patient's level of awareness to take their medication (16, 17). The Indonesian Family Life Survey (IFLS) is a longitudinal socioeconomic and health survey in Indonesia, which covers ~83% of the Indonesian population (18). Previous studies that employed IFLS-4 or IFLS-5 mostly examined the prevalence of DM and its sociodemographic risk factors (19–22). Other research evaluated the association between socioeconomic characteristics and the incidence of diabetes (23, 24). Until now, no study has investigated psychosocial and sociodemographic factors associated with medication awareness among DM patients using IFLS. It is still unclear as to which components or focal points are necessary to increase DM medication awareness in Indonesia. Therefore, addressing its fundamental causes is vital for developing effective intervention strategies. This study aims to identify sociodemographic and behavioural factors associated with low awareness of DM medication in Indonesia.

2. Methods

2.1. Study design

The cross-sectional study design utilised in this study is based on secondary data, IFLS-5. The IFLS is a longitudinal study that used a multistage stratified sample design to represent 83% of the Indonesian population (18). The IFLS sampling strategy stratified on provinces and urban/rural locations and then sampled randomly within these strata. Provinces were chosen to optimise population representation, represent Indonesia's cultural and socioeconomic variety and be cost-effective to survey, given the country's size and geography. Therefore, 13 of the 27 provinces that existed at the time were included. The IFLS randomly selected 321 enumeration areas (EA) within each of the 13 provinces, oversampling urban EAs and EAs in smaller provinces to facilitate urban–rural and Javanese–non-Javanese comparisons. Twenty households from each urban EA and 30 households from each rural EA were selected (18). The IFLS gathered sociodemographic, economic, and health status characteristics, including self-reported health status, symptoms and pain assessments, as well as biomarker assessments. Preliminary testing of the IFLS questionnaire ensured its reliability and validity before the full-scale survey was conducted (18). The ethical review boards of the RAND's Human Subjects Protection Committee (s0064-06-01-CR01) approved IFLS research. Before data collection, every respondent provided written consent (18). Approval was sought from the research ethics committee of Universitas Padjadjaran, Indonesia, which waived the requirement because this study uses anonymous data from the IFLS.

2.2. Study population

Data were collected from IFLS-5 from individuals aged at least 15 years after the survey. Individuals with available data on HbA1c and medication for chronic diseases were included.

2.3. Outcome measure

Patients with DM were defined as having an HbA1c value of ≥6.5% (25). The blood samples examined were obtained from dried blood spots (DBSs) and taken through the capillaries at the fingertips (18). These blood samples are easier to obtain than those taken through intravenous vessels and are more durable in terms of storage (26). However, the results of the HbA1c examination from DBS will first be converted to whole blood HbA1c, the gold standard of HbA1c examination, so that it can be used in the diagnosis of DM (18).

Awareness of DM medication was determined by the responses to the following question posed by the surveyor: *Are you currently taking prescription medication weekly to manage your DM?* Those who responded with a "yes" were considered to have a good awareness of DM medication, whereas those who responded with a "no" were

thought to have low awareness. This questionnaire has been validated and involved extensive pretests and analysis of the pretest data (18).

2.4. Potential factors associated with low awareness of DM medication

Sociodemographic and behavioural risk factors were analysed as potential contributors to low awareness of DM medication. Age, gender, educational level, marital status, residency, economic status, health insurance coverage, religiosity and comorbidity status were categorised into sociodemographic and behavioural factors. At the same time, behavioural factors included blood glucose monitoring status, general medical check-ups, health care satisfaction, happiness status, insomnia and depressive symptom status.

Sociodemographic factors were age after the survey, gender (male/female), level of education (no education, elementary school, junior high school, senior high school and university), marital status (currently married and currently unmarried), residency (urban and rural) and health insurance coverage (yes and no). To assess the economic status, we divided the annual household income in rupiah by family size during the previous 12 months (per capita income). Capita income was categorised per quintile. The quintile is categorised by sorting the per capita income from lowest to highest and then dividing them into five equal groups (the first quintile = an income $\leq$ \$77.01; the second quintile = \$77.01–\$256.70; the third quintile = \$256.70–\$483.55; the fourth quintile = \$483.55–\$924.7; the fifth quintile $\geq$ \$924.7). To evaluate religiosity, we asked the question *How religious are you?* The individuals who said they were extremely religious or religious were classified as religious, whereas those who said they were somewhat religious or not religious were classified as non-religious (27).

Depressive symptoms were assessed using a self-reported Centre for Epidemiologic Studies Depression (CES-D) scale (18). The CES-D consists of 10 items highly linked to the presence of depressive symptoms (28). Eight of the questions examine the negative symptoms of depression (e.g., *I felt fearful and lonely*), whereas the other two examined the positive symptoms (e.g., *I felt hopeful about the future)*. The respondents stated how frequently each item applied to them in the previous week using a four-point Likert-type scale (0 = rarely or never, 1 = some or little, 2 = moderately or much of the time, and 3 = frequently or almost always). After reversing the positive mood items, the total score is obtained by summing all elements. An individual with a total score of $\geq$10 is deemed to have depressive symptoms (29). The CES-D questionnaire was translated into Indonesian (forward translation) and then re-translated separately into English by two translators (back translation) (18).

Ten Patient-Recorded Outcomes Measurement Information System (PROMIS) questions were used to assess the severity of insomnia (18). Each measure of sleep quality and sleep impairment during the previous week was determined using a set of five items (30). Each item was rated using a five-point Likert-type scale (0 = never/not at all; 1 = a little bit; 2 = somewhat; 3 = quite a bit; 4= always/very much). Insomnia was defined as a total score of $\geq$21–40 (31). The PROMIS questionnaire was translated into Indonesian (forward translation) and then re-translated separately into English by two translators (back translation) (18).

The question *Has a doctor/paramedic/nurse/midwife ever told you that you have the following chronic ailments or diseases?* was used to evaluate whether the individuals had comorbid disease status, with the following potential responses: hypertension; DM; TB; asthma and other chronic lung diseases; cardiac disease (heart attack/coronary heart disease/angina or other heart diseases); liver disease; stroke; cancer or other malignancies; gout/uric acid; depression and vision and hearing abnormalities (27). Individuals who responded only with DM were classified as having no comorbidity. Individuals who responded with DM and one to three additional chronic diseases were classified as having 1–3 comorbidities, whereas individuals with four or more comorbidities were classified as having $\geq$4 comorbidities.

Individuals' response to the question *How regularly do you have your blood glucose checked?* determined the classification of blood glucose monitoring status as either regular or irregular. Individuals' response to *Have you had a general check-up in the recent 5 years?* as either yes or no determined one's status as having/not having had a general medical check-up. Happiness level was determined by asking *Overall, how would you describe the current state of these days? Would you say you are very happy, happy, unhappy or very unhappy?* (27). Those who responded with very happy or happy were categorised as happy, whereas those who responded with unhappy or very unhappy were categorised as unhappy. Satisfaction with healthcare was measured with the question *In relation to your healthcare, which of the following is true: it is less than adequate for my needs; it is just adequate for my needs or it is more than adequate for my needs?* (27). Low healthcare satisfaction corresponds to the response "it is just less than adequate for my needs," and high healthcare satisfaction corresponds to "it is just adequate for my needs" or "it is more than adequate for my needs."

2.5. Statistical analysis

Descriptive statistics were used to summarise the characteristics of the individuals. Awareness of DM medication was estimated for each age group and gender. The Little test was performed to assess whether the incomplete data type was classified as Missing Completely At Random (MCAR). The Little test assumes that the missing-ness of the data is independent of both observed and unobserved data, thus, a $p > 0.05$ is considered MCAR because it is asymptotically distributed under the null hypothesis that there are no differences between the observed and unobserved data (32). Since the missing data were MCAR ($p > 0.005$), complete case analyses could be carried out (32). A Chi-square test was conducted to evaluate the bivariate relationship between the individuals' characteristics and outcomes. The potential factors related to the outcome in the bivariate analysis at a significance threshold of $p < 0.25$ were included in the initial multivariate model. To determine the odds ratio (OR) with a 95% confidence interval (95% CI), multivariate logistic regression with manual backward elimination was used. The p-values for the factors included in the final model were all fixed to $p < 0.05$. The Hosmer–Lemeshow test was used to assess the goodness-of-fit statistic; R-squared is a number ranging from 0 to 1, indicating how much the combination of independent factors influences the value of the dependent variable at the same time (33). All statistical analyses were performed using Stata software version 14.0 for Windows.

3. Results

3.1. General characteristics of the study population

A total sample of 706 individuals without missing data on awareness of DM medication was included in this study. The majority of the respondents were female (58.8%) and aged 55–65 years (28.8%; Table 1). A high proportion of individuals had irregular blood glucose monitoring (90.1%), with most of them (88.4%) not having undergone a general medical check-up. Approximately half of the individuals were not covered by health insurance (50.4%) and had no comorbidities (50.1%). Out of the total respondents, 22.1% experienced depression, whereas 85.9% had insomnia.

3.2. Risk factor of low awareness of DM medication

The prevalence of low awareness of DM medication was 87.9% in females and 86.0% in males. Gender, educational level, marital status, economic status, religiosity, happiness status, insomnia and depressive symptoms were not statistically significant differences between the low-awareness DM medication group and the high-awareness DM medication group (Table 1). Age, residency, health insurance coverage, blood glucose monitoring status, comorbidities and general medical check-up were selected as potential factors associated with low awareness of DM medication on the basis of bivariate analyses. In the multivariate model, irregular blood glucose monitoring (OR: 23.61, 95% CI 11.46–48.65; $p < 0.001$), having no comorbidity (OR 2.03, 95% CI 1.05–3.90; $p = 0.034$), not having undergone any general medical check-up (OR 2.52, 95% CI 1.12–5.36; $p = 0.016$), 26–35 years of age (OR 4.96, 95% CI 1.06–23.19; $p = 0.042$), 36–45 years of age (OR 5.04, 95% CI 1.17–21.69; $p = 0.030$) and having no health insurance coverage (OR 2.08, 95% CI 1.12–3.87; $p = 0.021$) were significantly associated with low awareness of diabetes medication (Table 2). The goodness-of-fit p-value of the model was 0.552, with an R-squared value of 34.71%.

4. Discussion

This study revealed that more than three-quarters of DM patients had low awareness of their medication therapy. This implies that only one out of four patients had high awareness of their medication therapy. Age and healthcare insurance coverage are sociodemographic factors associated with awareness of DM medication, whereas blood glucose monitoring status, comorbidity status, and having a routine medical check-up are behavioural factors associated with awareness of DM medication.

We observed that the young (26–35 years old) and middle (36–45 years old) adulthood were associated with low awareness of DM medication, similar to what a Malaysian study revealed (34). This might be due to the old misconception that DM is a disease that primarily only affects the elderly (35). However, the prevalence of type 2 diabetes in adolescents and adults is dramatically increasing due to unhealthy lifestyles and obesity, which has a more aggressive disease profile, leading to premature complications that affect the quality of life and long-term outcomes (36).

In this study, coverage of health insurance and awareness of DM medication were observed to be in significant association. Individuals with no health insurance were twice as likely to have low awareness of their medication therapy when compared with those who have health insurance. This might be because self-paying DM patients may have high substantial medical expenses or financial issues. In turn, this may lead them to forego diabetic care as well as DM medication that would otherwise help them survive their conditions (37). Another possible explanation for this is that patients without medical insurance more likely skip regular medical care (38) and do not acquire better education, which may lead to low awareness of DM medication therapy.

We further observed that the number of comorbidities had a significant association with awareness of DM medication. This study revealed that individuals with no comorbidities were twice as likely to have low awareness of their DM medication when compared with those with 1–3 comorbidities. This finding is in line with the results of a study conducted in Malaysia reporting that patients with comorbidities had a high level of awareness; however, their level of self-care practise for diabetes remained low (39). Other recent qualitative research showed that patients with no comorbidities felt they may have prevented disease progression had they been given a more detailed explanation of their situation earlier as they were unaware of the risk factors, complications, and comorbidities (40).

DM individuals with irregular blood glucose monitoring likely had low awareness of their medication therapy when compared with individuals with regular blood glucose monitoring. The possible reason is perhaps that patients with regular blood glucose monitoring were more conscious of the consequences of not taking the drugs appropriately (41). DM patients who never had general medical check-ups were twice as likely to have low awareness of DM medication when compared with those who had had a general medical check-up. This might be because general medical check-ups provide health-related information, help identify issues early, assist in planning treatments as well as improve the awareness of medication (42).

A majority of the sociodemographic factors were not associated with low awareness of DM medication. Sociodemographic factors, such as marital status, may be overly generic when predicting an individual's DM medication awareness. A study revealed no correlation between gender and low awareness of DM medication (43). However, males were reported to be less aware of DM than females were (44, 45). Furthermore, in our study, educational level is not related to medication awareness as health literacy may be more essential than educational level (46). By contrast, another study found individuals with higher educational levels to have more awareness (47). The current study also found residency not to be associated with low awareness of DM medication, contradicting the results of a previous study, which reported a correlation between urban residence and high awareness of DM medication as urban residents seek therapy more often and have easier access to care (48). Moreover, religiosity was not associated with DM medication awareness in this study. This finding is contrary to the results of previous studies suggesting that, in terms of providing assistance and coping with a disease condition, religiosity played a significant role (49, 50). We further observed that depressive symptoms, happiness, insomnia and satisfaction with healthcare are not associated with low awareness of DM medication therapy. Previous studies have reported that depressive symptoms (51), insomnia (52), and happiness (53)

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TABLE 1 Baseline characteristics of the study population.

Characteristic	Low awareness of diabetes mellitus medication		High awareness of diabetes mellitus medication		*p*-value	Total respondent ($n = 706$)	
	n	%	*n*	%		*n*	%
Total respondent	619	87.7	87	12.3		706	100%
Gender					0.467		
Female	348	87.9	48	12.1		396	58.8
Male	239	86.0	39	14.0		278	41.2
Missing	32	–	–	–		32	–
Age					<0.001*		
15–25 years old	48	100	0	0		48	7.1
26–35 years old	85	96.6	3	3.4		88	13.0
36–45 years old	103	97.2	3	2.8		106	15.7
46–55 years old	89	82.4	19	17.6		108	16.0
55–65 years old	153	78.9	41	21.1		194	28.8
>65 years old	109	83.8	21	16.2		130	19.3
Missing	32	–	–	–		32	–
Education level					0.237*		
No education	73	93.6	5	6.4		78	11.8
Elementary school	227	88.0	31	12.0		258	38.9
Junior high school	93	86.1	15	13.9		108	16.3
Senior high school	10	83.3	2	16.7		12	1.8
University	173	83.6	34	16.4		207	31.2
Missing	43	–	–	–		43	–
Marital status					0.105*		
Not currently Married	163	90.6	17	9.4		180	26.7
Currently married	424	85.8	70	14.2		494	73.3
Missing	32	–	–	–		32	–
Economic status					0.231*		
Quintile 1	127	87.6	18	12.4		153	22.7
Quintile 2	103	86.5	16	13.5		107	15.9
Quintile 3	120	88.2	16	11.8		134	19.9
Quintile 4	111	91.7	10	8.3		126	18.7
Quintile 5	126	82.3	27	17.7		154	22.8
Missing	32	–	–	–		32	–
Residency					<0.001*		
Rural	247	93.6	17	6.4		264	37.4
Urban	372	84.2	70	15.8		442	62.6
Missing	–	–	–	–		–	–
Coverage of health insurance					<0.001*		
No	320	91.2	31	8.8		351	52.6
Yes	261	82.6	55	17.4		316	47.4
Missing	38	–	1	–		39	–
Religiosity					0.916		

(Continued)

TABLE 1 (Continued)

Characteristic	Low awareness of diabetes mellitus medication		High awareness of diabetes mellitus medication		*p*-value	Total respondent ($n = 706$)	
	n	%	*n*	%		*n*	%
Not religious	86	86.9	13	13.1		99	16.3
Religious	445	87.2	65	12.8		510	83.7
Missing	88	–	9	–		97	–
Having comorbidities	<0.001*						
No comorbid	313	93.7	21	6.3		334	50.1
1–3 Comorbidities	263	81.1	61	18.9		323	48.4
≥4 Comorbidities	5	55.6	4	44.4		10	1.5
Missing	38	–	1	–		39	–
Blood glucose control status					<0.001*		
Irregularly	514	93.3	37	6.7		551	90.3
Regularly	18	30.5	41	69.5		59	9.7
Missing	87	–	9	–		96	–
Having general medical check-up					<0.001*		
No	484	89.8	55	10.2		539	88.4
Yes	48	67.6	23	32.4		71	11.6
Missing	96	–	–	–		96	–
Health care satisfaction	0.836						
Low	122	87.8	17	12.2		139	22.7
High	412	87.1	61	12.9		473	77.3
Missing	85	–	9	–		94	–
Happiness status					0.726		
Unhappy	62	88.6	8	11.4		70	11.4
Happy	472	87.1	70	12.9		542	88.6
Missing	85	–	9	–		94	–
Insomnia					0.994		
Yes	75	87.2	11	12.8		525	85.9
No	458	87.2	67	12.8		86	14.1
Missing	86	–	9	–		95	–
Depressive symptoms status					0.213*		
Not depressed	410	86.3	65	13.7		475	77.9
Depressed	122	90.4	13	9.6		135	22.1
Missing	87	–	9	–		96	–

*Included in the initial multivariate model.

were not associated with awareness of DM medication, which is in line with the present results. By contrast, in previous studies, the individuals more satisfied with their healthcare were possibly more aware of their DM medication (54, 55).

The awareness of DM patients about their medication is crucial for ensuring that they take their DM medication as prescribed to avoid any complications or associated morbidities and mortality (56). These findings may help us understand the significance of the issue of awareness of DM medication as well as offer potential solutions. During the early stages of the disease, it may be necessary for patients to be better informed to increase their awareness of DM medication, particularly among those at high risk of developing comorbidities and complications (57). To raise patients' awareness of DM medication, healthcare professionals are essential information resources and play a leading role in the awareness-raising effort (58). Patient education can be improved by first determining the individual's learning needs and then providing them with individualised educational interventions tailored to meet their requirements (59). This study,

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TABLE 2 Association between sociodemographic and behavioural factors and low awareness of diabetes mellitus medication.

Characteristic	Univariate		Multivariate[a]	
	Crude OR [95% CI]	*p*-value	Adjusted OR [95% CI][b]	*p*-value
Age				
15–25 years old	1		1	
26–35 years old	5.46 [1.58–18.91]	0.007*	4.96 [1.06–23.19]	0.042*
36–45 years old	6.61 [1.92–22.84]	0.003*	5.04 [1.17–21.69]	0.030*
46–55 years old	0.90 [0.46–1.78]	0.768	1.07 [0.41–2.76]	0.895
55–65 years old	0.72 [0.40–1.28]	0.265	0.61 [0.27–1.40]	0.244
>65 years old	Reference		Reference	
Coverage of health insurance				
Yes	Reference		Reference	
No	2.18 [1.36–3.48]	0.001*	2.08 [1.12–3.87]	0.021*
Comorbidity status				
No comorbid	3.47 [2.06–5.85]	<0.001*	2.03 [1.05–3.90]	0.034*
1–3 Comorbidities	Reference		Reference	
≥4 Comorbidities	0.35 [0.95–1.28]	0.111	1.12 [0.65–19.72]	0,933
Blood glucose control status				
Regularly	Reference		Reference	
Irregularly	31.64 [16.57–60.42]	<0.001*	23.61 [11.46–48.65]	0.000*
Having general medical check-up				
Yes	Reference		Reference	
No	4.22 [2.38–7.46]	<0.001*	2.52 [1.12–5.36]	0.016*
Education level				
No education	2.87 [1.08–7.63]	0.035*	-	-
Elementary school	1.44 [0.85–2.43]	0.174	-	-
Junior high school	1.22 [0.63–2.35]	0.556	-	-
Senior high school	0.98 [0.21–4.69]	0.982	-	-
University	Reference		-	-
Marital status				
Not currently married	1.58 [0.90–2.77]	0.108	-	-
Currently married	Reference		-	-
Economic status				
Quintile 1	1.51 [0.79–2.88]	0.209	-	-
Quintile 2	1.38 [0.71–2.70]	0.347	-	-
Quintile 3	1.61 [0.82–3.13]	0.163	-	-
Quintile 4	2.38 [1.10–5.13]	0.027	-	-
Quintile 5	Reference		-	-
Residency				
Rural	2.73 [1.57–4.76]	<0.001*	-	-
Urban	Reference		-	-
Depressive symptoms status				
Depressed	1.49 [0.79–2.79]	0.216	-	-
Not depressed	Reference			

[a]Goodness-of-fit p-value of final model: 0.552; pseudo-R-squared: 34.71%.

[b]Final multivariate model.

*$p < 0.05$ at the 5% level of significance.

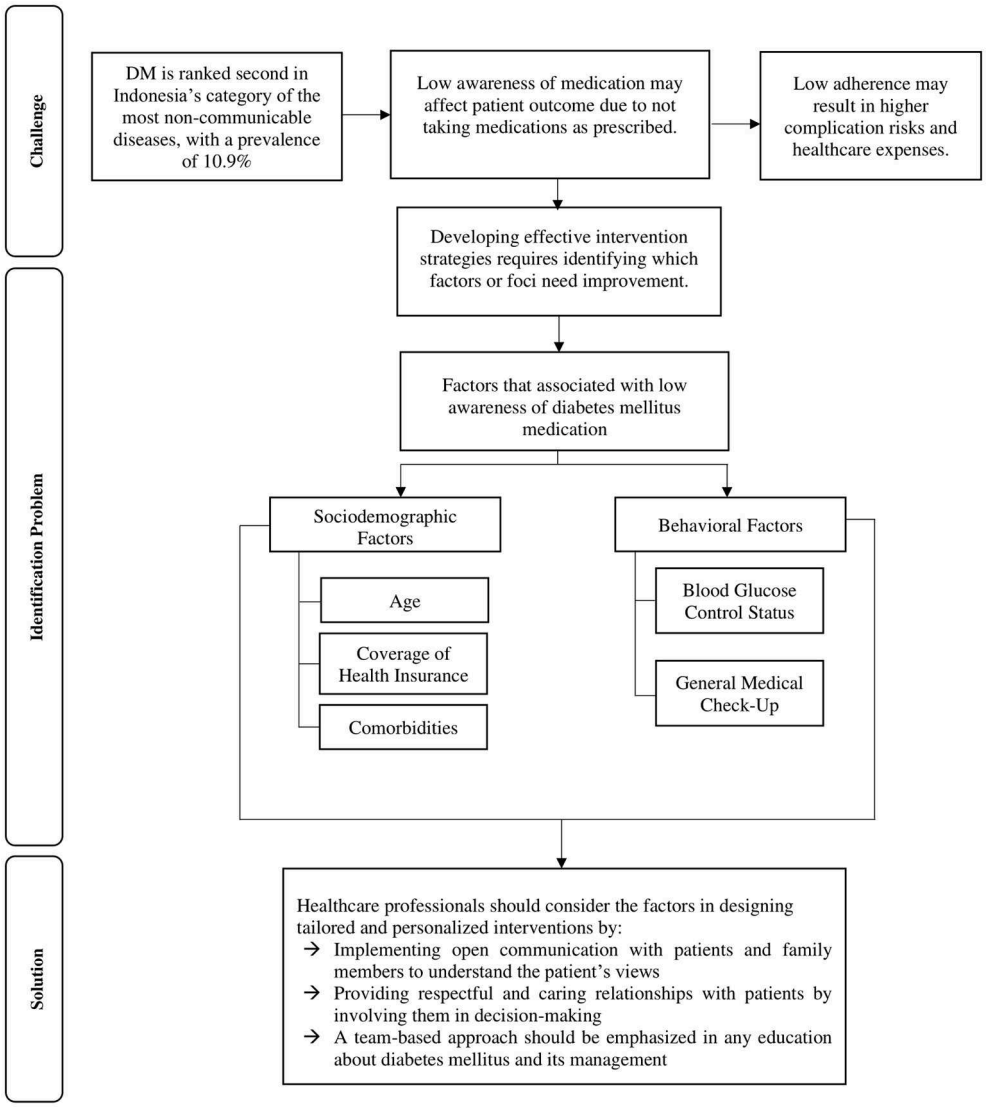

FIGURE 1
Problems, challenges and solutions to increase the level of awareness of diabetes mellitus medication in Indonesia.

thus, advocates for better management of DM by inquiring about medication adherence during clinical consultations and improving the quality of DM care. Furthermore, patient education, counselling and behavioural support are critical for achieving successful DM medication therapy. These tailored interventions could be effective in clarifying misconceptions and help clear up any misunderstandings to increase their level of awareness (60). In addition, healthcare professionals should monitor blood glucose, perform general medical check-ups regularly, prioritise patient satisfaction by ensuring that they receive appropriate care and establish a respectful and caring relationship with patients by involving them in decision-making (61). The current findings may be useful as a point of reference for healthcare professionals, addressing factors related to low awareness of DM medication (Figure 1).

Until now, the present study appears to be the first to assess the awareness of DM medication and its associated factors in Indonesia. The study's strength is that we used the IFLS data, which represents 83% of the Indonesian population with an attrition rate of only 6%. The IFLS provides numerous benefits, including large samples, which are relatively heterogenous, less expensive than collecting new data and representative of the Indonesia setting. Besides, in this analysis, individuals were included on the basis of an objective measurement of HbA1c >6.5%, providing a more objective individual selection and avoiding selection bias. Despite its strengths, the study certainly has certain limitations related to methodological issues. First, notably, the cross-sectional design of the study precludes any causal inferences regarding the relationship between sociodemographic and behavioural factors and low awareness of DM medication. Second, since we performed a complete case analysis, it may have reduced the statistical power, probably increasing the possibility of bias in our estimation, which might be an overestimation or underestimation of conclusions. Third, we have a wide CI value, indicating a greater likelihood of uncertainty regarding whether we have precisely estimated the strength of the association. Fourth, this study was at risk to recall bias due to disparities in accuracy in recalling past events based on self-reported answers from several variable independents. Fifth, this study was unable to distinguish between DM types 1 and 2. Sixth, as we relied on a secondary database that provides binary

outcomes for the awareness, it might not be adequate to explain the multidimensional aspects of behavioural science (62). Further studies are needed to consider these aspects when assessing awareness of medication. Seventh, our model's overall association was low, indicating the possibility of other unmeasured factors influencing the low awareness of DM medication, such as another comorbid disease such as kidney disease (63), healthy lifestyle (64), education about DM (65), the number of medicines in the therapy (66), duration of DM (67), ethnic background (68) or medication beliefs (69).

5. Conclusion

Healthcare professionals should monitor blood glucose, perform general medical check-ups regularly, prioritise patient satisfaction by ensuring that they receive appropriate care and establish a respectful and caring relationship with patients by involving them in the decision-making process. Patient education can be improved by first determining the individual's learning needs and then providing them with individualised educational interventions tailored to meet their requirements in order to improve their awareness of DM medication. Therefore, our findings reveal the need to develop intervention strategies targeting those who irregularly monitor their blood glucose level; who irregularly undergo general medical check-ups, with multiple comorbidities; who have no health insurance coverage and who are young.

Data availability statement

Publicly available datasets were analysed in this study. This data can be found here: https://www.rand.org/well-being/social-and-behavioral-policy/data/FLS/IFLS.html.

Ethics statement

The Ethical Review Boards of the RAND's Human Subjects Protection Committee (s0064-06-01-CR01) approved IFLS research. Research Ethics Committee of Universitas Padjadjaran, Indonesia, waived the requirement because this study uses anonymous data from the IFLS.

Author contributions

QK wrote the first draft of this manuscript. SA and RA participated in the design of the study. QK and SA participated in data analysis and interpretation. QK, SA, and RA revised the manuscript. All authors approved the final manuscript.

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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.

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References

1. WHO. *Fact Sheets: Diabetes. World Heal Organ.* (2021). Available online at: https://www.who.int/news-room/fact-sheets/detail/diabetes#::text=In2019%2Cdiabeteswasthedirectcauseof1.5million,ageof70fromdiabetes (accessed February 21, 2022).

2. IDF. *IDF Diabetes Atlas 10th Edition*. Belgium: International Diabetes Federation (2021).

3. Kemenkes RI. *Hasil Riset Kesehatan Dasar Tahun 2018*. Jakarta: Kementrian Kesehatan Republik Indonesia (2018).

4. IDF. *IDF Diabetes Atlas: Indoensia Diabetes Report 2000 – 2045*. (2021). Available online at: https://www.diabetesatlas.org/data/en/country/94/id.html (accessed October 29, 2022).

5. Shi Y, Vanhoutte PM. Macro- and microvascular endothelial dysfunction in diabetes. *J Diabetes.* (2017) 9:434–49. doi: 10.1111/1753-0407.12521

6. ADA. *Standards of Medical Care in Diabetes-2020 Abridged for Primary Care Providers. United States*. American Diabetes Association (2020).

7. AlShayban DM, Naqvi AA, Alhumaid O, AlQahtani AS, Islam MA, Ghori SA, et al. Association of disease knowledge and medication adherence among out-patients with type 2 diabetes mellitus in Khobar, Saudi Arabia. *Front Pharmacol.* (2020) 11:1–9. doi: 10.3389/fphar.2020.00060

8. Gafoor AK. Considerations in measurement of awareness. *Natl Semin Emerg Trends Educ.* (2012) 1–6. Available online at: https://files.eric.ed.gov/fulltext/ED545374.pdf (accessed November 11, 2022).

9. Gordon K, Smith F, Dhillon S. Effective chronic disease management: patients' perspectives on medication-related problems. *Patient Educ Couns.* (2007) 65:407–15. doi: 10.1016/j.pec.2006.09.012

10. Vrijens B, De Geest S, Hughes DA, Przemyslaw K, Demonceau J, Ruppar T, et al. A new taxonomy for describing and defining adherence to medications. *Br J Clin Pharmacol.* (2012) 73:691–705. doi: 10.1111/j.1365-2125.2012.04167.x

11. Araya EM, Gebrezgabiher HA, Tekulu GH, Alema NM, Getnet D, Gebru HT, et al. Medication non-adherence and associated factors among diabetic patients visiting general hospitals in the eastern zone of Tigrai, Northern Ethiopia. *Patient Prefer Adherence.* (2020) 14:2071–83. doi: 10.2147/PPA.S278148

12. Polonsky WH, Henry RR. Poor medication adherence in type 2 diabetes: recognizing the scope of the problem and its key contributors.

Patient Prefer Adherence. (2016) 10:1299–306. doi: 10.2147/PPA.S106821

13. Feldhaus I, Nagpal S, Verguet S. Alleviating the burden of diabetes with health equity funds: economic evaluation of the health and financial risk protection benefits in Cambodia. *PLoS ONE.* (2021) 16:e0259628. doi: 10.1371/journal.pone.0259628

14. McPherson ML, Smith SW, Powers A, Zuckerman IH. Association between diabetes patients' knowledge about medications and their blood glucose control. *Res Soc Adm Pharm.* (2008) 4:37–45. doi: 10.1016/j.sapharm.2007.01.002

15. Okuyan B, Sancar M, Izzettin FV. Assessment of medication knowledge and adherence among patients under oral chronic medication treatment in community pharmacy settings. *Pharmacoepidemiol Drug Saf.* (2012) 22:209–14. doi: 10.1002/pds.3275

16. Alkatheri AM, Albekairy AM. Does the patients' educational level and previous counseling affect their medication knowledge? *Ann Thorac Med.* (2013) 8:105–8. doi: 10.4103/1817-1737.109823

17. Abdu-Aguye SN, Labaran KS, Danjuma NM, Mohammed S. An exploratory study of outpatient medication knowledge and satisfaction with medication counselling at selected hospital pharmacies in Northwestern Nigeria. *PLoS ONE.* (2022) 17:1–14. doi: 10.1371/journal.pone.0266723

18. Strauss J, Witoelar F, Sikoki B. *The fifth wave of the Indonesia family life survey: overview and field report: volume 1.* RAND Corporation (2016) 94p.

19. Kamilah FZ, Habibie F, Rahma GR, Sofyan MNF, Isnaini NS, Nadhilah ND, et al. Analysis of the determinants of diabetes mellitus in Indonesia: a case study of the 2014 indonesian family life survey. *Dis Prev Public Heal J.* (2021) 15:88. doi: 10.12928/dpphj.v15i2.3079

20. Safitri AZ, Fajariyah RN, Astutik E. Risk factors of diabetes mellitus in urban communities in Indonesia (IFLS 5). *J Berk Epidemiol.* (2021) 9:184. doi: 10.20473/jbe.V9I22021.184-191

21. Tanoey J, Becher H. Diabetes prevalence and risk factors of early-onset adult diabetes: results from the Indonesian family life survey. *Glob Health Action.* (2021) 14:2001144. doi: 10.1080/16549716.2021.2001144

22. Asrullah M, L'Hoir M, Feskens EJM, Melse-Boonstra A. Trend in age at menarche and its association with body weight, body mass index and non-communicable disease prevalence in Indonesia: evidence from the Indonesian Family Life Survey (IFLS). *BMC Public Health.* (2022) 22:1–10. doi: 10.1186/s12889-022-12995-3

23. Indrahadi D, Wardana A, Pierewan AC. The prevalence of diabetes mellitus and relationship with socioeconomic status in the Indonesian population. *J Gizi Klin Indones.* (2021) 17:103. doi: 10.22146/ijcn.55003

24. Finkelstein EA, Chay J, Bajpai S. The economic burden of self-reported and undiagnosed cardiovascular diseases and diabetes on Indonesian households. *PLoS ONE.* (2014) 9:e99572. doi: 10.1371/journal.pone.0099572

25. ADA. Classification and diagnosis of diabetes : standards of medical care in diabetes — 2022. *Diabetes Care.* (2022) 45:17–38. doi: 10.2337/dc22-S002

26. Mastronardi CA, Whittle B, Tunningley R, Neeman T, Paz-Filho G. The use of dried blood spot sampling for the measurement of HbA1c: a cross-sectional study. *BMC Clin Pathol.* (2015) 15:1–7. doi: 10.1186/s12907-015-0013-5

27. IFLS-5. "Buku III B," in *Survei Aspek Kehidupan Rumah Tangga Indonesia 2014.* Jakarta: RAND.

28. Vilagut G, Forero CG, Barbaglia G, Alonso J. Screening for depression in the general population with the center for epidemiologic studies depression (ces-d): a systematic review with meta-analysis. *PLoS ONE.* (2016) 11:1–17. doi: 10.1371/journal.pone.0155431

29. Björgvinsson T, Kertz SJ, Bigda-Peyton JS, McCoy KL, Aderka IM. Psychometric properties of the CES-D-10 in a psychiatric sample. *Assessment.* (2013) 20:429–36. doi: 10.1177/1073191113481998

30. Yu L, Buysse DJ, Germain A, Moul DE, Stover A, Dodds NE, et al. Development of short forms from the PROMIS sleep disturbance and sleep-related impairment item banks. *Behav Sleep Med.* (2011) 10:6–24. doi: 10.1080/15402002.2012.636266

31. Peltzer K, Pengpid S. Prevalence, social and health correlates of insomnia among persons 15 years and older in Indonesia. *Psychol Heal Med.* (2019) 24:757–68. doi: 10.1080/13548506.2019.1566621

32. Li C. Little's test of missing completely at random. *Stata J.* (2013) 13:795–809. doi: 10.1177/1536867X1301300407

33. Dodge Y. Chi-square Goodness of Fit Test. In: *The Concise Encyclopedia of Statistics.* New York, NY: Springer New York, 72–6.

34. Ho BK, Jasvindar K, Gurpreet K, Ambigga D, Suthahar A, Cheong SM, et al. Prevalence, awareness, treatment and control of diabetes mellitus among the elderly: the 2011 National Health and Morbidity Survey, Malaysia. *Malaysian Fam Physician.* (2014) 9:12–9.

35. Song SH. Emerging type 2 diabetes in young adults. *Adv Exp Med Biol.* (2012) 771:51–61. doi: 10.1007/978-1-4614-5441-0_7

36. Lascar N, Brown J, Pattison H, Barnett AH, Bailey CJ, Bellary S. Type 2 diabetes in adolescents and young adults. *Lancet Diabetes Endocrinol.* (2018) 6:69–80. doi: 10.1016/S2213-8587(17)30186-9

37. Al-Sanaani EA, Ismail A, Manaf MRA, Suddin LS, Mustafa N, Sukor N, et al. Health insurance status and its determinants among patients with type 2 diabetes mellitus in a tertiary teaching hospital in Malaysia. *PLoS ONE.* (2022) 17:1–17. doi: 10.1371/journal.pone.0267897

38. Tipirneni R, Politi MC, Kullgren JT, Kieffer EC, Goold SD, Scherer AM. Association between health insurance literacy and avoidance of health care services owing to cost. *JAMA Netw open.* (2018) 1:e184796. doi: 10.1001/jamanetworkopen.2018.4796

39. Abdulghani HM, AlRajeh AS, AlSalman BH, AlTurki LS, AlNajashi NS, Irshad M, et al. Prevalence of diabetic comorbidities and knowledge and practices of foot care among diabetic patients: a cross-sectional study. *Diabetes, Metab Syndr Obes Targets Ther.* (2018) 11:417–25. doi: 10.2147/DMSO.S171526

40. Lopez-Vargas PA, Tong A, Howell M, Phoon RKS, Chadban SJ, Shen Y, et al. Patient awareness and beliefs about the risk factors and comorbidities associated with chronic kidney disease : a mixed-methods study. *Nephrology.* (2017) 22:374–81. doi: 10.1111/nep.12829

41. Pascal IGU, Ofoedu JN, Uchenna NP, Nkwa AA, Uchamma GUE. Blood glucose control and medication adherence among adult type 2 diabetic Nigerians attending a primary care clinic in under-resourced environment of eastern Nigeria. *N Am J Med Sci.* (2012) 4:310–5. doi: 10.4103/1947-2714.98590

42. Ngo TT, Hoang PN, Pham HV, Nguyen DN, Bui HTT, Nguyen AT, et al. Routine medical check-up and self-treatment practices among community-dwelling living in a mountainous area of Northern Vietnam. *Biomed Res Int.* (2021) 2021:8734615. doi: 10.1155/2021/8734615

43. Shahzad A, Ahmad MM, Anwer I, Ijaz N, Shahzad M, Usman M. Gender-specific knowledge of diabetes and its management among patients visiting outpatient clinics in Faisalabad, Pakistan. *Cureus.* (2018) 10:2–10. doi: 10.7759/cureus.3119

44. Siddiqui M, Khan M, Carline T. Gender differences in living with diabetes mellitus. *Mater Socio Medica.* (2013) 25:140. doi: 10.5455/msm.2013.25.140-142

45. Obirikorang Y, Obirikorang C, Anto EO, Acheampong E, Batu EN, Stella AD, et al. Knowledge of complications of diabetes mellitus among patients visiting the diabetes clinic at Sampa Government Hospital, Ghana: a descriptive study. *BMC Public Health.* (2016) 16:1–8. doi: 10.1186/s12889-016-3311-7

46. Zowgar AM, Siddiqui MI, Alattas KM. Level of diabetes knowledge among adult patients with diabetes using diabetes knowledge test. *Saudi Med J.* (2018) 39:161–8. doi: 10.15537/smj.2017.2.21343

47. Al-Rasheedi AAS. The role of educational level in glycemic control among patients with type II diabetes mellitus. *Int J Health Sci.* (2014) 8:177–87. doi: 10.12816/0006084

48. Deepa M, Bhansali A, Anjana R, Pradeepa R, Joshi S, Joshi P, et al. Knowledge and awareness of diabetes in urban and rural India: the Indian council of medical research india diabetes study (phase i): Indian council of medical research india diabetes 4. *Indian J Endocrinol Metab.* (2014) 18:379–85. doi: 10.4103/2230-8210.131191

49. Watkins YJ, Quinn LT, Ruggiero L, Quinn MT, Choi Y-K. Spiritual and religious beliefs and practices, and social support's relationship to diabetes self-care activities in African Americans. *Diabetes Educ.* (2013) 39:231–9. doi: 10.1177/0145721713475843

50. Duke N. Type 2 diabetes self-management: spirituality, coping and responsibility. *J Res Nurs.* (2021) 26:743–60. doi: 10.1177/17449871211026958

51. Egede LE, Ellis C. The effects of depression on diabetes knowledge, diabetes self-management, and perceived control in indigent patients with type 2 diabetes. *Diabetes Technol Ther.* (2008) 10:213–9. doi: 10.1089/dia.2007.0278

52. Schipper SBJ, Van Veen MM, Elders PJM, van Straten A, Van Der Werf YD, Knutson KL, et al. Sleep disorders in people with type 2 diabetes and associated health outcomes: a review of the literature. *Diabetologia.* (2021) 64:2367–77. doi: 10.1007/s00125-021-05541-0

53. Liu SY, Huang J, Dong QL, Li B, Zhao X, Xu R, Yin HF. Diabetes distress, happiness, and its associated factors among type 2 diabetes mellitus patients with different therapies. *Medicine.* (2020) 99:e18831. doi: 10.1097/MD.0000000000018831

54. White RO Eden S, Wallston KA, Kripalani S, Barto S, Shintani A, Rothman RL. Health communication, self-care, and treatment satisfaction among low-income diabetes patients in a public health setting. *Patient Educ Couns.* (2015) 98:144–9. doi: 10.1016/j.pec.2014.10.019

55. Boels AM, Vos RC, Hermans TGT, Zuithoff NPA, Müller N, Khunti K, et al. What determines treatment satisfaction of patients with type 2 diabetes on insulin therapy? An observational study in eight European countries. *BMJ Open.* (2017) 7:e016180. doi: 10.1136/bmjopen-2017-016180

56. Al-Qazaz HK, Sulaiman SA, Hassali MA, Shafie AA, Sundram S, Al-Nuri R, et al. Diabetes knowledge, medication adherence and glycemic control among patients with type 2 diabetes. *Int J Clin Pharm.* (2011) 33:1028–35. doi: 10.1007/s11096-011-9582-2

57. Chatterjee S, Davies MJ, Heller S, Speight J, Snoek FJ, Khunti K. Diabetes structured self-management education programmes: a narrative review and current innovations. *Lancet Diabetes Endocrinol.* (2018) 6:130–42. doi: 10.1016/S2213-8587(17)30239-5

58. Tan E, Khoo J, Gani LU, Malakar RD, Tay TL, Tirukonda PS, et al. Effect of multidisciplinary intensive targeted care in improving diabetes

mellitus outcomes: a randomized controlled pilot study - the integrated diabetes education, awareness and lifestyle modification in Singapore (IDEALS) Program. *Trials.* (2019) 20:1–10. doi: 10.1186/s13063-019-3601-3

59. Adam L, O'Connor C, Garcia AC. Evaluating the impact of diabetes self-management education methods on knowledge, attitudes and behaviours of adult patients with type 2 diabetes mellitus. *Can J Diabetes.* (2018) 42:470–7.e2. doi: 10.1016/j.jcjd.2017.11.003

60. Cruz-Cobo C, Santi-Cano MJ. Efficacy of diabetes education in adults with diabetes mellitus type 2 in primary care: a systematic review. *J Nurs Scholarsh.* (2020) 52:155–63. doi: 10.1111/jnu.12539

61. Tamhane S, Rodriguez-Gutierrez R, Hargraves I, Montori VM. Shared decision-making in diabetes care. *Curr Diab Rep.* (2015) 15:1–10. doi: 10.1007/s11892-015-0688-0

62. Trevethan R. Deconstructing and assessing knowledge and awareness in public health research. *Front Public Heal.* (2017) 5:16–9. doi: 10.3389/fpubh.2017.00194

63. Nordheim E, Jenssen TG. Chronic kidney disease in patients with diabetes mellitus. *Endocr Connect.* (2021) 10:R151–9. doi: 10.1530/EC-21-0097

64. Ang BW, Tan MY, Goh CM, Rahardja S, Lim BY, Chiew W, et al. Impact of knowledge and attitudes on lifestyle practices in preventing type 2 diabetes mellitus. *Ann Acad Med Singapore.* (2019) 48:247–63. doi: 10.47102/annals-acadmedsg.V48N8p247

65. Foma MA, Saidu Y, Omoleke SA, Jafali J. Awareness of diabetes mellitus among diabetic patients in the Gambia: a strong case for health education and promotion. *BMC Public Health.* (2013) 13:1–8. doi: 10.1186/1471-2458-13-1124

66. Shams N, Amjad S, Kumar N, Ahmed W, Saleem F. Drug non-adherence in type 2 diabetes mellitus; predictors and associations. *J Ayub Med Coll Abbottabad.* (2016) 28:302–7.

67. Ghouse J, Isaksen JL, Skov MW, Lind B, Svendsen JH, Kanters JK, et al. Effect of diabetes duration on the relationship between glycaemic control and risk of death in older adults with type 2 diabetes. *Diabetes, Obes Metab.* (2020) 22:231–42. doi: 10.1111/dom.13891

68. Taylor YJ, Davis ME, Monahan S, Robertson S, Robinson MD. Awareness of racial disparities in diabetes among primary care residents and preparedness to discuss disparities with patients. *J Racial Ethn Heal Disparities.* (2019) 6:237–44. doi: 10.1007/s40615-018-0518-6

69. Alfian SD, Annisa N, Fajriansyah F, Perwitasari DA, Abdulah R, Hak E, et al. Modifiable factors associated with non-adherence to antihypertensive or antihyperlipidemic drugs are dissimilar: a multicenter study among patients with diabetes in Indonesia. *J Gen Intern Med.* (2020) 35:2897–906. doi: 10.1007/s11606-020-05809-y

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Perceptions and responses to cognitive decline in people with diabetes: A systematic review of qualitative studies

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Objective: We aimed at summarizing the perceptions and responses to cognitive decline, assessing the disease management, identifying deficiencies and proposing new strategies for improvement in people with diabetes (PWDs).

Methods: A comprehensive search was performed in the following nine databases: PubMed, EMBASE, Web of Science, The Cochrane Library, PsycINFO, CINAHL, WanFang, CNKI, and VIP. The Joanna Briggs Institute (JBI) Critical Appraisal Tool for qualitative research was utilized to evaluate the quality of included studies. Descriptive texts and quotations relating to patient experience were extracted from the included studies and thematically analyzed.

Results: Eight qualitative studies met the inclusion criteria and 2 overarching themes were identified: (1) self-perception of cognitive decline referred to perceived cognitive symptoms, lack of knowledge and, impaired self-management and coping in multiple methods; (2) reported benefits of cognitive interventions referred to how cognitive interventions improved disease management, attitudes and needs of PWDs.

Conclusion: PWDs described misconceptions about their cognitive decline and suffered from them during disease management. This study provides a patient-specific reference for cognitive screening and intervention in PWDs, supporting disease management with cognitive decline in clinical practice.

KEYWORDS

diabetes mellitus, cognitive decline, perception, qualitative research, systematic review

1. Introduction

Diabetes mellitus is a chronic metabolic disease that damages human health (1). Owing to an aging population and the growing number of people with obesity, diabetes has affected ~537 million people worldwide with an increasing prevalence (2). The comorbidities accompanied with diabetes also deteriorate people's quality of life, especially cognitive impairment which is associated with increasing risk of mortality (3). Studies have shown that cognitive decline in people with diabetes (PWDs) progresses twice as fast as normal aging, and more likely develops into Alzheimer's disease and dementia (4–6).

PWDs have to hold complex and perpetual self-management to maintain their health and independent lifestyle, all of which may be disorganized by cognitive decline (7, 8). PWDs with worse self-management and other complications such as hypoglycemia and depression are more

prone to cognitive impairment (9). A recent population-based cohort study suggested that poorly controlled diabetes was associated with double the risk of cognitive impairment and triple the risk of cognitive impairment progressing to dementia (10). PWDs will be caught in a vicious circle and unable to coexist with the disease along with decreasing quality of life, shorter life expectancy and higher mortality due to cognitive decline (11, 12). Therefore, it is critical for PWDs to timely and effectively improve and strength their disease management ability weakened due to cognitive decline.

Although cognitive decline can have deleterious effects, it is not irreparable. Studies have shown that ∼10–40% of people with mild cognitive impairment (MCI) may return to normal cognitive performance within ∼4–5 years (13). Cognitive interventions may also have a positive effect on cognitive decline in PWDs (14). However, there are various types of cognitive impairment, and different people have great heterogeneity in the symptoms they perceive and the ways they deal with cognitive impairment. A number of studies have explored the impact of cognitive decline on specific components of self-management in PWDs. Evidence has shown that global cognitive decline is mainly associated with poor medication management in PWDs (15), such as lower insulin self-injection knowledge (16), less responsibility for self-medication (17), improper filling (18) and being less likely to take oral medications on time (19). PWDs are also less likely to engage in glucose self-monitoring and use health care clinics properly (17, 19). Furthermore, a significant association was found between global cognitive decline and diet adherence (20). Nevertheless, specific symptoms and performances of disease management among PWDs with cognitive decline have not been systematically evaluated and synthesized.

Neuropsychological tests are now commonly used to assess PWDs' cognitive function. Common instruments are based on theoretical knowledge, statistical methods and diagnostic criteria, without a systematic qualitative research, which probably leads to omissions when mild cognitive changes are assessed (21, 22). Qualitative methods have the strength of addressing highly nuanced and contextualized aspects of a subjective experience (23). There is already a substantial literature dealing with the qualitative exploration of the lived experience of people with dementia (24–27). However, the qualitative description of the experiences underlying cognitive complaints has only recently been pursued with PWDs.

This study aimed at summarizing the perceptions and responses to cognitive decline in PWDs, assessing the disease management, identifying deficiencies and proposing new strategies for improvement.

2. Methods

This study adhered to the Preferred Reporting Items for Systematic review and Meta-Analysis guideline (PRISMA) (28). The study protocol was registered in PROSPERO with the registration number CRD42022301334.

2.1. Data source and search strategy

Two independent reviewers performed a comprehensive search in the following nine databases: PubMed, EMBASE, Web of Science, The Cochrane Library, PsycINFO, CINAHL, WanFang, CNKI, and VIP. The search period ended in January 2022. Search strategies for all databases are listed in Supplementary Table S1.

2.2. Eligibility criteria

We included qualitative studies conducted in individual with type 1 diabetes or type 2 diabetes published in a peer-reviewed journal in either English or Chinese language. Studies including people without diabetes were considered only if they specifically reported results for PWDs. The included studies should examine perceptions and/or experiences of cognitive decline, thoughts, attitudes, feelings and views. Studies were excluded if they did not meet the above criteria.

2.3. Data selection and extraction

Two reviewers independently screened all papers according to the eligibility criteria (defined earlier), extracted and cross-checked the data. The following information was extracted: the surname of the first author and publication year; location; sample; PWDs' age; method/theory; data collection methods; research objective. Themes from the study's result section and participants' direct quotations were extracted as findings. For studies without direct quotes, the researchers extracted appropriate text after repeatedly reading the narrative. Extracted data were then imported into MS Excel for further coding and integration. Discrepancies between reviewers were resolved through discussion or by referring to a third reviewer.

2.4. Quality assessment

The methodological quality of the included studies was independently assessed for quality by two reviewers using the criteria based on the JBI Critical Appraisal Tool for qualitative research. Each checklist item was graded as "Yes," "No," and "Unclear." The two reviewers shared the results of the checklist and arrived at a consensus.

2.5. Data synthesis and analysis

Thematic synthesis was used to analyze the qualitative data from the included papers (29). Two reviewers coded text fragments for similarity. All extracted results were read repeatedly to extract concepts for coding. Individual codes were then combined

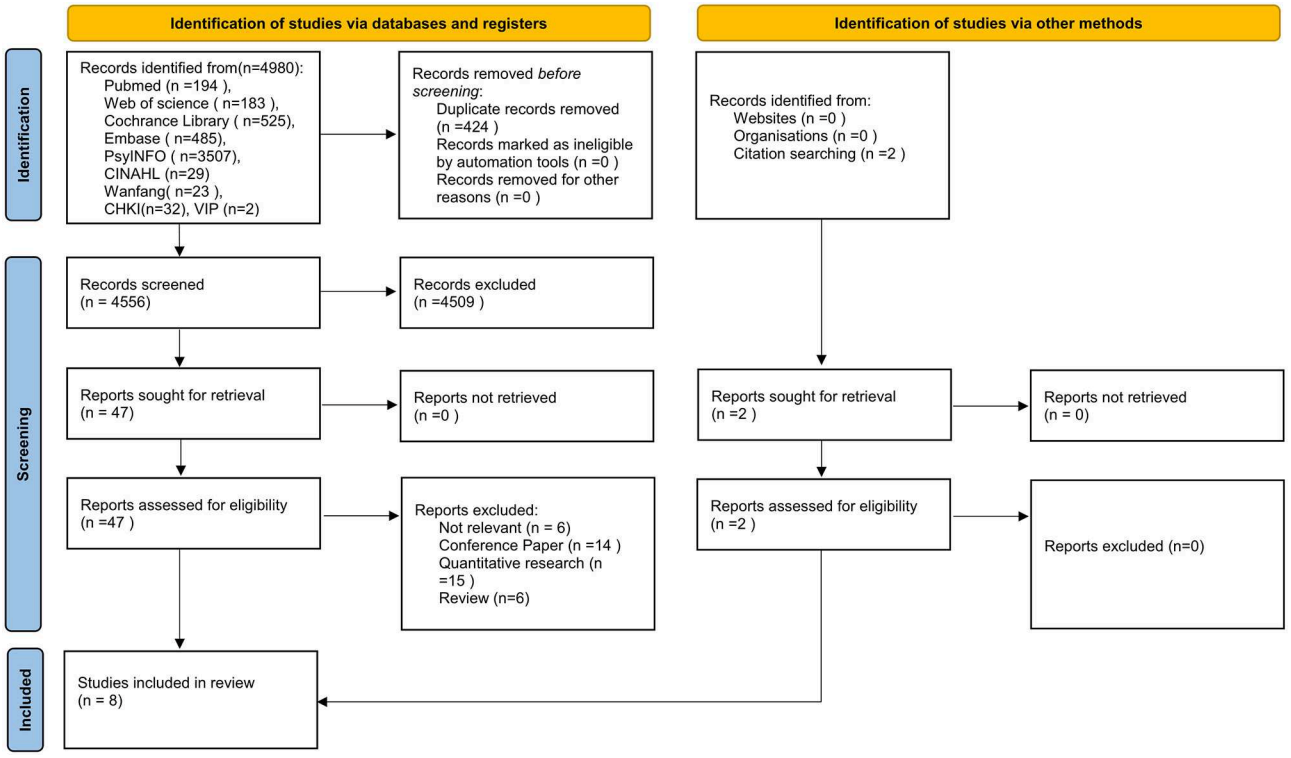

FIGURE 1
Flowchart of the study.

into groups and summarized by descriptive themes. Subthemes were used to further refine and categorize descriptive themes. Finally, distinct analytical themes were defined. The contents were organized into a structured hierarchy reflecting the content of the included studies. Distinct analytical themes were defined. The synthesized results reinforced the of current knowledge and generated new insights.

3. Results

3.1. Study selection

The initial search produced 4,556 articles after excluding duplicates, which were further reduced by 4,509 after excluding articles based on reviewing the titles and abstracts. Full texts of the remaining 49 articles were retrieved and a further 41 articles were excluded after review. Figure 1 shows the document selection process. Two of the papers were selected from the reference list of the included studies. Overall, eight qualitative studies were selected for inclusion.

3.2. Study characteristics

Inclusive study characteristics are shown in Table 1. Studies were conducted in the United Kingdom ($n = 2$), United States ($n = 3$), China ($n = 1$), Germany ($n = 1$), and New Zealand ($n = 1$). All included studies reported their sample size, which varied from 7 to 30 participants. All eight papers had participants with diabetes making a total sample size of 127. The JBI total score for each study is provided in Table 2. The quality of included studies varied considerably, with scores ranging from 60 to 90%.

3.3. Data synthesis

Two overarching themes were identified: self-perception of cognitive decline, and reported benefits of cognitive interventions. Supplementary Table S2 provides an overview of the two overarching themes and their sub-themes, with illustrative quotes from participants and a list of the codes.

3.3.1. Self-perception of cognitive decline

3.3.1.1. Underestimatione of cognitive decline associated with diabetes

Two studies reported participants' dissociative cognitive decline with diabetes and their unawareness of the link between diabetes and cognitive decline (33, 35). Unsurprisingly, cognitive problems were attributed to aging and considered to be a normal part of the aging process, albeit some participants were not elderly (33–35, 37). Persons appeared to treat the corresponding symptoms as common and ordinary and not as a problem with their cognitive health (37). In three studies, health care providers did not provide information about the association between diabetes and cognitive decline, which participants rarely acquired through other means

TABLE 1 Characteristics of included studies.

References	Location	Sample	Age (years)	Methodology/ theory orientation	Method of data collection	Research questions or objectives
Hasseler et al. (30)	Germany	7 T2DM patients	55–76	Symbolic interactionism	Problem-centered interview	To identify diabetes-related coping strategies and problems of adjustment to the disease from the perspective of PWDs.
Wilson (31)	UK	25 T1DM or T2DM patients	72–84	None stated	Telephone interview	To explore the views of older PWDs about the care they received from healthcare professionals.
Speight et al. (32)	UK	17 T1DM patients	35–57	Adapted grounded theory	Semi-structured interview	To explore individual experiences of severe hypoglycaemia occurring in daily life, and understand barriers to the prevention of hypoglycaemia.
Cuevas et al. (33)	USA	10 T2DM patients	44–70	None stated	Narrative interview	To explore the perceptions of people with T2DM regarding cognitive changes they experienced and examine informants' recommendations for modifications of existing cognitive rehabilitation interventions.
Cuevas et al. (34)	USA	19 T2DM patients	40–70	None stated	Focus group	To describe and focus specifically on the perceptions of people with T2DM in a cognitive rehabilitation intervention.
Hu and Zhang (35)	China	9 T2DM patients	41–57	Phenomenology	Semi-structured interview	To explore the cognition and feeling of MCI in PWDs, evaluate their performance and demand for cognitive rehabilitation intervention.
Chepulis et al. (36)	New Zealand	10 T2DM patients	26–75	None stated	Semi-structured interview	To provide an up-to-date assessment of challenges to diabetes care and glycemic control, particularly in patients with T2DM who have severe glycemic control.
Cuevas et al. (37)	USA	30 T2DM patients	Mean age = 66	None stated	One-on-one interview	To explore the meaning of cognitive health from the perspectives of Latinx adults with T2DM.

NB: T2DM, Type 2 diabetes mellitus; T1DM, Type 1 diabetes mellitus.

(34, 35, 37). Only a few participants knew a little about the dangers of hypoglycaemia on cognitive function (33, 35).

3.3.1.2. Suffering from cognitive symptoms

The most common symptom was deterioration in memory capacity. PWDs found themselves had significantly reduced memory capacity, especially short-term memory (32–35). Inadequate attention was another common symptom, with participants reporting the inability to concentrate and lack of interest (32). PWDs reported difficulties in generating thoughts and responses and/or maintaining psychomotor skills (31, 32, 37). In a single study, some PWDs who were aware of their cognitive dysfunction developed a sense of shame, remaining silent for fear of stigma (32).

3.3.1.3. Impaired diabetes self-management

Cognitive decline causes many difficulties for PWDs in the disease management and their daily activities. This extends to patients' ability to use and implement new knowledge in their everyday life (30). Medication non-adherence was a common finding as participants forgot to take their medication, resulting in unstable blood glucose levels and hospitalization (31, 33, 35, 36). A planned diets also became difficult to implement as plans are time-consuming and easy to forget (32, 33, 35). Participants often forgot the sequence of care routines leading to a loss of confidence in self-management of the disorder (31, 37). In addition, cognitive decline affected the their ability to care for family, work, and maintain social relationships, leading to problems with social functioning and the patient's quality of life (37).

3.3.1.4. Coping in multiple methods

Although cognitive decline disabling, many PWDs devised useful ways to actively cope with the problem. Some PWDs educated themselves through books or online to compensate for their lack of knowledge on the disease and the knowledge from health care providers (33, 37). Many

TABLE 2 Quality assessments.

References	Criteria										Score (%)
	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10	
Hasseler et al. (30)	U	Y	Y	Y	Y	Y	N	N	N	Y	60%
Wilson (31)	U	Y	Y	Y	Y	N	N	Y	Y	Y	70%
Speight et al. (32)	U	Y	Y	Y	Y	Y	Y	Y	Y	Y	90%
Cuevas et al. (33)	Y	Y	Y	Y	Y	U	Y	Y	Y	Y	90%
Cuevas et al. (34)	U	Y	Y	Y	Y	U	Y	Y	Y	Y	80%
Hu and Zhang (35)	Y	Y	Y	Y	Y	N	U	Y	Y	Y	80%
Chepulis et al. (36)	U	Y	Y	Y	Y	Y	U	Y	Y	Y	80%
Cuevas et al. (37)	Y	Y	Y	Y	Y	Y	U	Y	Y	Y	90%

Y, Yes; N, No; U, Unclear; Q1, Congruity between the stated philosophical perspective and the research methodology; Q2, Congruity between the research methodology and the research question or objectives; Q3, Congruity between the research methodology and the methods used to collect data; Q4, Congruity between the research methodology and the representation and analysis of data; Q5, There is congruence between the research methodology and the interpretation of results; Q6, Locating the researcher culturally or theoretically; Q7, Influence of the researcher on the research, and vice-versa, is addressed; Q8, Representation of participants and their voices; Q9, Ethical approval by an appropriate body; Q10, Relationship of conclusions to analysis, or interpretation of the data.

PWDs implemented compensatory strategies such as list-making or other mnemonic devices to mitigate deficits in attention/orientation (33, 34, 37). This ensured work or life balance (37). Many other methods were used to promote cognitive health such as having a hobby, doing physical activity, playing video games, dieting and having social interactions (33, 37). Participants often acknowledged the cognitive benefits of these approaches without verifying whether these methods really work.

3.3.2. Reported benefits from cognitive interventions

3.3.2.1. Benefiting from cognitive interventions

PWDs received cognitive interventions in two studies, including educational sessions teaching compensatory cognitive strategies and online brain training programs (33, 34). They benefited from these interventions, supporting an improved diabetes management. Some participants learned the content of the intervention and applied the cognitive strategies to their long-term practice (33). After taking educational sessions, PWDs used to interact better with their health care providers, for example asking to assess a potential vitamin B12 deficiency associated with metformin use or requesting to perform additional tests for the measurement of cognitive deficits (34). Through practice of cognitive strategies in the class, PWDs were also facilitated to think about and use cognitive strategies in the process of disease self-management thus helping them to manage their progress as well as consider the basis for many other methods (34). Some PWDs realized that the effects of the intervention were both short-term and long-term. The short-term effect resulted from learning to think better and using cognitive strategies, while the long-term effect was related to better diabetes management such as improved blood glucose, cholesterol, blood pressure, and cognitive function (34). Most participants had a sense of achievement and felt that the intervention had helped to improve their "mental capacity and flexibility in planning cognitive strategies" (34).

3.3.2.2. Attitude to cognitive interventions

Most participants had a positive attitude and a strong interest in cognitive interventions. They anticipated learning a myriad of better ways of cognitive brain function through the intervention, believing it would improve cognition (33). In addition, most participants appeared to appreciate the clarification of cognitive content, as information overload proved overwhelming as it introduced uncertainty and fears about being misled. The ability of health care providers to clarify this information made them feel reassured and helpful (34). Participants were not just interested in cognitive interventions but appeared willing and ready to participate (33). PWDs' attitudes were also influenced by the content and form of the intervention (33). Mainly, PWDs had intrinsic and extrinsic motivations to participate in the intervention. Encouragement and support from health care professionals also were important drivers (34).

However, significant barriers also existed, such as the time factor. People's attendance was adversely affected when course or group meeting times clashed with participants' working hours or appointments (33, 35). Additionally, some people had difficulty planning or implementing their plans, which prevented them from attending sessions on time (34). Others found it difficult to change habits. Consequently, this made the intervention ineffective and caused participants to lose faith in the intervention, and finally, drop out of the sessions (34).

3.3.2.3. Preference for cognitive interventions

PWDs were expected to learn better cognitive strategies to improve cognitive functioning (34, 37). A wide range of content was recommended as specific areas of interest, which included understanding how diabetes affects cognitive function, cognitive decline coping skills during diabetes self-management, discussing the association between diabetes-related stress and cognitive function, and learning how to integrate a "brain-healthy" lifestyle, especially as it relates to diet, in routine activities (33). Participants appeared keen to understand the link between diabetes and cognitive decline and be informed at the time of diagnosis for early preparation for the onset of cognitive problems (33). PWDs with

cognitive problems needed information about treatment options such as diet and medication (35). The studies also recommended focusing on teaching cognitive strategies that can improve quality of life (33). Patients' inattentiveness as a result of cognitive dysfunction creates a sense of helplessness over their illness and triggers anxiety, therefore exercises are required to improve these problems as a matter of priority such as meditation and deep breathing (34). Some people preferred a group format for interventions, as they wanted to "learn cognitive strategies from each other and share ideas" and acquire new knowledge (33).

4. Discussion

This is the first study that systematically reviewed the performances of disease management in PWDs during their cognitive decline. The findings showed that PWDs often experienced cognitive symptoms without recognizing pre-existing cognitive decline. In terms of self-response, they tried various methods to deal with cognitive decline with no certainty that these approaches could be effective. The cognitive interventions supported PWDs with increasing knowledge and practical strategies to better manage their disease.

In our findings, PWDs often perceived their own cognitive problems they were experiencing as normal part of aging, although some people did not fit this profile. Consistently with our findings, a review of the qualitative literature found that normal aging was the most common cause attribute to the self-perceived cognitive changes (21). Age, vascular and metabolic risk factors are related to mild cognitive impairment and dementia (38). As a most common metabolic risk factor, diabetes may aggravate cognitive decline with age, which is obviously ignored by PWDs. Lack of knowledge prevents them from recognizing cognitive decline in time and reduces the efficacy of self-management, including medication compliance, a proper diet and the application of new knowledge (39). Therefore, providing a timely education to PWDs may support them in preventing and managing the development of cognitive impairment.

Although PWDs reported many methods used to cope with cognitive decline, these methods appear to be applicable to various scenarios of daily life and not be diabetes-specific. Our results cannot reveal how PWDs adapt diabetes self-management strategies to cope with cognitive decline, which requires more in-depth research to explore. However, it may also suggest that PWDs cannot cope with the disruptions in diabetes management caused by cognitive decline on their own and they need external help and support.

Furthermore, based on our findings, PWDs with cognitive impairment did not receive adequate medical, educational, or emotional support from their health care providers. Although they adapted a variety of methods to help themselves cope with difficulties caused by cognitive impairment, support from health care providers means a lot to them. There are several possible reasons for this. First, a lack of awareness and comprehension of the co-occurrence of these disorders among health and social care providers might result in their inability to recognize and promptly treat cognitive impairment (40). Health and social care providers generally lack the awareness of the bidirectional relationship between diabetes management and cognitive impairment, resulting in an increased risk of diagnostic and treatment deficits (40, 41). Although the benefits of routine cognitive screening in PWDs have not been determined (42), health care providers should be alerted to memory complaints developed in PWDs or their caregivers. Thus, health care workers need to improve their understanding of the relationship between diabetes and cognitive impairment to reduce the impact of the disease and ameliorate clinical outcomes.

Secondly, although several guidelines have provided some relevant management recommendations, there is a lack of a comprehensive guidance for the clinical management of patients with diabetes and cognitive impairment. The American Diabetes Association (43), a UK Multidisciplinary National Expert Working Group (40), the American Association of Diabetes Educators (44) and the Chinese Medical Association (45) provide optimal practice guidance for healthcare professionals caring for patients with diabetes combined with cognitive impairment or dementia. Global guidelines from the International Diabetes Federation on managing older patients with type 2 diabetes provide the earliest suggestions for looking after patients with different functional deprivations, including frailty and dementia. While the guidelines have been well-received, their recommendations are not based on evidence of effectiveness in clinical practice (46). These guidelines need to be updated and improved as more contemporary evidence-based results become available.

Currently, there is no specific treatment plan for PWDs with mild cognitive impairment or dementia. The novel SGLT2 inhibitors have the potential to prevent and improve the cognitive decline associated with type 2 diabetes. The mechanisms underlying the development of cognitive impairment in PWDs have not been fully elucidated, but the available evidence suggests a possible combination of vascular damage, chronic inflammation and neurodegenerative pathology (11). Animal experiments showed that SGLT2 inhibitors have neuroprotective, anti-inflammatory, oxidative stress-reducing and anti-atherosclerotic effects (47). Carmen et al. found that empagliflozin reduced vascular damage and cognitive impairment in a mixed murine model of Alzheimer's disease and type 2 diabetes (48). However, we have little information on how SGLT2 inhibitors affect cognitive decline in clinical diabetes (49). Serena et al. found that a SGLT2 inhibitor was positively associated with better cognitive scores in a cohort of patients with diabetes (50). A prospective study showed significant beneficial effects of empagliflozin on cognitive and physical decline in frail older adults with diabetes and heart failure with preserved ejection fraction (51). New studies are needed to substantiate the benefits of SGLT2 inhibitors on cognitive impairment in people with type 2 diabetes.

Cognitive training was beneficial in PWDs. Cognitive training is a common non-pharmacological intervention to treat people with cognitive impairment (45). An eight-week, nurse-led study of a cognitive training intervention conducted in people with type 2 diabetes found that 58% of participants stated the intervention helped their diabetes self-management, and 74% expressed the desire to continue using the learned cognitive strategies (52). Another online cognitive intervention study

found that individuals with diabetes improved scores on self-management, cognition and self-efficacy, with an increased adherence to a proper diet and medications (53). These findings are consistent with our results. We showed that PWDs were particularly interested in cognitive training, as well as lifestyle interventions aimed at improving cognitive function. Cognitive training allows PWDs to gain a sense of accomplishment, learn new skills and reduce anxiety. In previous studies, cognitive training improved cognitive skills or daily activities in the average population with mild cognitive impairment or mild to moderate dementia (54, 55). However, some studies have shown different results. Wong et al. found that the combination of patient empowerment and cognitive training did not improve glycemic control or self-care activities in older PWDs with memory complaints (56). A systematic review found moderate strength of evidence that cognitive training may improve performance in trained cognitive domains (57). The reasons for these opposite findings may be due to different study participants and methodological discrepancies, including incompatible treatments and dissimilar treatment durations. Large-scale and high-quality studies are needed in the future to demonstrate the types of cognitive interventions that can be successfully utilized in clinical practice.

5. Strengths and limitations

To our knowledge, this is the first qualitative systematic review that specifically addresses cognitive problems in PWDs by identifying how they perceive and experience their cognitive problems and describing the impact of cognitive problems on their daily lives. The explicit and comprehensive search strategy reported quality appraisal of the included studies and data synthesis process. Nevertheless, the study had several limitations. The first is the small sample size of the included articles. However, the included articles were heterogeneous in design so they were representative of diverse patient populations although they cannot be generalized. Second, we included three studies published by Cuevas et al., suggesting a potential bias. However, we conducted a comprehensive literature search with a rigorous screening, ensuring the reliability of our results. The thematic analysis was an interpretative process and the outcomes were validated by the co-authors, thus there was the potential for other interpretations. Finally, the exclusion of languages other than English or Chinese meant relevant studies published in other languages may have been overlooked.

6. Conclusions

This study showed that cognitive problems often occur among PWDs, seriously affecting their self-management and daily life activities. This phenomenon has received little attention from healthcare professionals, with limited patient education or treatment interventions. Given the deleterious effects of cognitive impairment on PWDs, healthcare providers should focus more on cognitive performance, facilitating and supporting treatments and interventions for cognitive impairment. Health education in PWDs might help them self-monitor and identify cognitive impairment. New studies will explore effective cognitive interventions in large-scale trials.

Author contributions

YC, MW, and XG contributed to the study design, data acquisition, and analysis and manuscript revision. MW, XG, and JY contributed to data acquisition, interpretation of data, and manuscript revision. NM, RT, and XLi contributed to quality assessments, manuscript drafting, and revision. XLv and FY contributed to manuscript drafting and revision. YC is the guarantor of this work. All authors significantly contributed to the manuscript and approved the final version for publication.

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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.

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Supplementary material

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References

1. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, et al. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the International Diabetes Federation Diabetes Atlas, 9(th) edition. *Diab Res Clin Pract*. (2019) 157:107843. doi: 10.1016/j.diabres.2019.107843

2. Hong S, Pouya S, Suvi K, Moritz P, Katherine O, Bruce B D, et al. IDF Diabetes Atlas: Global, regional and country-level diabetes prevalence estimates for 2021 and projections for 2045. *Diabetes Res Clin Pract.* (2022) 183:109119. doi: 10.1016/j.diabres.2021.109119

3. An J, Li H, Tang Z, Zheng D, Guo J, Liu Y, et al. Cognitive impairment and risk of all-cause and cardiovascular disease mortality over 20-year follow-up: results from the BLSA. *J Am Heart Assoc*. (2018) 7:e008252. doi: 10.1161/JAHA.117.008252

4. Biessels GJ, Nobili F, Teunissen CE, Simó R, Scheltens P. Understanding multifactorial brain changes in type 2 diabetes: a biomarker perspective. *Lancet Neurol*. (2020) 19:699–710. doi: 10.1016/S1474-4422(20)30139-3

5. Rawlings AM, Sharrett AR, Albert MS, Coresh J, Windham BG, Power MC, et al. The association of late-life diabetes status and hyperglycemia with incident mild cognitive impairment and dementia: the ARIC study. *Diab Care*. (2019) 42:1248–54. doi: 10.2337/dc19-0120

6. Areosa Sastre A, Vernooij RW, Gonzalez-Colaco Harmand M, Martinez G. Effect of the treatment of Type 2 diabetes mellitus on the development of cognitive impairment and dementia. *Cochrane Database Syst Rev*. (2017) 6:CD003804. doi: 10.1002/14651858.CD003804.pub2

7. Cuevas H, Stuifbergen A. Perceived cognitive deficits are associated with diabetes self-management in a multiethnic sample. *J Diabetes Metab Disord*. (2017) 16:7. doi: 10.1186/s40200-017-0289-3

8. Hewitt J, Smeeth L, Chaturvedi N, Bulpitt CJ, Fletcher AE. Self management and patient understanding of diabetes in the older person. *Diabet Med*. (2011) 28:117–22. doi: 10.1111/j.1464-5491.2010.03142.x

9. Xu W, Hu X, Zhang X, Ling C, Wang C, Gao L. Cognitive impairment and related factors among middle-aged and elderly patients with type 2 diabetes from a bio-psycho-social perspective. *Diab Metab Syndrome Obes Targets Ther*. (2021) 14:4361–9. doi: 10.2147/DMSO.S333373

10. Dove A, Shang Y, Xu W, Grande G, Laukka EJ, Fratiglioni L, et al. The impact of diabetes on cognitive impairment and its progression to dementia. *Alzheimers Dement*. (2021) 17:1769–78. doi: 10.1002/alz.12482

11. Srikanth V, Sinclair AJ, Hill-Briggs F, Moran C, Biessels GJ. Type 2 diabetes and cognitive dysfunction-towards effective management of both comorbidities. *Lancet Diab Endocrinol*. (2020) 8:535–45. doi: 10.1016/S2213-8587(20)30118-2

12. Díaz-Venegas C, Schneider DC, Myrskylä M, Mehta NK. Life expectancy with and without cognitive impairment by diabetes status among older Americans. *PLoS ONE*. (2017) 12:e0190488. doi: 10.1371/journal.pone.0190488

13. Force USPST, Owens DK, Davidson KW, Krist AH, Barry MJ, Cabana M, et al. Screening for cognitive impairment in older adults: US preventive services task force recommendation statement. *JAMA*. (2020) 323:757–63. doi: 10.1001/jama.2020.0435

14. Jamali A, Shahrbanian S, Morteza Tayebi S. The effects of exercise training on the brain-derived neurotrophic factor (BDNF) in the patients with type 2 diabetes: a systematic review of the randomized controlled trials. *J Diab Metab Disord*. (2020) 19:633–43. doi: 10.1007/s40200-020-00529-w

15. Min Jung K, Fritschi C. Relationships between cognitive impairment and self-management in older adults with type 2 diabetes: an integrative review. *Res Gerontol Nurs*. (2021) 14:104–12. doi: 10.3928/19404921-20201117-01

16. Omori K, Kawamura T, Urata M, Matsuura M, Kusama M, Imamine R, et al. Effect of re-coaching on self-injection of insulin in older diabetic patients - impact of cognitive impairment. *Diab Res Clin Pract*. (2017) 130:34–42. doi: 10.1016/j.diabres.2017.05.011

17. Sinclair AJ, Girling AJ, Bayer AJ. Cognitive dysfunction in older subjects with diabetes mellitus: impact on diabetes self-management and use of care services. All Wales Research into Elderly (AWARE) Study. *Diab Res Clin Pract*. (2000) 50:203–12. doi: 10.1016/S0168-8227(00)00195-9

18. Anderson K, Willmore C, Doran E, Oki N, Vonnahme J, Gates BJ. Cognitive and literacy screening as predictors of ability to fill a pillbox using two pillbox assessment scoring methods. *Consult Pharm*. (2014) 29:304–16. doi: 10.4140/TCP.n.2014.304

19. Rosen MI, Beauvais JE, Rigsby MO, Salahi JT, Ryan CE, Cramer JA. Neuropsychological correlates of suboptimal adherence to metformin. *J Behav Med*. (2003) 26:349–60. doi: 10.1023/A:1024257027839

20. Feil DG, Zhu CW, Sultzer DL. The relationship between cognitive impairment and diabetes self-management in a population-based community sample of older adults with Type 2 diabetes. *J Behav Med*. (2012) 35:190–9. doi: 10.1007/s10865-011-9344-6

21. Buckley RF, Saling MM, Frommann I, Wolfsgruber S, Wagner M. Subjective cognitive decline from a phenomenological perspective: a review of the qualitative literature. *J Alzheimers Dis*. (2015) 48(Suppl. 1):S125–40. doi: 10.3233/JAD-150095

22. Miebach L, Wolfsgruber S, Frommann I, Fliessbach K, Jessen F, Buckley R, et al. Cognitive complaints in memory clinic patients and in depressive patients: an interpretative phenomenological analysis. *Gerontologist*. (2019) 59:290–302. doi: 10.1093/geront/gnx208

23. Denny E, Weckesser A. Qualitative research: what it is and what it is not: study design: qualitative research. *BJOG*. (2019) 126:369. doi: 10.1111/1471-0528.15198

24. Harman G, Clare L. Illness representations and lived experience in early-stage dementia. *Qual Health Res*. (2006) 16:484–502. doi: 10.1177/1049732306286851

25. Clare L, Rowlands J, Bruce E, Surr C, Downs M. 'I don't do like I used to do': a grounded theory approach to conceptualising awareness in people with moderate to severe dementia living in long-term care. *Soc Sci Med*. (2008) 66:2366–77. doi: 10.1016/j.socscimed.2008.01.045

26. Johansson MM, Marcusson J, Wressle E. Cognitive impairment and its consequences in everyday life: experiences of people with mild cognitive impairment or mild dementia and their relatives. *Int Psychogeriatr*. (2015) 27:949–58. doi: 10.1017/S1041610215000058

27. Koppara A, Wagner M, Lange C, Ernst A, Wiese B, König HH, et al. Cognitive performance before and after the onset of subjective cognitive decline in old age. *Alzheimers Dement*. (2015) 1:194–205. doi: 10.1016/j.dadm.2015.02.005

28. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *PLoS Med*. (2021) 18:e1003583. doi: 10.1371/journal.pmed.1003583

29. Thomas J, Harden A. Methods for the thematic synthesis of qualitative research in systematic reviews. *BMC Med Res Methodol*. (2008) 8:45. doi: 10.1186/1471-2288-8-45

30. Hasseler MK, Von Der Heide M, Indefrey S. Resources for and barriers to effective diabetes care management- experiences and perspectives of people with type 2 diabetes [Article]. *J Public Health*. (2011) 19:65–71. doi: 10.1007/s10389-010-0354-6

31. Wilson V. Evaluation of the care received by older people with diabetes. *Nurs Older People*. (2012) 24:33–7. doi: 10.7748/nop2012.05.24.4.33.c9071

32. Speight J, Barendse SM, Singh H, Little SA, Rutter MK, Heller SR, et al. Cognitive, behavioural and psychological barriers to the prevention of severe hypoglycaemia: a qualitative study of adults with type 1 diabetes. *SAGE Open Med*. (2014) 2:2050312114527443. doi: 10.1177/2050312114527443

33. Cuevas HE, Stuifbergen AK, Brown SA, Rock JL. Thinking about cognitive function: perceptions of cognitive changes in people with type 2 diabetes. *Diabetes Educ*. (2017) 43:486–94. doi: 10.1177/0145721717729806

34. Cuevas HE, Stuifbergen AK, Ward C. Participant perspectives of cognitive rehabilitation for type 2 diabetes: expectations and impact. *J Aging Res*. (2018) 2018:6563457. doi: 10.1155/2018/6563457

35. Hu Y, Zhang W. Qualitative research on diabetes patients experiencing mild cognitive impairment events. *J Nurs Rehabil*. (2019) 18:9–12. doi: 10.3969/j.issn.1671-9875.2019.06.003

36. Chepulis L, Morison B, Cassim S, Norman K, Keenan R, Paul R, et al. Barriers to diabetes self-management in a subset of New Zealand adults with type 2 diabetes and poor glycaemic control. *J Diabetes Res*. (2021) 2021:5531146. doi: 10.1155/2021/5531146

37. Cuevas H, Zuniga J. Latinx with type 2 diabetes: perceptions of cognitive health. *J Immigr Minor Health*. (2021) 23:337–43. doi: 10.1007/s10903-020-00995-7

38. Kivipelto M, Mangialasche F, Ngandu T. Lifestyle interventions to prevent cognitive impairment, dementia and Alzheimer disease. *Nat Rev Neurol*. (2018) 14:653–66. doi: 10.1038/s41582-018-0070-3

39. Smalls BL, Walker RJ, Hernandez-Tejada MA, Campbell JA, Davis KS, Egede LE. Associations between coping, diabetes knowledge, medication adherence and self-care behaviors in adults with type 2 diabetes. *Gen Hosp Psychiatry*. (2012) 34:385–9. doi: 10.1016/j.genhosppsych.2012.03.018

40. Sinclair AJ, Hillson R, Bayer AJ, National Expert Working G. Diabetes and dementia in older people: a Best Clinical Practice Statement by a multidisciplinary National Expert Working Group. *Diab Med*. (2014) 31:1024–31. doi: 10.1111/dme.12467

41. Ojo O, Brooke J. Evaluating the association between diabetes, cognitive decline and dementia. *Int J Environ Res Public Health*. (2015) 12:8281–94. doi: 10.3390/ijerph120708281

42. Patnode CD, Perdue LA, Rossom RC, Rushkin MC, Redmond N, Thomas RG, et al. Screening for cognitive impairment in older adults: an evidence update for the U.S. preventive services task force. *JAMA*. (2020) 323:764–85. doi: 10.1001/jama.2019.22258

43. Young-Hyman D, de Groot M, Hill-Briggs F, Gonzalez JS, Hood K, Peyrot M. Psychosocial care for people with diabetes: a position statement of the american diabetes association. *Diab Care*. (2016) 39:2126–40. doi: 10.2337/dc16-2053

44. Gonzalvo JD, Hamm J, Eaves S, Muñoz CE, De Groot M, Hill-Briggs F, et al. A practical approach to mental health for the diabetes educator. *AADE Pract*. (2019) 7:29–44. doi: 10.1177/2325160319826929

45. Branch CMAE. Expert consensus on diabetic cognitive dysfunction. *Chin J Diab*. (2021) 13:678–94. doi: 10.3760/cma.j.cn115791-20210527-00291

46. Dunning T, Sinclair A, Colagiuri S. New IDF Guideline for managing type 2 diabetes in older people. *Diab Res Clin Pract*. (2014) 103:538–40. doi: 10.1016/j.diabres.2014.03.005

47. Pawlos A, Broncel M, Wozniak E, Gorzelak-Pabiś P. Neuroprotective effect of SGLT2 inhibitors. *Molecules*. (2021) 26:7213. doi: 10.3390/molecules26237213

48. Hierro-Bujalance C, Infante-Garcia C, Del Marco A, Herrera M, Carranza-Naval MJ, Suarez J, et al. Empagliflozin reduces vascular damage and cognitive impairment in a mixed murine model of Alzheimer's disease and type 2 diabetes. *Alzheimers Res Ther*. (2020) 12:40. doi: 10.1186/s13195-020-00607-4

49. Rizzo MR, Di Meo I, Polito R, Auriemma MC, Gambardella A, di Mauro G, et al. Cognitive impairment and type 2 diabetes mellitus: focus of SGLT2 inhibitors treatment. *Pharmacol Res*. (2022) 176:106062. doi: 10.1016/j.phrs.2022.106062

50. Low S, Goh KS, Ng TP, Moh A, Ang SF, Wang J, et al. Association between use of sodium-glucose co-transporter-2 (SGLT2) inhibitors and cognitive function in a longitudinal study of patients with type 2 diabetes. *J Alzheimers Dis*. (2022) 87:635–42. doi: 10.3233/JAD-215678

51. Mone P, Lombardi A, Gambardella J, Pansini A, Macina G, Morgante M, et al. Empagliflozin improves cognitive impairment in frail older adults with type 2 diabetes and heart failure with preserved ejection fraction. *Diab Care*. (2022) 45:1247–51. doi: 10.2337/dc21-2434

52. Cuevas HE, Stuifbergen AK, Brown SA, Ward C. A nurse-led cognitive training intervention for individuals with type 2 diabetes. *Res Gerontol Nurs*. (2019) 12:203–12. doi: 10.3928/19404921-20190612-01

53. Cuevas H, Carter S. Online cognitive training: an adaptation of the memory, attention, and problem solving skills for persons with diabetes intervention. *Comp Inform Nurs*. (2020) 39:162–9. doi: 10.1097/CIN.0000000000000663

54. Bahar-Fuchs A, Martyr A, Goh AM, Sabates J, Clare L. Cognitive training for people with mild to moderate dementia. *Cochrane Database Syst Rev*. (2019) 3:Cd013069. doi: 10.1002/14651858.CD013069.pub2

55. Martin M, Clare L, Altgassen AM, Cameron MH, Zehnder F. Cognition-based interventions for healthy older people and people with mild cognitive impairment. *Cochrane Database Syst Rev*. (2011) 2011:Cd006220. doi: 10.1002/14651858.CD006220.pub2

56. Wong CW, Wai-Tsun William O, Wong KS, Ma R, Hui E, Kwok CT. Randomized trial of a patient empowerment and cognitive training program for older people with diabetes mellitus and cognitive impairment. *Geriatr Gerontol Int*. (2020) 20:1164–70. doi: 10.1111/ggi.14062

57. Kane RL, Butler M, Fink HA, Brasure M, Davila H, Desai P, et al. *Interventions To Prevent Age-Related Cognitive Decline, Mild Cognitive Impairment, and Clinical Alzheimer's-Type Dementia. Comparative Effectiveness Review No. 188. (Prepared by the Minnesota Evidence-based Practice Center under Contract No. 290-2015-00008-I.) AHRQ Publication No. 17-EHC008-EF*. Rockville, MD: Agency for Healthcare Research and Quality (2017). doi: 10.23970/AHRQEPCCER188

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Lifestyle intervention reduces risk score for cardiovascular mortality in company employees with pre-diabetes or diabetes mellitus – A secondary analysis of the PreFord randomized controlled trial with 3 years of follow-up

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Aim: To evaluate the effects of a multimodal intervention (including exercise training, psychosocial interventions, nutrition coaching, smoking cessation program, medical care) on the health and long-term cardiovascular disease (CVD) mortality risk of company employees with pre-diabetes or diabetes mellitus (DM) at high CVD risk.

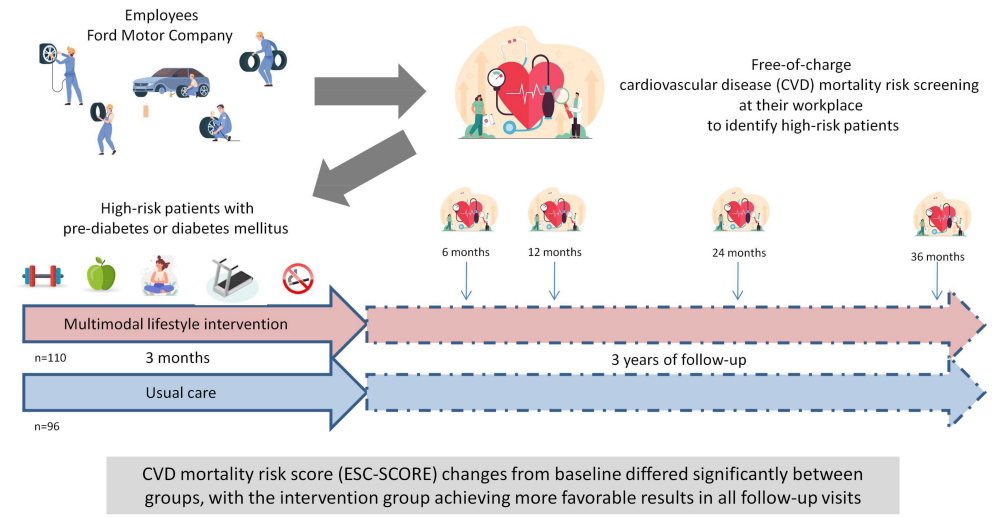
Methods: In the PreFord study, German company employees (n=4196) participated in a free-of-charge CVD mortality risk screening at their workplace. Based on their European Society of Cardiology – Systematic Coronary Risk Evaluation score (ESC-SCORE), they were subdivided into three risk groups. High-risk patients (ESC-SCORE≥5%) were randomly assigned to a 15-week lifestyle intervention or usual care control group. Data from patients with pre-DM/DM were analyzed intention-to-treat (ITT: n=110 versus n=96) and per protocol (PP: n=60 versus n=52).

Results: Body mass index, glycated hemoglobin, total cholesterol, low-density lipoprotein, triglyceride levels as well as systolic and diastolic blood pressure improved through the intervention (ITT, PP: p<0.001). The ESC-SCORE markedly decreased from pre- to post-intervention (ITT, PP: p<0.001). ESC-SCORE changes from baseline differed significantly between the groups, with the intervention group achieving more favorable results in all follow-up visits 6, 12, 24 and 36 months later (at each time point: ITT: p<0.001; PP: p ≤ 0.010).

Conclusion: The study demonstrates the feasibility of attracting employees with pre-DM/DM at high CVD mortality risk to participate in a multimodal lifestyle program following a free CVD mortality risk screening at their workplace. The lifestyle intervention used in the PreFord study shows high potential for improving health of company employees with pre-DM/DM in the long term. ISRCTN23536103.

KEYWORDS

exercise, nutrition, cardiovascular risk assessment, employees, diabetes



GRAPHICAL ABSTRACT

1 Introduction

Cardiovascular diseases (CVDs) are the leading cause of premature death (1, 2). Thus, reducing the incidence of CVDs is of high public health importance. A meta-analysis from observational studies has shown that a healthy lifestyle can reduce the risk of developing CVDs by up to 66% (3). Preventive measures aimed at lifestyle changes can therefore be helpful to reduce individual mortality risk. In the PreFord study (4), German company employees of the Ford Motor Company (n=4196) participated in a free-of-charge CVD mortality risk screening at their workplace. The participants were then subdivided into three risk groups based on their risk factors, quantified by the European Society of Cardiology – Systematic Coronary Risk Evaluation score (ESC-SCORE), which is an established metric to estimate the risk of fatal cardiovascular events with a high accuracy for Germans and other Europeans (5, 6). Employees with a high risk score (ESC-SCORE ≥ 5%) were randomly assigned to a multimodal lifestyle intervention group (receiving exercise training, psychosocial interventions, nutrition coaching, smoking cessation program, medical care) or to a usual care group (receiving medical care only).

Large-scale observational studies show that patients with diabetes mellitus (DM) have a drastically increased risk of cardiovascular events and CVD mortality (7–9). As lifestyle changes can help reduce cardiovascular risk, patients with pre-DM and DM should optimize their lifestyle as early as possible. Unfortunately, these patients are often very difficult to motivate for lifestyle changes; moreover, there might be several psycho-social barriers (10). When they participate in an intervention program, achieving sustainable effects is usually challenging, due to low program adherence and high drop-out rates (11).

This secondary analysis of the PreFord study data explores the direct effects of the study's 15-week multimodal lifestyle intervention on the ESC-SCORE and other health-related variables in the pre-DM/DM subgroup. Long-term effects on the individual cardiovascular risks and the program's efficiency for patients with pre-DM/DM are discussed, considering that aggressive programs for lifestyle changes are urgently needed to account for an increasing incidence and prevalence of DM (12, 13).

2 Methods

2.1 PreFord study

2.1.1 Study design

The PreFord trial was designed as a randomized controlled, multicenter clinical study. The study design has already been

described in detail (4). The study protocol in line with good clinical practice has been approved by the Ethics Committee of the University of Cologne (ref: 03-217) and the Ethics Committee of the North Rhine Medical Association (Ärztekammer Nordrhein, ref: 2004079). Subjects gave their written informed consent prior to the start of the study.

2.1.2 Subjects

Employees of the Ford Motor Company Germany (>15.000) were invited to participate in a free-of-charge cardiovascular medical check-up (T0) and to determine their ESC-SCORE which reflects personal risk of cardiovascular events. The score was calculated by an independent statistics institution (Institute of Medical Statistics, Informatics and Epidemiology, University of Cologne). Age, blood pressure, smoking habits and total cholesterol values were recorded for risk assessment. Inclusion criteria were defined as follows: an ESC-SCORE ≥ 5% (high-risk group) and the ability to exercise. Exclusion criteria were defined as follows: exercise-limiting diseases, history of cardiovascular disease, cancer, pregnancy or severe mental disorders.

2.1.3 Lifestyle intervention

Subjects were randomly assigned to the intervention (INT) group or the usual care control (CON) group by block randomization 1:1. The computer-generated random list was provided by the Clinical Trial Center Cologne. Study personnel assigned participants to the INT or CON group according to this random list. The 15-week multimodal lifestyle intervention (Table 1) was supervised by professional health care specialists (medical doctors, exercise physiologists, psychologists, and nutritional coaches). The intervention was performed in small groups twice a week for 2.5-3 hours per session in two rehabilitation centers in Cologne, Germany. Further details about the program are available in the publication of Gysan et al. (4). All employees who participated in the intervention program were examined immediately after the intervention (T1). The CON group participants received usual care from their general practitioners.

2.1.4 Follow-up

All company employees who participated in the study, in either the INT or ON group, were invited for follow-up medical check-ups 6 (T2), 12 (T3), 24 (T4) and 36 (T5) months after start of the study. The study ended after completion of the last follow-up.

2.2 Secondary data analysis

2.2.1 Subjects

The secondary data analysis is reported in accordance with the CONSORT statement (14). Only employees diagnosed with diabetes mellitus (and receiving pharmacological treatment) and/or with glycated hemoglobin (HbA1c) levels ≥ 5.7% were included in this analysis (Figure 1). In total, the datasets of n=142 persons with pre-DM (HbA1c levels ≥ 5.7% and < 6.5% without anti-diabetic medication) and n=64 patients with manifest DM (HbA1c levels ≥ 6.5% and/or treated with anti-diabetic medication) were considered. The HbA1c thresholds correspond to the American Diabetes Association cutoffs for the diagnoses of pre-DM and DM (15).

2.2.2 Primary and secondary outcomes

The ESC-SCORE was defined as the primary outcome. It was determined in the INT and CON group at every follow-up examination and thus helped assess the long-term effectiveness of the intervention. The same ESC-SCORE algorithm in its initially published form was used throughout the study (5). The ESC-SCORE provides an accurate prediction of cardiovascular events in Europeans without a history of severe cardiovascular diseases (e.g., coronary heart disease, stroke, peripheral artery disease, heart failure, heart arrhythmia).

To determine the intervention's direct effectiveness, body weight, body mass index (BMI), waist circumference, glycated hemoglobin (HbA1c), high-sensitive C-reactive protein (hsCRP), total cholesterol, high-density lipoprotein (HDL), low-density lipoprotein (LDL), triglycerides, systolic and diastolic blood pressure and exercise capacity pre- and post-intervention were defined as secondary outcomes.

2.2.3 Statistical analyses

Data are presented as mean values ± standard deviations (SD) and 95% confidence intervals (95%-CI). The "SPSS" program (v. 28.0, IBM Corporation, Armonk, New York, USA) was used for the

TABLE 1 Lifestyle intervention.

Components	Hours planned	Subgroup data: Actual time spent (percentage of planned hours)
Aerobic endurance and resistance training	37.00	90.0%
Nutrition coaching Information/Education in Mediterranean-style diet and practical training in preparing a meal	11.00	73.1%
LifeSkills according to Williams and Williams	13.50	74.5%
Progressive relaxation training	6.00	90.7%
Smoking cessation program	0.45	11.1% (5 persons)
Medical care with guideline-based pharmacotherapy	4.75	90.7%
Information/Education Healthy lifestyle management	8.00	121.9%

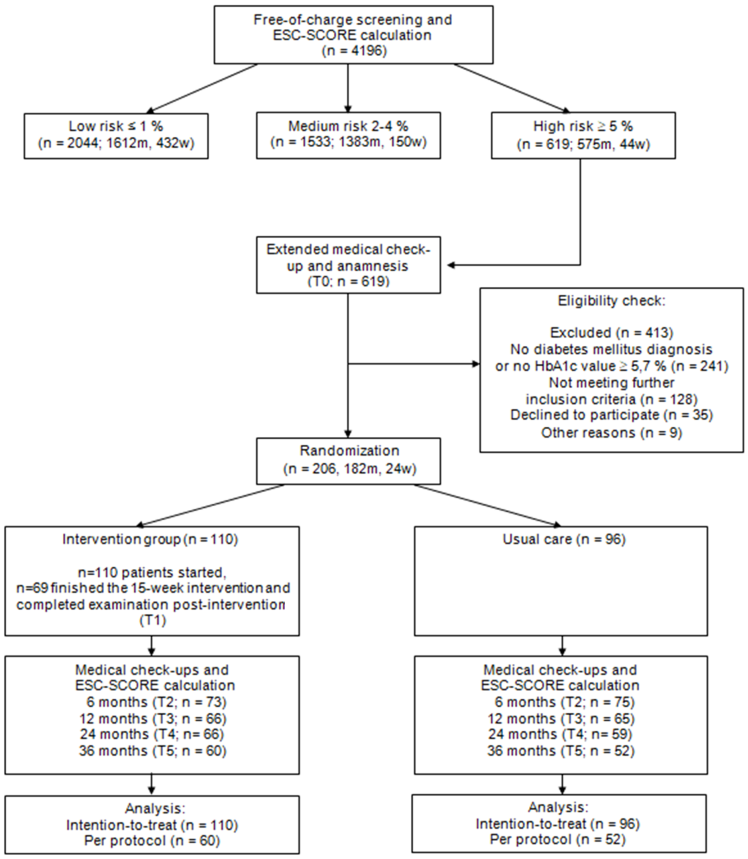


FIGURE 1
Study flow chart.

statistical analyses. Parametric tests were used throughout. When assumptions were violated and when appropriate, non-parametric (rank-based) hypotheses tests were conducted. For baseline comparisons of interval-scaled variables between the two groups, the Student's t test or the Mann-Whitney U test for unpaired samples were performed. The Chi^2 test was used to assess differences in the distribution of nominal-scaled variables between the groups. For pre-post-comparisons of interval-scaled variables within the INT group, the Student's t test or the Wilcoxon signed rank test for paired samples were used. For follow-up analyses within each group (INT and CON), the Friedman test was carried out. To compare changes from baseline between the two groups at the different follow-up time points, the Student's t test or the Mann-Whitney U test for unpaired samples were used. Data were analyzed intention-to-treat and per protocol. The intention-to-treat cohort included all patients. Missing values in the intention-to-treat analysis were replaced by the last observation carried forward (29.9% missing ESC-SCORE data, 33.2% missing HbA1c data). The per protocol cohort included only those patients who fully adhered to the study protocol. In addition, data from all measurement time points had to be available. Significance was considered at $p \leq 0.05$.

2.2.4 Sample size and power calculation

A sample size calculation was performed for the original study *a priori* (4). For this subgroup data analysis, a second power analysis was performed for the ESC-SCORE as the primary outcome *a posteriori* using G Power (v. 3.1.9.7., University of Düsseldorf, Düsseldorf, Germany). For the intention-to-treat analysis, a power of 100% was calculated for the comparison between the ESC-SCORE pre- and post-intervention of the INT group and a power of 94% for the comparison of ESC-SCORE changes from baseline between the INT and CON groups during the follow-up medical check-up 36 months later. For the per protocol analysis, statistical power values of 100% and 83% were calculated, respectively.

3 Results

3.1 Baseline data

The baseline (T0) data of the subjects of the INT and CON groups are presented in Table 2. The ratio of men and women roughly reflects the ratio of employees in the company. The groups were almost perfectly matched for the ESC-SCORE and also did not significantly differ in any other variable, except BMI.

3.2 Direct effects of the multimodal lifestyle intervention on the ESC-SCORE and important health variables

Pre-post-intervention data (T0-T1) are presented in Table 3. The ESC-SCORE decreased significantly, irrespective of the type of analysis

conducted (intention-to-treat or per protocol). Nearly all other health-related variables (body weight, BMI, waist circumference, HbA1c, total cholesterol, LDL, triglycerides, systolic and diastolic blood pressure, exercise capacity) also improved significantly. HDL levels remained unchanged and hsCRP levels increased significantly, but very slightly.

3.3 Follow-up and long-term effects of the multimodal lifestyle intervention on the ESC- SCORE

There was a significant overall time effect for the ESC-SCORE in each group (INT and CON) from T0 across all follow-up time points (T2, T3, T4, T5) (Friedman test: $p<0.001$), which was evident in both the intention-to-treat and per protocol analyses (Supplemental Data File: ESM 1). It appears that the ESC-SCORE of the INT group increased only very slightly in the long term after the intervention, while it increased more in the CON group. This is reflected in the changes from baseline. The delta values differed significantly between the groups (INT and CON) at each time point (T2, T3, T4, T5), with the intervention group achieving more favorable results in the intention-to-treat (Figure 2) as well as the per protocol analysis (Figure 3).

To clarify whether there is a difference in the primary outcome between pre-DM and DM patients, a further subgroup analysis was performed for ESC-SCORE changes (Supplemental Data File: ESM 2). The intention-to-treat analysis revealed that the pre-DM patients'

TABLE 2 Study participants´ baseline (T0) characteristics.

	Intervention group n=110	Usual care group n=96	p-value
ESC-SCORE result [%]	8.07 ± 5.17 (7.09-9.05)	8.03 ± 4.83 (7.06-9.01)	0.959 ◊
Sex [m/f, n]	96/14	86/10	0.606 †
Age [years]	60.1 ± 8.7 (58.4-61.7)	60.2 ± 7.7 (58.6-61.7)	0.994 ◊
Body weight [kg]	89.6 ± 15.3 (86.7-92.4)	86.4 ± 14.3 (83.5-89.3)	0.076 ◊
BMI [kg/m^2]	29.62 ± 4.55 (28.76-30.48)	28.19 ± 3.85 (27.41-28.97)	0.019 ◊
Waist circumference [cm]	103.8 ± 10.6 (101.8-105.8) n=109	101.0 ± 12.4 (98.5-103.5) n=95	0.083 #
HbA1c [%]	6.41 ± 0.86 (6.25-6.57)	6.18 ± 0.57 (6.06-6.29)	0.103 ◊
hsCRP [mg/l]	0.31 ± 0.56 (0.20-0.41) n=108	0.33 ± 0.54 (0.22-0.44) n=95	0.752◊
Total cholesterol [mg/dl]	238.9 ± 48.5 (229.7-248.1)	237.4 ± 48.5 (227.6-247.2)	0.908 ◊
HDL [mg/dl]	53.5 ± 12.3 (51.1-55.8)	54.8 ± 12.8 (52.2-57.4)	0.338 ◊
LDL [mg/dl]	150.6 ± 34.4 (144.1-157.1)	149.7 ± 33.6 (142.9-156.5)	0.928 ◊
Triglycerides [mg/dl]	218.0 ± 160.2 (187.7-248.2)	204.5 ± 138.6 (176.5-232.6)	0.982 ◊
Systolic BP [mmHg]	139.8 ± 17.7 (136.4-143.1)	138.1 ± 15.0 (135.1-141.2)	0.417 ◊
Diastolic BP [mmHg]	87.9 ± 11.1 (85.8-90.0)	88.7 ± 9.9 (86.7-90.7)	0.720 ◊
Exercise capacity [W/kg]	1.73 ± 0.47 (1.64-1.82) n=103	1.69 ± 0.43 (1.60-1.78) n=86	0.715 #
Smokers			
Non-smokers	42 (38.2%)	39 (40.6%)	
Current smokers	23 (20.9%)	21 (21.9%)	0.866 †
Ex-smokers	45 (40.9%)	36 (37.5%)	
Anti-diabetic drugs			
Insulin	8 (7.3%)	10 (10.4%)	0.425 †
Oral antidiabetic agents	19 (17.3%)	9 (9.4%)	0.099 †
Other drugs			
ASS	13 (11.8%)	20 (20.8%)	0.078 †
Statins	26 (23.6%)	15 (15.6%)	0.151 †
Anti-hypertensive agents	55 (50.0%)	36 (37.5%)	0.072 †

ESC-SCORE, European Society of Cardiology Systematic Coronary Risk Evaluation; BMI, body mass index; HbA1c, glycated hemoglobin; hsCRP, high-sensitive C-reactive protein; HDL, high-density lipoprotein; LDL, low-density lipoprotein; BP, blood pressure. Means ± standard deviations (SD) and 95% confidence intervals. ◊ Mann-Whitney U test # Student´s t test (unpaired samples) † Chi^2 test.

TABLE 3 Study participants´ characteristics pre (T0) and post (T1) -intervention.

	Intention-to-treat analysis Intervention group Pre-intervention n=110	Intention-to-treat analysis Intervention group Post-intervention n=110	p-value	Per protocol analysis Intervention group Pre-intervention n=69	Per protocol analysis Intervention group Post-intervention n=69	p-value
ESC-SCORE result [%]	8.07 ± 5.17 (7.09-9.05)	6.33 ± 4.31 (5.52-7.15)	<0.001 ○	8.62 ± 5.29 (7.35-9.89)	5.85 ± 3.87 (4.92-6.78)	<0.001 ○
Body weight [kg]	89.6 ± 15.3 (86.7-92.4)	87.8 ± 15.1 (85.0-90.7)	<0.001 ■	86.8 ± 14.7 (83.2-90.3)	84.0 ± 13.7 (80.7-87.3)	<0.001 ■
BMI [kg/m^2]	29.62 ± 4.55 (28.76-30.48)	29.02 ± 4.36 (28.20-29.84)	<0.001 ○	29.08 ± 4.46 (28.01-30.16)	28.13 ± 3.97 (27.17-29.08)	<0.001 ○
Waist circumference [cm]	103.8 ± 10.6 (101.8-105.8) n=109	101.9 ± 10.4 (99.9-103.9) n=109	<0.001 ■	102.0 ± 10.7 (99.4-104.6) n=68	99.0 ± 9.7 (96.6-101.3) n=68	<0.001 ■
HbA1c [%]	6.41 ± 0.86 (6.25-6.57)	6.26 ± 0.87 (6.09-6.42)	<0.001 ○	6.29 ± 0.80 (6.09-6.48)	6.04 ± 0.75 (5.86-6.22)	<0.001 ○
hsCRP [mg/l]	0.31 ± 0.56 (0.20-0.41) n=108	0.32 ± 0.64 (0.20-0.41) n=108	0.035 ○	0.24 ± 0.33 (0.16-0.32) n=67	0.26 ± 0.52 (0.14-0.39) n=67	0.035 ○
Total cholesterol [mg/dl]	238.9 ± 48.5 (229.7-248.1)	219.7 ± 49.5 (210.3-229.0)	<0.001 ○	232.2 ± 44.2 (221.6-242.9)	201.5 ± 36.6 (192.8-210.3)	<0.001 ○
HDL [mg/dl]	53.5 ± 12.3 (51.1-55.8)	53.8 ± 13.0 (51.3-56.2)	0.625 ○	54.8 ± 12.8 (51.7-57.9)	55.3 ± 13.8 (52.0-58.6)	0.625 ○
LDL [mg/dl]	150.6 ± 34.4 (144.1-157.1)	134.3 ± 34.3 (127.9-140.8)	<0.001 ○	147.6 ± 33.6 (139.5-155.6)	121.6 ± 26.4 (115.3-128.0)	<0.001 ○
Triglycerides [mg/dl]	218.0 ± 160.2 (187.7-248.2)	188.6 ± 149.5 (160.3-216.8)	<0.001 ○	191.2 ± 130.2 (159.9-222.4)	144.3 ± 90.7 (122.5-166.1)	<0.001 ○
Systolic BP [mmHg]	139.8 ± 17.7 (136.4-143.1)	132.7 ± 13.9 (130.1-135.3)	<0.001 ■	142.5 ± 19.2 (137.8-147.1)	131.2 ± 13.6 (127.9-134.5)	<0.001 ■
Diastolic BP [mmHg]	87.9 ± 11.1 (85.8-90.0)	84.5 ± 9.6 (82.7-86.3)	<0.001 ○	88.5 ± 11.7 (85.7-91.3)	83.1 ± 9.1 (80.9-85.3)	<0.001 ■
Exercise capacity [W/kg]	1.73 ± 0.47 (1.64-1.82) n=103	1.89 ± 0.50 (1.80-1.99) n=103	<0.001 ○	1.78 ± 0.51 (1.66-1.91) n=65	2.04 ± 0.50 (1.92-2.17) n=65	<0.001 ■

ESC-SCORE, European Society of Cardiology Systematic Coronary Risk Evaluation; BMI, body mass index; HbA1c, glycated hemoglobin; hsCRP, high-sensitive C-reactive protein; HDL, high-density lipoprotein; LDL, low-density lipoprotein; BP, blood pressure. Means ± standard deviations (SD) and 95% confidence intervals. ○ Wilcoxon signed rank test ■ Student´s t test (paired samples).

(INT: n=72, CON: n=70) results were quite similar to those of all patients (pre-DM/DM patients). Delta values differed significantly between the groups (INT and CON) at each time point (T2, T3, T4, T5), with the intervention group achieving more favorable results. In DM patients (INT: n=38, CON: n=26), a significant difference in ESC-SCORE changes was evident after the lifestyle intervention, with better results in the INT group. However, from T3 onward, there was no longer a significant difference in delta values between the groups (INT and CON). It should be noted that ESC-SCORE baseline values were significantly higher in pre-DM than in DM patients in both groups (INT: pre-DM: 9.16 ± 4.99% (95%-CI: 7.99-10.33%), DM: 6.00 ± 4.95% (95%-CI: 4.38-7.63%), U test: p<0.001; CON: pre-DM: 8.54 ± 4.12% (95%-CI: 7.55-9.52%), DM: 6.67 ± 6.24% (95%-CI: 4.15-9.20%), U test: p=0.017). Due to the small number of included DM patients (INT: n=15, CON: n=12), no subgroup analysis was performed in the per protocol cohort.

3.4 Follow-up and long-term effects of the multimodal lifestyle intervention on glycemic control

There was a significant overall time effect for the HbA1c levels in the INT group from T0 across all follow-up time points (T2,T3,T4,T5) (Friedman test: p<0.001), which was evident in both the intention-to-treat and per protocol analyses (Supplemental Data File: ESM 3). There were no significant HbA1c changes in the CON group. Of all pre-DM patients from the per protocol cohort, 5% developed manifest DM in the INT and 22% in the CON group (from T0 to T5). Half of them started treatment with anti-diabetic medication.

3.5 Adverse events during the intervention

There were no adverse events during the intervention.

4 Discussion

DM can drastically increase the risk of CVDs. The INTERHEART study, which collected data from more than 27,000 subjects in 52 countries, identified DM as a strong risk factor for acute myocardial infarction (9). Other famous large-scale studies such as the Framingham study or the San Antonio Heart Study found increased CVD mortality rates in DM patients compared with non-diabetic subjects from the general population (7, 8). Furthermore, the understanding of the pathogenesis of CVDs in the context of DM improves continuously, with hyperglycemia, hyperinsulinemia and

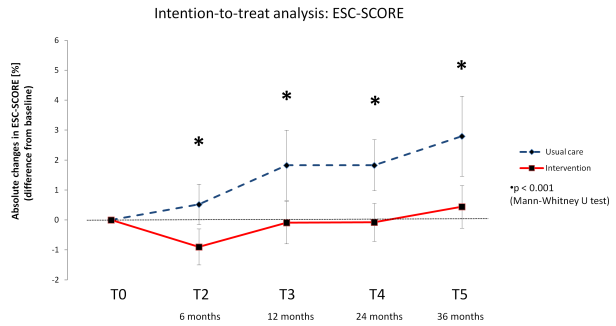


FIGURE 2
Delta values of the European Society of Cardiology – Systematic Coronary Risk Evaluation score (ESC-SCORE) – Intention-to-treat analysis. Means with 95% confidence intervals.

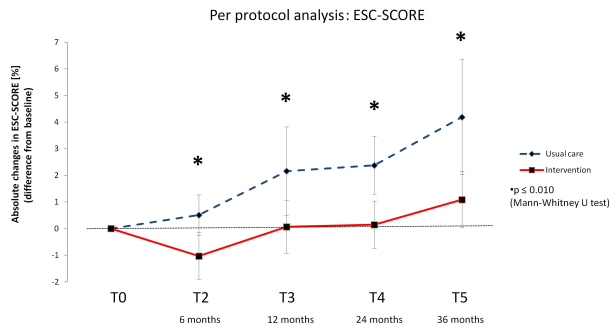


FIGURE 3
Delta values of the European Society of Cardiology – Systematic Coronary Risk Evaluation score (ESC-SCORE) –Per protocol analysis. Means with 95% confidence intervals.

hypercoagulability playing important roles in increased CVD risk and mortality (16, 17). Lifestyle interventions that can prevent the development of CVDs or that have a positive effect on their progression should therefore be strongly recommended as preventive measures not only for patients with manifest DM, but also for those with pre-DM (18, 19).

The secondary data analysis of the PreFord study shows that the cardiovascular risk of persons with pre-DM/DM can be substantially reduced through the multimodal lifestyle program applied in the study. There was a direct effect on several health variables and the ESC-SCORE after 15 weeks. Over the next 3 years of follow-up, there were more favorable results in the INT group.

The ESC-SCORE reflects the probability of dying in the next 10 years from a cardiovascular event (5). The ESC-SCORE used in this study is calculated based on age, systolic blood pressure, smoking habits and total cholesterol values (5, 6). Although the algorithm does not consider pre-DM or diabetes status, the ESC-SCORE is nonetheless suitable for a rough assessment of the cardiovascular risk in the subgroup studied, because the relationship of the other risk factors with CVDs are almost parallel in individuals with and without DM (5, 20). However, the risk of persons with DM is generally higher. According to the ESC- SCORE's instructions, it should be considered for the interpretation that the calculated risk at every risk factor combination can be at least twice as high in men and up to 4-fold higher in women with manifest DM (5). It must therefore be assumed that the actual CVD risk tends to be underestimated by the ESC-SCORE value for the subgroup studied, but because many more pre-DM patients than patients with manifest DM were included in the analysis, the underestimation should not be too far-reaching.

The overall results suggest a clear positive health effect of the intervention for the subgroup studied, which is very similar to the effect for the entire study cohort group (4). Persons in the intervention group generally benefited from the multimodal lifestyle program, which was reflected in more favorable ESC-SCORE changes compared to those in the usual care control group over the course of the study. Multimodal interventions that also target self-empowerment, such as the program in the PreFord study, promise long-term effectiveness, which in turn may also be cost-effective (21). Kähm et al. (22) estimated the costs for diabetic complications in German patients. End-stage renal disease, amputations, stroke, myocardial infarction and ischemic heart disease were deemed very cost-intensive. Indirect costs related to lost productivity and work ability due to diabetes and its complications are also very high (23). Magliano et al. (24) demonstrated that "productivity-adjusted life years" were reduced by 11.6% and 10.5% among men and women with DM, respectively. Interventions that focus on persons with pre-DM and DM and which are initiated early in working life could thus help reduce work absenteeism and protect the workforce by preventing the development of disease complications.

A closer look at the long-term effects on the ESC-SCORE changes (intention-to-treat analysis) implies that pre-DM patients in particular benefited from the lifestyle intervention. Further measures may be necessary to achieve more beneficial effects in patients with manifest DM. However, it should be noted that the pre-DM patients already had higher values at the beginning of the study, so that possible improvements may be more pronounced in them than in the DM patients. However, the result should not be overestimated, as only 42% of the DM patients of the intention-to-treat analysis fully adhered to the study protocol.

The strategy for raising awareness of CVD risk at the workplace through flyers and offering a quick medical check-up free of charge could—as demonstrated in the present study—motivate workers with pre-DM and DM to participate in multimodal therapy. Despite the noted drop-out rate during the intervention of 37% among those with pre-DM and DM (for all study participants, the rate was 32%), the intention-to-treat analysis nevertheless indicated significant and clinically meaningful improvements post-intervention, underscoring the program's overall efficacy.

The PreFord study has some limitations, which have already been pointed out in the initial publication (4). One limitation, for example, is the fact that there could be concerns against the employer who pushed the study, so that some employees did not participate in the CVD mortality risk screening due to concerns that their health data could be misused. Therefore, the representativeness of the results for the entire company cannot be guaranteed. Another limitation is that very few women were included. Therefore, the question is to what extent the results are gender-specific. This cannot be clarified based on the present data.

An additional point that might be of interest, especially for the secondary analysis, is that no distinction was made between the types

of DM. Among the 13 insulin-dependent patients, some patients with type 1 DM may have been included. However, Juutilainen et al. (25) showed in an 18-year observational study that there was no major difference between middle-aged individuals with T1DM and T2DM in terms of their CVD mortality risk (onset of the disease was > 30 years in both groups). However, other data suggest a greater mortality risk for T2DM patients compared with T1DM patients when the age of onset of the diabetic disease is earlier in both groups (15-30 years) (26).

Furthermore, there was a minor, but statistically significant difference in BMI values between the INT and CON group, which might have affected the development of health values. However, for the primary outcome (ESC-SCORE), the groups were almost perfectly matched.

5 Conclusion

In conclusion, attracting company employees who are at high CVD mortality risk to participate in a multimodal lifestyle program following a free CVD mortality risk screening at their workplace may be a successful strategy for CVD prevention, particularly in patients with pre-DM/DM. The multimodal intervention used in the PreFord study was suitable for improving the health of company employees with pre-DM/DM and for reducing their CVD mortality risk in the long term.

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 Ethics Committee of the University of Cologne. The patients/participants provided their written informed consent to participate in this study.

Author contributions

CB had the idea for this paper. CB and HH performed the statistical analyses. CB wrote and revised the manuscript. All other authors (D-BG, CA, SM, BB-W, JL, GH, KW, CH, MS, H-GP) have contributed substantially to the design, acquisition, analysis and interpretation of study data from the PreFord study and gave their intellectual input to the present manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of interest

CB is a member of the Abbott Diabetes Care Advisory Board and has received research grants and honoraria from Abbott. GH was employed by the Health Service of the Ford Motor Company GmbH.

The remaining 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/fendo.2023.1106334/full#supplementary-material

References

1. GBD 2017 Causes of Death Collaborators. Global, regional, and national age-sex-specific mortality for 282 causes of death in 195 countries and territories, 1980-2017: A systematic analysis for the global burden of disease study 2017. *Lancet* (2018) 392:1736–88. doi: 10.1016/S0140-6736(18)32203-7

2. Jagannathan R, Patel SA, Ali MK, Narayan KMV. Global updates on cardiovascular disease mortality trends and attribution of traditional risk factors. *Curr Diabetes Rep* (2019) 19:44. doi: 10.1007/s11892-019-1161-2

3. Barbaresko J, Rienks J, Nöthlings U. Lifestyle indices and cardiovascular disease risk: A meta-analysis. *Am J Prev Med* (2018) 55:555–64. doi: 10.1016/j.amepre.2018.04.046

4. Gysan DB, Millentrup S, Albus C, Bjarnason-Wehrens B, Latsch J, Gohlke H, et al. Substantial improvement of primary cardiovascular prevention by a systematic score-based multimodal approach: A randomized trial: The PreFord-study. *Eur J Prev Cardiol* (2017) 24:1544–54. doi: 10.1177/2047487317718081

5. Conroy RM, Pyörälä K, Fitzgerald AP, Sans S, Menotti A, de BG, et al. Estimation of ten-year risk of fatal cardiovascular disease in Europe: The SCORE project. *Eur Heart J* (2003) 24:987–1003. doi: 10.1016/S0195-668X(03)00114-3

6. Elsner LK, von JB, Grün D, JS W, Weferling M, Diouf K, et al. Prognostic performance of the ESC SCORE and its German recalibrated versions in primary and secondary prevention. *Eur J Prev Cardiol* (2020) 27:2166–9. doi: 10.1177/2047487319868034

7. Kannel WB. Diabetes and cardiovascular disease. *JAMA* (1979) 241:2035. doi: 10.1001/jama.1979.03290450033020

8. Wei M, Gaskill SP, Haffner SM, Stern MP. Effects of diabetes and level of glycemia on all-cause and cardiovascular mortality. *San Antonio Heart Study Diabetes Care* (1998) 21:1167–72. doi: 10.2337/diacare.21.7.1167

9. Yusuf S, Hawken S, Ôunpuu S, Dans T, Avezum A, Lanas F, et al. Effect of potentially modifiable risk factors associated with myocardial infarction in 52 countries

(the INTERHEART study): Case-control study. *Lancet* (2004) 364:937–52. doi: 10.1016/S0140-6736(04)17018-9

10. Fuller H, Alberti H. Barriers to lifestyle changes in people with diabetes. *Br J Gen Pract* (2017) 67:61. doi: 10.3399/bjgp17X689005

11. MacDonald CS, Ried-Larsen M, Soleimani J, Alsawas M, Lieberman DE, Ismail AS, et al. A systematic review of adherence to physical activity interventions in individuals with type 2 diabetes. *Diabetes Metab Res Rev* (2021) 37:e3444. doi: 10.1002/dmrr.3444

12. Echouffo-Tcheugui JB, Selvin E. Prediabetes and what it means: The epidemiological evidence. *Annu Rev Public Health* (2021) 42:59–77. doi: 10.1146/annurev-publhealth-090419-102644

13. Tinajero MG, Malik VS. An update on the epidemiology of type 2 diabetes: A global perspective. *Endocrinol Metab Clin North Am* (2021) 50:337–55. doi: 10.1016/j.ecl.2021.05.013

14. Begg C, Cho M, Eastwood S, Horton R, Moher D, Olkin I, et al. Improving the quality of reporting of randomized controlled trials: The CONSORT statement. *JAMA* (1996) 276:637–9. doi: 10.1001/jama.1996.03540080059030

15. ADA American Diabetes Association. 2. classification and diagnosis of diabetes: Standards of medical care in diabetes-2021. *Diabetes Care* (2021) 44:S15–33. doi: 10.2337/dc21-S002

16. Paul S, Ali A, Katare R. Molecular complexities underlying the vascular complications of diabetes mellitus – a comprehensive review. *J Diabetes Complications* (2020) 34:107613. doi: 10.1016/j.jdiacomp.2020.107613

17. Qazi MU, Malik S. Diabetes and cardiovascular disease: Original insights from the framingham heart study. *Glob Heart* (2013) 8:43–8. doi: 10.1016/j.gheart.2012.12.008

18. Narayan KMV, Imperatore G, Benjamin SM, Engelgau MM. Targeting people with pre-diabetes. *BMJ (Clinical Res ed.)* (2002) 325:403–4. doi: 10.1136/bmj.325.7361.403

19. Tuso P. Prediabetes and lifestyle modification: Time to prevent a preventable disease. *Perm J* (2014) 18:88–93. doi: 10.7812/TPP/14-002

20. Stamler J, Vaccaro O, Neaton JD, Wentworth D. The multiple risk factor intervention trial research group. diabetes, other risk factors, and 12-yr cardiovascular mortality for men screened in the multiple risk factor intervention trial. *Diabetes Care* (1993) 16:434–44. doi: 10.2337/diacare.16.2.434

21. Rodriguez-Mañas L, Laosa O, Vellas B, Paolisso G, Topinkova E, Oliva-Moreno J, et al. Effectiveness of a multimodal intervention in functionally impaired older people with type 2 diabetes mellitus. *J Cachexia Sarcopenia Muscle* (2019) 10:721–33. doi: 10.1002/jcsm.12432

22. Kähm K, Laxy M, Schneider U, Rogowski WH, Lhachimi SK, Holle R. Health care costs associated with incident complications in patients with type 2 diabetes in Germany. *Diabetes Care* (2018) 41:971–8. doi: 10.2337/dc17-1763

23. Riddle MC, Herman WH. The cost of diabetes care-an elephant in the room. *Diabetes Care* (2018) 41:929–32. doi: 10.2337/dci18-0012

24. Magliano DJ, Martin VJ, Owen AJ, Zomer E, Liew D. The productivity burden of diabetes at a population level. *Diabetes Care* (2018) 41:979–84. doi: 10.2337/dc17-2138

25. Juutilainen A, Lehto S, Rönnemaa T, Pyörälä K, Laakso M. Similarity of the impact of type 1 and type 2 diabetes on cardiovascular mortality in middle-aged subjects. *Diabetes Care* (2008) 31:714–9. doi: 10.2337/dc07-2124

26. Constantino MI, Molyneaux L, Limacher-Gisler F, Al-Saeed A, Luo C, Wu T, et al. Long-term complications and mortality in young-onset diabetes: Type 2 diabetes is more hazardous and lethal than type 1 diabetes. *Diabetes Care* (2013) 36:3863–9. doi: 10.2337/dc12-2455

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Influence of state of health and personality factors of resilience and coping in healthy subjects and those with diabetes

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Introduction: Currently, the most common chronic metabolic disease in our society is Diabetes Mellitus. The diagnosis of Diabetes Mellitus supposes an impact for the patient, since it requires a modification in the lifestyle, which demands a great capacity for adaptation and modification of habits. The aim of the study was to determine whether personality factors and health status influence resilience and coping strategies in a sample of healthy and diabetic subjects.

Methodology: The sample included a total of 401 subjects (201 patients with Diabetes and 200 without pathology). The instruments applied for data collection were: Sociodemographic data questionnaire, the Resilience Scale, the Coping Strategies Questionnaire and The "Big Five" factor taxonomy. The data collection period was approximately 2 years (between February 2018 and January 2020).

Results: Certain personality factors, such as Emotional Stability, Integrity, Conscientiousness and Extraversion, were positively related to Resilience. Additionally, Emotional Stability, Integrity, and Extraversion were positively associated with Rational Coping. On the other hand, emotional stability, agreeableness and extraversion were negatively related to emotional coping. In relation to health status, the absence of pathology is related to the use of rational strategies more than to the diagnosis of diabetes. Therefore, the participants in this study present different psychological patterns depending on personality and health status.

Conclusions: The present study shows that the subjects of the sample present different psychological patterns depending on Personality and health status.

KEYWORDS

resilience, coping strategies, personality factors, diabetes mellitus, chronic diseases

1. Introduction

According to the latest studies, the most frequent chronic metabolic disease in our society is diabetes mellitus (1). In 2002, the WHO announced a worldwide prevalence of diabetes of 3%, which corresponds to 170 million people in the world diagnosed with this pathology. It was even estimated that this figure would double by 2025 (2). Today, these forecasts have already been exceeded. The latest figures provided by the International Diabetes Federation (IDF), corresponding to 2019, showed that 9.3% of adults have diabetes, which corresponds to a total of 463 million people. They also indicated that 1.1 million children and adolescents under the age of 20 live with type 1 diabetes. In addition, the IDF estimates that in 2030, 578 million adults will be living with the disease. In 2045, it is estimated that the figure will rise to 700 million (3).

These data are of great importance because the diagnosis, prognosis and treatment of the disease have a great emotional impact on the patient. This is associated with the need to assume a pathology that will accompany the subject throughout their life and the subject's obligation to modify their life habits in order to obtain a better quality of life, thus reducing complications deriving from it (4).

There is even research that details the psychological repercussions that accompany diabetes (5, 6). Thus, it has been stated that said pathology can be associated with depression and anxiety. These two diseases can arise regardless of the type of diabetes, especially in the presence of clinical complications. Therefore, it is essential that health workers use programmes in their clinical practice that address the emotional demands detected. Even the American Diabetes Association has incorporated new medical care recommendations, with the intention of including the assessment of the psychological and social situation of subjects diagnosed with diabetes (7, 8).

Based on the aforementioned theoretical aspects, some research has been oriented toward the identification of those psychological mediators that could contribute to achieving a better quality of life in subjects with chronic disease. Thus, the impact of resilience, personality and coping strategies on the health of subjects with chronic pathologies has been studied.

Therefore, research studying resilience and coping strategies has increased (9–11). Resilience is defined from the health field as the ability of individuals to maintain health and quality of life in a dynamic and challenging environment. Therefore, it is considered relevant variable for in the area of health due to its capacity to buffer stress (12–16). In the study by Pasantes et al. interventions were carried out with diabetic patients aimed at promoting their level of resilience. These incidents had a positive result on hemoglobin A1C levels (17). The type of coping strategies that people use to adapt to their illness can anticipate the impact caused by said illness. Therefore, certain coping styles can mediate and buffer the effects of stress. It is stated that active coping strategies are positively related to health (18).

In addition, personality may also play a fundamental role in the way subjects deal with the disease, directly influencing their wellbeing. Thus, personality modulates the way in which people face and adapt to a chronic disease, favoring the development of resilience and the use of coping strategies (19–21). The psychological aspect of diabetes is considered an important part of the treatment and management of this condition in the modern world. Thus, the assessment of personality traits can play a substantial role in the proper treatment of diabetics. The study by Esmaeilinasab et al. was determined that extraversion in diabetic patients is associated with better disease control (21). In addition, this may be relevant if we take into account that some studies indicate that patients with diabetes have different personality traits than subjects without pathology (22). In the context of this study, personality is approached through the Big Five model in Spanish, which hierarchically orders five personality factors: emotional stability, agreeableness, integrity, conscientiousness and extraversion (23).

Based on the scientific evidence outlined above, the objective of this study was to determine whether health status and personality factors influence resilience and coping strategies in a sample of healthy and diabetic subjects. For this reason, we consider it essential to know which diseases predict a worse adaptation, as well as those personality characteristics that favor the development of resilience, in order to focus on effective and individualized health programme.

The novelty of this study is justified in the use of a clinical sample. This allowed us to assess the influence of the Personality, but also the health status of the subjects to explain the Resilience and Coping. In most studies, only the relationships between these psychological variables have been investigated, through samples with healthy population, such as university students (24, 25).

2. Methodology

2.1. Aim and design of the study

The aim of this study was to determine whether health status and personality factors influence resilience and coping strategies in a sample of healthy and diabetic subjects. The study had a non-experimental cross-sectional design with a correlational objective.

2.2. Participants

These samples were selected at the University Assistance Complex of Salamanca. Four hundred and thirty six subjects participated in the study, of which 35 were excluded for not completing the informed consent or not completing the questionnaires. The total sample consisted of 401 subjects (Figure 1).

The study participants were 200 healthy subjects and 201 patients with diabetes ($N = 401$). The majority of it is made up of men ($N = 285$) and are mainly aged between 44 and 50 years ($N = 118$). Most of the subjects are married/in a couple ($N = 247$) and only 85 subjects have higher education (Table 1).

Pearson's $\chi 2$ test was used, using Cramer's V to determine the effect size. Thus, it was detected that the subsamples of our study, no significant differences were detected in the sociodemographic variables ($p > 0.05$) (Table 1).

In both subsamples, to participate in the project, the following inclusion criteria must be met: The subjects can be of legal age and participate voluntarily in the study. In the case of subjects with diabetes, an additional inclusion criterion was having a confirmed diagnosis of said disease, regardless of its stage. An additional inclusion criterion in healthy patients was that they were not diagnosed with any disease. The exclusion criteria were: suffering from a disease that would prevent the patient from completing the study, not agree to participate in the study and have been diagnosed with an affective pathology that could bias the results.

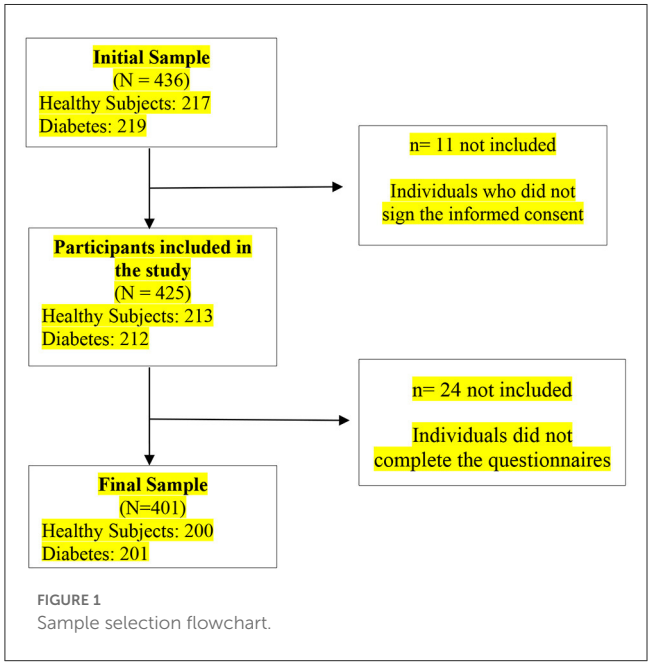

FIGURE 1
Sample selection flowchart.

2.3. Data collection

The samples were selected following a quota sample with equivalent age ranges, sex and educational level, with the aim of achieving homogeneous sub-samples. The sample was collected at the University Assistance Complex of Salamanca. The selection of the subsample made up of diabetic patients was carried out in the Diabetes Unit of the Clinical Hospital of Salamanca and the Internal Medicine hospitalization wards of the same hospital.

After obtaining the sample of subjects with diabetes, the sample of healthy subjects was selected. The selection of this subsample was carried out in different Salamanca health centers (namely, "Periurbana Sur" and "Capuchinos" Health Centers). These patients voluntarily participated in the study after attending their scheduled appointment in the nursing consultation.

The data collection period was ∼2 years (between February 2018 and January 2020), through the instruments detailed below.

2.3.1. Sociodemographic data questionnaire

Sociodemographic data were collected through an instrument made up of a series of questions of a socio-demographic nature and information on the presence of diabetes.

- Health status (subjects without pathology, subjects with diabetes).

TABLE 1 Sample description.

	Diabetes		Healthy		Total		Ji	TE	*p*
	N	%	*N*	%	*N*	%			
N° participants	201	50.1%	200	49.9%	401	100%			
Sex									
Woman	58	28.9%	58	29.0%	116	28.9%	0.001	0.002	0.975
Man	143	71.1%	142	71.0%	285	71.6%			
Age									
43 years or younger	46	22.9%	59	29.5%	105	26.2%	2.588	0.080	0.460
44 to 50 years	63	31.3%	55	27.5%	118	29.4%			
From 51 to 55 years old	44	21,9%	38	19.0%	82	20.4%			
56 years or older	48	23,9%	48	24.0%	96	23.9%			
Marital status									
Married/couple	123	61.2%	124	62.0%	247	61.6%	4.574	0.107	0.102
Single/widowed/other	64	31.8%	51	25.5%	115	28.7%			
Separated/divorced	14	07.0%	25	12.5%	39	9.7%			
Level of studies									
Secondary or lower	160	79.6%	156	78.0%	316	78.8%	0.154	0.020	0.695
Superior	41	20.4%	44	22.0%	85	21.2%			

- Socio-demographic variables studied (age, sex, marital status and educational level).

2.3.2. Personality: "Big Five" factor taxonomy

To assess personality factors, the "Taxonomic Proposal of the Big Five in Spanish" was produced by Iraegui and Quevedo-Aguado (26). This research consisted of a psycholinguistic approach to the study of personality following the "Big Five hypothesis." Principal component factor analysis was applied to the 150 mini-markers finally identified in this research as personality descriptors. The Kaiser rule was employed to select the number of factors to retain and varimax normalization was used as the rotation method.

The five factor solution required ten iterations for convergence and explained 19.36% of the total variance with a Cronbach's α of 0.88. For our study we have used a reduced scale of 50 personality descriptors, ten for each factor (five positive and five negative). These descriptors were chosen based on their correlations with the corresponding factor. In this investigation, the use of the reduced version was chosen due to its brevity and its adequate psychometric properties. The global reliability of the instrument is $\alpha = 0.884$, finding each of the five factors in indices that oscillate between $\alpha = 0.079$ and $\alpha = 0.89$ (26).

In this scale, the subjects have to evaluate these 50 descriptors depending on whether they are suitable or not for defining their personality traits. The response range was from 0, not suitable, to 4, very suitable. A total score was obtained for each of the factors.

2.3.3. Coping strategies questionnarie

The Coping Strategies Questionnaire scale was designed by the authors Sandín and Chorot in 2002. This questionnaire contains a scale made up of 42 items, which score from 0 (never) to 4 (almost always). Through this scale, two general dimensions of coping can be measured: emotional coping and rational coping. Also, based on this general classification, it allows assessing seven more specific coping dimensions. Thus, emotional coping includes negative self-focused coping and overt emotional expression. Rational coping included problem-solving coping, positive reappraisal, and seeking social support. Each coping factor/dimension includes seven items, with the total variance explained by the seven factors being 55.3% (27).

2.3.4. Resilience scale

Wagnild and Young created The Resilience Scale in 1993, adapted by Novella in 2002 into Spanish (28). This scale has 25 items. Each item ranges from 1 (strongly disagree) to 7 (strongly agree).

The scale assesses five resilience factors: personal satisfaction, equanimity, feeling good alone, self-confidence, and perseverance. Global internal consistency was measured using Cronbach's α coefficient ($\alpha = 0.88$).

2.4. Ethical considerations

This study received a positive report from the Clinical Research Ethics Committee of the University Hospital of Salamanca PIO02/01/2018.

2.5. Data analysis

Statistical analysis was performed using International Business Machines' (IBM) Statistical Package for the Social Sciences (SPSS) version 25 (IBM Corp., Armonk, NY, USA). To determine whether personality factors and health status influence resilience and coping strategies in a sample of healthy subjects and those with diabetes, linear regression analysis was performed.

This technique requires the fulfillment of five assumptions: independence, non-collinearity, linearity, homoscedasticity and normality. After verifying these assumptions, the linear regression analysis was applied. For this, the variables were grouped into two blocks. One of them contained the dummy variables, and the other the rest of the variables. For the first block the method was introduced, while for the second the stepwise regression was obtained. To study the fit of the model, the coefficient of determination (R^2) and the adjusted coefficient of determination (R^2_A) were used. In all statistical test, testing was significant when $p > 0.05$.

3. Results

3.1. Linear regression

For each dependent variable, a linear regression analysis was performed. These de-pendent variables (DVs) were resilience, rational coping and emotional coping. In contrast, the independent variables (IVs) were the factors of personality and the state of health of the subjects. In categorical VI with two or more levels, it was necessary to create dummy variables.

3.1.1. Resilience

In relation to resiliene, the regression revealed that the best model is the one in which four variables were included ($R^2 = 0.848$, $R^2_A = 0.719$, F (df1, df2) = 20.047 (1, 393), $p \leq 0.001$). These predictor variables included in the final model explained 71.6% of the variance in DVs.

Table 2 shows that emotional stability, integrity, conscientiousness and extraversion were positively related to resilience. Personality factor 1 (emotional stability) was the one with the greatest weight (B = 0.883, β = 0.457, t = 11.998, $p <$ 0.001). In relation to health status, the dummy of healthy subjects was not significant (B = 0.990, β = 0.875, t = 1.137, $p = 0.256$).

Finally, the existence of atypical and predominant cases was assessed. Table 3 shows the value of the most extreme cases in different measures. Two atypical cases are detected in RV but neither of them is predominant.

TABLE 2 Final model coefficients: resilience.

	B	Std error	β	t	p
Constant	32.589	3.938		8.279	<0.001
F1 emotional stability factor	0.883	0.074	0.457	11.998	<0.001
F3 integrity factor	0.663	0.115	0.221	5.756	<0.001
F4 conscientiousness factor	0.449	0.099	0.175	5.531	<0.001
F5 extraversion factor	0.384	0.078	0.166	4.936	<0.001
Healthy subjects	0.990	0.875	0.031	1.137	0.256

B, unstandardized regression coefficients; Std error, standard error; β, standardized regression coefficients; p, p-value.

TABLE 3 Atypical and predominant cases: resilience (linear regression).

	Maximum	Minimum	Cases out of range
Typified residues	4.166	−2.971	2
Waste diversion	4.288	−3.103	2
Leverage	0.075	-	0
Cook's distance	0.110	-	0

TABLE 4 Final model coefficients: rational coping.

	B	Std error	β	t	p
Constant	23.016	2.702		8.521	<0.001
F1 emotional stability factor	0.554	0.073	0.333	7.582	<0.001
F3 integrity factor	0.859	0.101	0.331	8.507	<0.001
F5 extraversion factor	0.491	0.080	0.247	6.161	<0.001
Healthy subjects	4.244	0.894	0.153	4.746	<0.001

B, Unstandardized Regression Coefficients; Std. error, standard error; β, Standardized Regression Coefficients; p, p-value.

3.1.2. Rational coping

The regression revealed that the best model is the one in which four variables were included ($R^2 = 0.776$, $R^2_A = 0.598$, F (df1, df2) = 22.527 (1, 393), $p < 0.001$). These predictor variables included in the final model explained 59.8% of the RV variance.

Table 4 shows that emotional stability, integrity and extraversion are positively related to rational coping. In relation to the state of health, the dummy of healthy subjects was significant, being the variable that had the greatest weight (B = 4.244, β = 0.153, $t = 4.746$, $p \leq 0.001$).

Finally, the existence of atypical and predominant cases was assessed. Table 5 shows the value of the most extreme cases in different measures. Two atypical cases are detected, but neither is predominant.

3.1.3. Emotional coping

The regression revealed that the best model is the one in which three variables were included ($R^2 = 0.457$, $R^2_A = 0.243$, F (df1, df2) = 5.539 (1, 394), $p = 0.019$). These predictor variables included in the final model explained 24.3% of the variance in DVs.

TABLE 5 Atypical and predominant cases: rational coping (linear regression).

	Maximum	Minimum	Cases out of range
Typified residues	4.116	−2.971	2
Waste diversion	4.288	−3.103	4
Leverage	0.075	-	0
Cook's distance	0.110	-	0

TABLE 6 Final model coefficients: emotional coping.

	B	Std error	β	t	p
Constant	25.231	1.309		19.278	<0.001
F1 Emotional Stability factor	−0.257	0.042	−0.378	−6.125	<0.001
F2 Agreeableness factor	−0.181	0.048	−0.204	−3.783	<0.001
F5 Extraversion factor	−0.106	0.046	−0.130	−2.323	0.021
Healthy subjects	−0.375	0.511	−0.033	−0.733	0.464

B, Unstandardized Regression Coefficients; Std. error, standard error; β, Standardized Regression Coefficients; p, p-value.

TABLE 7 Atypical and predominant cases: emotional coping (linear regression).

	Maximum	Minimum	Cases out of range
Typified residues	4.166	−2.973	2
Waste diversion	4.288	−3.103	3
Leverage	0.070	-	0
Cook's distance	0.110	-	0

Table 6 shows that emotional stability, agreeableness and extraversion were negatively related to emotional coping. In relation to health status, healthy subjects have negative coefficients, but this variable was not significant (B = −0.375, β = 0.033, $t = -0.733$, $p = 0.464$).

Finally, the existence of atypical and predominant cases was assessed. Table 7 shows the value of the most extreme cases in different measures. Two atypical cases are detected, but neither of them is predominant.

4. Discussion

Different studies have confirmed that the diagnosis of a chronic pathology and individual differences in personality traits can influence the development and maintenance of resilience and coping strategies (20, 26–29). Then, these results are compared with those of our project.

The results obtained in our research show that emotional stability, integrity, responsibility and extraversion were positively

related to resilience. We can highlight the fact that emotional stability turned out to be a significant predictor in all models. In addition, it was the variable with the highest weight for the resilience variable.

In a similar vein, numerous studies reflect the negative association between resilience and neuroticism (19, 25, 30–35). In order to compare this argument with the results of our re-search, it should be noted that the subjects with a low score in neuroticism are located on the opposite side of the emotional stability factor. Therefore, the results of the cited authors coincide with those of our study: the emotional stability factor was the one that was most related to higher levels of resilience.

Other research also reflects the positive relationship between extraversion, integrity and conscientiousness with levels of resilience (33, 35–39). This evidence also coincides with the results presented in our study, which reflect that the three factors contributed significantly to the prediction of resilience.

Also, the results of our research, in relation to the study of coping strategies, revealed that emotional stability, integrity and extraversion were positively related to rational coping. Emotional stability, agreeableness and extraversion were negatively associated with emotional coping. Other authors have also addressed the relationship between personality factors and coping strategies. The research found on this study topic indicates that the way in which an individual faces problem is influenced by their personality traits (40, 41). Thus, Mirnics et al. (42) found in their research that emotional stability was the trait that most significantly predicted coping strategies. Thus, emotional stability was associated positively with rational strategies and negatively with emotional strategies. In addition, extraversion and conscientiousness were found to be positively related to the use of rational strategies. Other authors, such as Afshar et al. (43), found similar results in their research, pointing out that subjects with a higher level of extraversion, integrity and emotional stability frequently use more rational strategies. This evidence is in line with the results obtained in our study. We also found results similar to those obtained in our research in the work of Leszko et al. (44), which indicated that agreeableness is negatively associated with emotional coping.

Therefore, the results obtained in our research are consistent with published studies on personality factors that predict resilience and coping. In relation to health status, our research shows that the absence of a pathology predicted a greater use of rational coping strategies.

However, there is little research focused on predicting the levels of resilience and coping strategies used based on the health status of the subjects. However, it has been described that the diagnosis of a chronic disease is a stress factor, hindering the development of resilience and threatening the coping capacity of the individual (14). Our results predicted that healthy and diabetic subjects would not present differences in the resilience variable. Thus, subjects with diabetes have learned how to face, overcome and transform themselves in the face of adversity.

It should be noted that there is very little research with which we can compare these results. Thus, few studies compare the level of resilience of subjects with diabetes and healthy subjects. However, we found similar results to those presented in our project in the research carried out by Novaes (45), which was conducted with a sample of subjects with diabetes mellitus and healthy subjects. In this study, it was found that there were no significant differences in the level of resilience between the groups. This finding coincides with that obtained in our study (45).

However, other studies have shown that healthy subjects have a higher level of resilience than patients with chronic pathologies (14). Thus, we consider it necessary to develop more research that evaluates and com-pares resilience in specific chronic diseases. It should be noted that each chronic disease has very different characteristics in terms of its development and therapeutic plan. For this reason, it is necessary to carry out more studies that compare the level of resilience according to the state of health. We consider it essential to investigate and learn about the pathologies associated with lower levels of resilience, since different studies coincide in believing that resilient people are more capable of coping with disease processes, both their own and those of others, and emerge stronger from the situation (13, 46).

Finally, the results of this study show that the state of health was also related to the type of coping strategies. The subsample of healthy subjects presented a greater use of rational cutting strategies. These strategies are associated with positive coping, coping with stress and trauma differently between individuals (47). Furthermore, rational coping, characterized by the mobilization of the patient to deal with the disease, is associated with greater adaptation to the disease and a higher quality of life (46–49). However, we find opposite conclusions in other studies, which state that diabetic subjects more frequently use rational coping strategies (50, 51).

4.1. Limitations

We point out as the main limitation that personality and health status only explained 24.3% of emotional coping. Therefore, variables that help improve our predictions are missing. However, there are investigations that state that resilience and gender can also predict the type of emotional coping used (52). Future research should take into account these variables not included in the models, which may be relevant for predicting the variables that are not well-explained.

5. Conclusion

Subjects present different psychological patterns depending on personality and health status. This conclusion may be useful in clinical practice for developing strategies, individually, focused on individuals with certain personality characteristics that predict a greater risk of maladjustment to their disease. Also, in an individualized way, strategies could be developed focused on individuals with certain personality characteristics that predict a greater risk of maladjustment to their disease. Therefore, the conclusions of this study show the importance of developing individualized health programs to address diabetes. However, it would be important to expand the study with other chronic diseases.

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 El Comité Ético de Investigación Con Medicamentos del Área de Salud de Salamanca. Código CEIC: PlO02/01/2018. The patients/participants provided their written informed consent to participate in this study.

Author contributions

Conceptualization: CR-P, MHB-C, and PMR-M; methodology: CR-P and JM-T; software: JM-T and JLS-G; validation: MHB-C, RJ-V, and CR-P; formal analysis: CR-P, RJ-V, and JLS-G; investigation: RJ-V and PMR-M; resources: PMR-M; data curation: CR-P, JLS-G, and JM-T; writing-original draft preparation: CR-P and PMR-M; writing-review and editing: CR-P and MPQ-A; visualization: CR-P and JM-T; supervision: JM-T and PMR-M; project administration: CR-P; funding acquisition: RJ-V, JM-T, and JLS-G. All authors have read and agreed to the published version of the manuscript.

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.

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References

1. Nadal J, Cases M, Puente D. Epidemiología y control clínico de la diabetes mellitus tipo 2 y sus comorbilidades en España (estudio e Control). *Medicina Clínica*. (2021) 147:1–7. doi: 10.1016/S0025-7753(17)30618-8

2. Ramirez S, Villa-Ruano N, García D. Epidemiología genética sobre las teorías causales y la patogénesis de la diabetes mellitus tipo 2. *Gaceta Médica de México*. (2017) 153:864–874. doi: 10.24875/GMM.17003064

3. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, et al. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: results from the International Diabetes Federation Diabetes Atlas, 9 th edition. *Diab Res Clin Pract*. (2019) 157:107843. doi: 10.1016/j.diabres.2019.107843

4. Beléndez M, Lorente I, Maderuelo M. Emotional distress and quality of life in people with diabetes and their families. *Gaceta Sanitaria*. (2015) 29:300–3. doi: 10.1016/j.gaceta.2015.02.005

5. Samaniego RA, García I, Sánchez FM, Del Río ML, Esparza ÓA. Coping and its relationship with quality of life in patients with type 2 diabetes mellitu. *European Journal of Health Research*. (2018) 4:19–29. doi: 10.30552/ejhr.v4i1.87

6. Wild S, Roglic G, Green A, Global prevalence of Diabetes estimates for the year 2000 and projections for 2030. *Diabetes Care*. (2004) 27:1047–53. doi: 10.2337/diacare.27.5.1047

7. Azzallini S, Vera BP, Vidal V, Benvenuto A, Ludmila F. Depression and anxiety in patients with diabetes type 2 and its relationship with the coping strategies used for the adherence to treatments. *Anuario de Investigaciones*. (2015) 22:287–91. Available online at: https://www.redalyc.org/articulo.oa?id=369147944029

8. Torales J, Jara G, Ruiz C, Villalba J. *Aspectos Psicopatológicos en el Paciente con Diabetes*. En Pie Diabético: Manual de Manejo, Flores J, Cappello J, Torales H, López H. (Eds.). Don Bosco. 2016 (pp. 16–25).

9. Ferrer L, Kirchner T. How do adolescents with Adjustment Disorder cope with stressful situations? Relationship with suicidal risk. *Revista de psiquiatria y salud mental*. (2020) 13:63–72. doi: 10.1016/j.rpsm.2018.11.002

10. Orozco-Gómez Á, Castiblanco-Orozco L. Factores Psicosociales e Intervención Psicológica en Enfermedades Crónicas No transmisibles. *Revista Colombiana de Psicol*. (2015) 24:203–17. doi: 10.15446/rcp.v24n1.42949

11. Quiceno JM, Vinaccia S. Resilience: a perspective from chronic disease in the adult population. *Pensamiento Psicol*. (2011) 9:69–82.

12. Garrido-Hernansaiz H, Murphy P.J, Alonso-Tapia J. Predictors of resilience and posttraumatic growth among people living with HIV: a longitudinal study. *AIDS and Behavior*. (2017) 21:3260–70. doi: 10.1007/s10461-017-1870-y

13. González I, Eduarda DS, Paiva L, Rossi LA, Dantas S, Alcalá D. Anxiety, depression, resilience and self-esteem in individuals with cardiovascular diseases. *Revista Latino-Americana de Enfermagem*. (2016) 24:2–10. doi: 10.1590/1518-8345.1405.2836

14. Gheshlagh RG, Ebadi A, Dalvandi A, Rezaei M, Tabrizi KN. A systematic study of resilience in patients with chronic physical diseases. *Nurs Midwifery Stud*. (2017) 6:36401. doi: 10.5812/nmsjournal.36401

15. Plascencia JC, Castellanos C. Resilience assessment in mexicans diagnosed with hiv: a comparative study. *Salud y Sociedad*. (2019) 10:52–64. doi: 10.22199/S07187475.2019.0001.00004

16. Fernández-Álvarez N, Fontanil Y, Alcedo Á. Resilience and associated factors in women survivors of Intimate Partner Violence: a systematic review. *Anal. Psicol*. (2022) 38:1631. doi: 10.6018/analesps.461631

17. Pesantes MA, Lazo-Porras M., Abu Dabrh AM, Ávila-Ramírez JR, Caycho M, Villamonte GY, et al. Resilience in vulnerable populations with type 2 diabetes mellitus and hypertension: a systematic review and meta-analysis. *The Canadian journal of cardiology*. (2015) 31:3. doi: 10.1016/j.cjca.2015.06.003

18. Pedraza GL, Vega CZ. Stress, coping, emotions and therapeutic adherence in diabetic patients. *Eureka*. (2018) 15:173–85. Available online at: https://www.psicoeureka.com.py/publicacion/15-2/articulo/8

19. Gong Y, Shi J, Ding H, Zhang M, Kang C, Wang K, Yu Y, Wei J, Wang S. Personality traits and depressive symptoms: the moderating and mediating effects of resilience in Chinese adolescents. *Journal of Affective Disorders*. (2020) 265:611–7. doi: 10.1016/j.jad.2019.11.102

20. Morell-Mengual V, Ruiz-Palomino E, Giménez-García, Castro-Calvo CJ, Díaz-Rodríguez I. The influence of personality in the perception of health care of Spanish youth. *Int J Develop Edu Psycho*. (2016) 2:173–80. doi: 10.17060/ijodaep.2016.n1.v2.199

21. Oshio A, Taku K, Hirano M, Saeed G. Resilience and big five personality traits: a meta-analysis. *Persona Individ Diff*. (2018) 127:54–60. doi: 10.1016/j.paid.2018.01.048

22. Esmaeilinasab M, Ebrahimi M, Mokarrar MH, Rahmati L, Mahjouri MY, Arzaghi SM. Type II diabetes and personality; a study to explore other psychosomatic aspects of diabetes. *J Diab Metabol Disord*. (2016) 15:3. doi: 10.1186/s40200-016-0281-3

23. Van Dooren FE, Denollet J, Verhey FR, Stehouwer CD, Sep SJ, Henry RM, et al. Psychological and personality factors in type 2 diabetes mellitus, presenting the rationale and exploratory results from The Maastricht Study, a population-based cohort study. *BMC Psychiatry*. (2016) 16:22. doi: 10.1186/s12888-016-0722-z

24. Richards, M. Optimism and Resilience in Adolescents. Refereed Scientific *Journal of the MenteClara Foundation*. (2022) 7:259. doi: 10.32351/rca.v7.259

25. Ramírez-Fernández E, Ortega-Martínez AR, Calero-García MJ. Optimism as a mediator between resilience and affective states in older adults. *Estudios de Psicología*. (2018) 39:1–19. doi: 10.1080/02109395.2018.1486360

26. Iraegui A, Quevedo-Aguado MP. Psycholinguistic approach to the study of personality in Spanish: A taxonomic proposal. *Iberpsicología: Revista Electrónica de la Federación española de Asociaciones de Psicología*. (2002) 7. Available online at: http://www.fedap.es/IberPsicologia/iberpsi7-1/iraegui/iraegui.htm

27. Sandín B., Chorot B. The Coping Strategies Questionnaire: Development and preliminary validation. *Revista de Piscopatoogía Piscologia Clínico*. (2002) 8:39–54. doi: 10.5944/rppc.vol.8.num.1.2003.3941

28. Novella A. *Incremento De La Resiliencia Luego De La Aplicación De Un Programa De Psicoterapia Breve En Madres Adolescentes*. Lima: Tesis Doctoral, Universidad Nacional Mayor de San Marcos (2002).

29. Kalisch R, Müller M, Tüscher O. A conceptual framework for the neurobiological study of resilience. *Behav Brain Sci*. (2015) 38:1–79. doi: 10.1017/S0140525X1400082X

30. Linnemann P, Berger K, Teismann, H. Associations between outcome resilience and sociodemographic factors, childhood trauma, personality dimensions and self-rated health in middle-aged adults. *Int J Behav Med*. (2022) 29:796–806. doi: 10.1007/s12529-022-10061-1

31. Martínez-Martí M.L, Ruch, W. Character strengths predict resilience over and above positive affect, self-efficacy, optimism, social support, self-esteem, and life satisfaction. *J Posit Psychol*. (2016) 12:1–10. doi: 10.1080/17439760.2016.1163403

32. Vizoso C, Arias-Gundín O. Relationship between resilience, optimism, and engagement in future educators. *Int J Edu Res Innovat*. (2019) 11:33–46. Available online at: https://dialnet.unirioja.es/servlet/articulo?codigo=6669316

33. Genise G, Genise N, Gómez M, Humeniuk A, Muiños FJ. Relationship Between Psychological Resilience and Personality Factors in Adolescent Population. *Revista Latinoamericana de Ciencia Psicológica*. (2018) 10:1–17. doi: 10.5872/psiencia/10.3.21

34. Holden CL. Characteristics of Veterinary Students: Perfectionism, Personality Factors, and Resilience. *Journal of Veterinary Medical Education ADVANCE*. (2020) 3:e0918111r. doi: 10.3138/jvme.0918-111r

35. Shi M, Liu L, Wang ZY, Wang L. The mediating role of resilience in the relationship between big five personality and anxiety among Chinese medical students: a cross-sectional study. *PLoS ONE*. (2015) 10:1–12. doi: 10.1371/journal.pone.0119916

36. González N, Valdez JL. Resilience and personality in adults. *Revista Electrónica de Psicología*. (2011) 14:295–316. Available online at: https://www.medigraphic.com/pdfs/epsicologia/epi-2011/epi114p.pdf

37. Jalilianhasanpour R, Williams B, Gilman I, Burke MJ, Glass S, Fricchione G, et al. Resilience linked to personality dimensions, alexithymia and affective symptoms in motor functional neurological disorders. *J Psycho Res*. (2018) 107:55–61. doi: 10.1016/j.jpsychores.2018.02.005

38. Rudow D, Lacoviello B, Charney D. Resilience and personality traits among living liver and kidney donors. *Progress Transplant*. (2014) 24:82–90. doi: 10.7182/pit2014448

39. Soriano J, Monsalve V. Profiles of personality and resilience in chronic pain: utility of the CD-RISC-10 to discriminate between resilient and vulnerable types. *Revista de La Sociedad Española Del Dolor*. (2019) 26:72–80. doi: 10.20986/resed.2018.3670/2018

40. Hengartner MP, Linden D. Van Der, Bohleber L, Von Wyl A. Big five personality traits and the general factor of personality as moderators of stress and coping reactions following an emergency alarm on a Swiss university campus. *Stress and Health*. (2016) 33:34–44. doi: 10.1002/smi.2671

41. Pereyra R. Coping and stress within the framework of the five personality factors model. *Review study PSocial*. (2017) 3:39–45. Available online at: https://publicaciones.sociales.uba.ar/index.php/psicologiasocial/article/view/2635

42. Mirnics Z, Bagdy G, Surány Z, Gonda X. The relationship between the big five personality dimensions and acute psychopathology: Mediating and moderating effects of coping strategies. *Psychiatr Danub*. (2013) 25:379–88.

43. Afshar H, Roohafza H, Keshteli AH, Mazaheri M. The association of personality traits and coping styles according to stress level. *Journal of Research in Medical Sciences*. (2015) 20:353–8.

44. Leszko M, Iwansky R, Jerzebinska A. The relationship between personality traits and coping styles among first-time and recurrent prisoners in Poland. *Front Psychol*. (2020) 10:1–8. doi: 10.3389/fpsyg.2019.02969

45. Novaes LE. Stress, resiliência e apoio social em indivíduos com hipertensão e diabetes mellitus. *Revista de Psicología*. (2019) 28:1–13. doi: 10.5354/0719-0581.2019.53954

46. Amar J, Martínez M, Utria L. New approach to health considering the resilience. *Salud Uninorte*. (2013) 29:124–33. Available online at: https://www.redalyc.org/articulo.oa?id=81728689014

47. Theleritis C, Psarros C, Mantonakis L, Roukas D, Papaioannou A, Paparrigopoulos T, et al. Coping and its relation to PTSD in Greek firefighters. *J Nerv Mental Dis*. (2020) 208:252–9. doi: 10.1097/NMD.0000000000001103

48. Furukori-Yasui N, Murakami H, Otaka H, Nakayama H, Murabayashi M, Muzushiri S, et al. Coping behaviors and depressive status in individuals with type 2 diabetes mellitus. *Annals General Psychiatry*. (2019) 18:1–8. doi: 10.1186/s12991-019-0235-5

49. Sanjuán P, Ávila M. Coping and motivation as predictors of subjective and psychological well-being. *Revista de Psicopatología y Psicología Clínica*. (2016) 21:1–10. doi: 10.5944/rppc.vol.21.num.1.2016.15401

50. Féki I, Turki M, Zitoun I, Sellami R, Baati I, Masmoudi J. Dépression et stratégies de coping chez les sujets âgés atteints de diabète de type 2. *L'Encephale*. (2019) 45:320–6. doi: 10.1016/j.encep.2019.01.005

51. Jourdan YY. Coping and quality of life in type 1 and 2 diabetic subjects from Argentina. *Revista ALAD*. (2016) 6:29–40. Available online at: https://www.revistaalad.com/files/alad-v6n1_029-040.pdf

52. Muñoz-Alonzo HM, González-Aguilar D, Ponce ME, Samayoa V, Paniagua WO. Coping and resilience in the context of oncological health care in Guatemala. *Ciencias Sociales y Humanidades*. (2018) 5:9–18. Available online at: https://revistas.usac.edu.gt/index.php/csh/article/view/685

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Feeding-induced hepatokines and crosstalk with multi-organ: A novel therapeutic target for Type 2 diabetes

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Hyperglycemia, which can be caused by either an insulin deficit and/or insulin resistance, is the main symptom of Type 2 diabetes, a significant endocrine metabolic illness. Conventional medications, including insulin and oral antidiabetic medicines, can alleviate the signs of diabetes but cannot restore insulin release in a physiologically normal amount. The liver detects and reacts to shifts in the nutritional condition that occur under a wide variety of metabolic situations, making it an essential organ for maintaining energy homeostasis. It also performs a crucial function in glucolipid metabolism through the secretion of hepatokines. Emerging research shows that feeding induces hepatokines release, which regulates glucose and lipid metabolism. Notably, these feeding-induced hepatokines act on multiple organs to regulate glucolipotoxicity and thus influence the development of T2DM. In this review, we focus on describing how feeding-induced cross-talk between hepatokines, including Adropin, Manf, Leap2 and Pcsk9, and metabolic organs (e.g.brain, heart, pancreas, and adipose tissue) affects metabolic disorders, thus revealing a novel approach for both controlling and managing of Type 2 diabetes as a promising medication.

KEYWORDS

type 2 diabetes, insulin resistance, glucolipid metabolism, feeding-induced hepatokines, multi-organ

1 Introduction

Diabetes is a major illness worldwide that is reaching an epidemic stage (1). Approximately 500 million individuals across the globe are living with diabetes, and it is anticipated that this figure will rise by 25% by the year 2030, and by 51% (roughly 700 million people) by the year 2045 (2). As a result, ensuring that the disease is avoided and

effectively treated is an ultimate importance to public health. Most cases of type 2 diabetes mellitus (T2DM) are caused by modifiable risk factors such as diet (3). Epidemiological surveys have found that the incidence of eating disorders in individuals with type 2 diabetes ranges from 1.2% to 14%, mainly manifesting as over-eating (4). Prior research has declared that nutritional behavior is mainly related to neural regulation in the brain (5). However, recent studies have shown that the liver maintains systemic metabolic homeostasis by transcriptionally controlling the expression of open organ factors in response to external signals such as feeding behavior (6).

In T2DM, when insulin secretion is insufficient, it first causes impaired glucose metabolism. Glycogen synthesis is reduced, and catabolism is increased, tissues' ability to take up and use glucose is reduced, which triggers fasting and postprandial hyperglycemia (7). The malfunction of glucose oxidation and the enhancement of lipolysis metabolism result in an elevation in free fatty acids in the blood, which enter the liver and promote synthesis and release of triglycerides and very-low-density lipoproteins (VLDL), resulting in disorders of lipid metabolism (8). Therefore, it is essential to regulate the metabolism of glucose and lipid in diabetic patients and to understand the treatment mechanism of diabetes, thus improving T2DM patients' life quality.

The liver is a vital organ in the modulation of energy homeostasis because it detects and reacts to shifts in the nutritional condition that occur in response to a wide range of metabolic circumstances (9). The majority of the attribution for the liver's function in the modulation of systemic glucolipid metabolism goes to the release of hepatokines that maintain metabolic homeostasis through autocrine, paracrine and endocrine pathways that regulate the connections between the liver and other organs (10). Of note, feeding can induce the release of hepatokines, which can act on other organs to influence the development of diabetes. Mechanistically, based on the available literature, these feeding-induced hepatokines act through one or more of the following metabolic organs (1) improving pancreatic β-cell cholesterol accumulation, reducing endoplasmic reticulum stress, (2) reducing white adipose tissue inflammation and inhibiting lipid accumulation, (3) inhibiting the brain feeding center and regulating energy homeostasis (Supplemental Table 1).

In this review, We focus on the feeding-induced hepatokines, including Adropin, Manf, Leap2 and Pcsk9, Which participate in the occurrence and development of diabetes. We also highlight the potential mechanisms by which these hepatokines mediate crosstalk between the liver and other organs (brain, heart, adipose tissue, and pancreas) and the possibility of using them as new treatments for T2DM (Figure 1).

2 Adropin

Adropin was originally thought to be a liver-derived peptide implicated in both the homeostasis of energy and the metabolism of glucolipids. The energy homeostasis-related (ENHO) gene encodes a 43-amino acid polypeptide (residues 34-76) that is used to produce this factor (11). There is a possibility that adropin functions as a secretory product of the hepatic biological clock, coordinating metabolic and circadian rhythms and responding to a large number of nutrients and energy balances in the diet. According to preliminary studies, sufficient energy is required for adropin expression in the liver (12). DIO decreased adropin levels in the serum, which were elevated in the nutritional condition (12, 13). As per the current work, the highest expression of Adropin in mice was associated with transcriptional stimulation by RORα/γ, while the minimum expression was associated with Re-verb transcription. Small molecules influencing Rev-erb blocking ability and transcriptional stimulation by RORα/γ quickly modified the expression of ENHO in human HepG2 cells (14). Impressively, it was observed that a high-fat diet that enhances hepatic Adropin expression also elevates liver ROR expression (15). Participation in these nuclear receptors also appears to offer a feasible mechanism for nutritional sensing in the modulation of

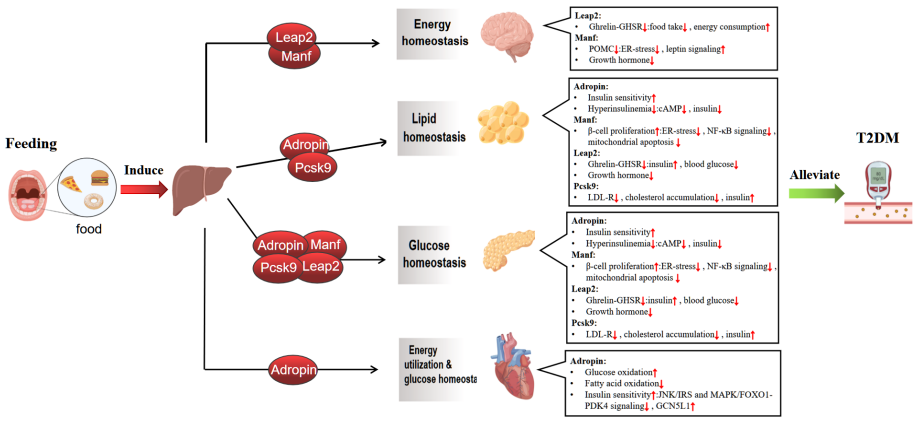

FIGURE 1
Mechanisms of feeding-induced hepatokines amelioration of Type 2 diabetes in target tissues. The liver plays a central role in regulating systemic metabolic homeostasis by sensing nutrient availability and altering metabolite and energy production for use by various organ systems. The act of feeding induces the liver to release hepatokines, which regulate glucolipid metabolism and maintain energy balance by affecting multiple metabolic organs, including the brain, adipose tissue, pancreas and heart, thereby improving type 2 diabete.↑ increase, ↓ decrease. Graphics from http://Biorender.com.

Adropin expression. Although the link between plasma Adropin levels, nutrition, and mealtime is lesser understood in people, plasma Adropin levels in nonhuman primates (constant monkeys) have also peaked at feeding times in available studies (15). In addition, levels of serum Adropin were decreased in patients with T2DM, and elevated circulating Adropin values are related to a decreased hazard of diabetes-related complications (16–18). Here, we summarize the crosstalk of feeding-induced hepatokines on multiple extra-liver organs (i.e., adipose tissue, pancreas, brain, and heart). We hypothesize that it is participated in the mechanisms of reduced obesity, insulin resistance, and glucolipid metabolism, thereby alleviating T2DM glycolipotoxicity.

2.1 Impact of Adropin on energy metabolism

The heart of diabetic patients relies more on the oxidation of fatty acids for producing energy and exhibits impaired glucose uptake and insulin signaling. These changes in cardiac metabolic activity of energy play a part in heart illness intensity (19, 20). Adropin induces significant alterations in cardiac energy metabolic activity *via* boosting glucose metabolism and inhibiting fatty acid oxidation while inhibiting the JNK/IRS-1 S307 phosphorylation axis improves insulin sensitivity, hence boosting metabolic statuses and cardiac efficiency (21). Adropin is hypothesized to have an insulin-sensitizing action to reduce the downregulation of pyruvate dehydrogenase (PDH) negative regulator PDK4 expression levels through MAPKs and FOXO1 signaling mechanisms (21, 22) or modulates the expression of the mitochondrial acetyltransferase GCN5L1, which alters the acetylation condition and the energy activities of the metabolizing enzymes to promote glucose oxidation (23). It also inhibits fatty acid uptake in muscle membranes and mitochondria at the transcriptional level by reducing the protein levels of fatty acid transporter CD36 and carnitine palmitoyltransferase 1 (CPTI) (24, 25). These findings show that adropin plays a significant part in modulating the preference for cardiac energy substrates. In clinical practice, low levels of the serum protein Adropin are connected with many cardiovascular diseases like endothelial malfunction (26–28), heart failure (29, 30), acute myocardial infarction (31), coronary atherosclerosis (32–34), and type X heart syndrome (35). Low levels of Adropin also serve as a separate threat variable and indicator for most illnesses. As a result, there is a good chance of a connection between the levels of Adropin and diabetes-related cardiovascular illness and energy metabolism. This connection needs to be fully investigated.

2.2 Effect of Adropin on lipid metabolism

Previous studies demonstrated that adropin could be participated in the control of adipose tissue function. Adrpin overexpression delayed weight gain in mice fed high-fat meals compared with wild-type animals (11). *In vitro*, through ERK1/2 and AKT-dependent signaling, adropin stimulates the growth of 3T3-L1 cells and mice preadipocytes. Additionally, adropin reduces the lipid deposition and expression of lipogenic genes in these cells. (Pparγ, Fabp4, C/ebpα), bringing about a final reduction in their differentiating process to mature adipocytes (36). Similarly, adropin stimulated brown adipose tissue (BAT) preadipocyte proliferation in Wistar rats through an AKT-dependent pathway, but inhibited preadipocyte maturation by downregulating lipogenic genes (C/ebpα, C/ebpβ, Pgc1α, Pparγ, and Prdm16). Additionally, this study found that adropin decreased lipid accumulation in BAT and increased glycerol and free fatty acid release. It also promoted hormone-sensitive lipase (HSL) activity (37). Of note, the hormone network is complex, and in addition to acting directly on the liver, hormones can interact with other hormones to regulate metabolic homeostasis. Recent studies have shown that adropin slightly promotes lipolysis in rat adipocytes and 3T3-L1 cells but does not affect glucose uptake. In addition, adropin may exert an ameliorative insulin resistance and anti-inflammatory effect by upregulating the expression of adiponectin and inhibiting the expression of resistin and visfatin (38, 39). Overall, Adropin inhibits adipogenesis as well as intracellular lipid accumulation, suggesting that it may improve diabetes by regulating lipid metabolism in adipose tissue as well as modulating the release of other adipokines. Although to further understand the function of Adropin in regulating adipose metabolic mechanism and the development of adipose tissues *in vivo*, additional research is required.

2.3 Effect of Adropin on glycolipid homeostasis

Adropin depletion is linked to higher intensity of glucose homeostasis imbalance as well as abnormalities of lipid metabolic activities when observed *in vivo*. A functional investigation of Adropin knockout (AdrKO) was carried out in C57BL/6J mice by Chen et al. The findings demonstrated that WT mice had normal blood glucose levels significantly lower than those of AdrKO mice when given a conventional diet for one year ($P < 0.0001$). It is interesting to note that after 30 weeks, almost all AdrKO mice developed T2DM when subjected to a high-fat initiation and impaired glycosphingolipid biosynthesis. In addition, a significant number of adipocytes infiltrated the pancreas, a hallmark of a fatty pancreas (FP) (40). Furthermore, the serum levels of Adropin were shown to be significantly reduced in individuals with FP and T2DM compared with healthy individuals, and the levels of relative modulatory T cells (Treg) were also found to be significantly lower and positively connected with Adropin levels ($r=0.7220$, $P=0.0001$) (40). Furthermore, the serum levels of Adropin were shown to be significantly reduced in individuals with FP and T2DM compared with healthy individuals, and the levels of relative modulatory T cells (Treg) were also found to be significantly lower and positively connected with Adropin levels ($r=0.7220$, $P=0.0001$) (40). Treg functions as a negative modulator of the inflammatory condition of adipocytes and were discovered to minimize IR, thereby controlling insulin sensitivity (41). In a model animal with IR caused by a high-fat diet, Adropin can reduce insulin mRNA expression and secretion by affecting the

synthesis of cyclic adenosine monophosphate (cAMP) in pancreatic cells without affecting β-cell viability or proliferation (42). Overall, these findings point to the possibility that Adropin enhances insulin sensitivity and reduces IR by altering T_{reg} number or function and modulating insulin secretion.

3 Manf

Although midbrain astrocyte-derived neurotrophic factor (Manf) was basically classified as a neurotrophic indicator, the protein does not structurally or functionally resemble a true neurotrophic factor. Neurotrophic factors act by the interaction with similar receptors found in the plasma membrane, although no cell surface receptors for Manf were identified (43). Recently Wu et al. demonstrated that RNA sequencing investigation of the livers of mice that were fasted and then fed revealed that Manf is a feeding-induced hepatokine. Manf, which is generated from hepatocytes, raises the body's rate of energy consumption, which combats diet-induced obesity. It would indicate that Manf is directly responsible for the browning of white adipose tissue (WAT) in the groin[13]. Under typical circumstances, the protein Manf can be found in the lumen of the endoplasmic reticulum (44). Endoplasmic reticulum stress (ERS) can increase its expression in different cells and tissues (44–46). The aggregation of unfolded or misfolded proteins in the endoplasmic reticulum causes ER-stress (47). It does this by activating a cellular defensive reaction known as the unfolded protein response (UPR). This response is a signaling cascade that restore endoplasmic reticulum stability (48). It is interesting to note that Apostolou et al. confirmed that Manf is a UPR gene that is able to reduce the amount of apoptosis caused by endoplasmic reticulum stress (46). Moreover, the serum levels of Manf were shown to be lower in patients who had T2DM and had a correlation with the metabolism of lipids and glucose (49). Here, we hypothesize that Manf may mitigate the progression of T2DM by modulating lipid metabolism, inflammation, apoptosis, and proliferation in the liver, adipose, and pancreatic tissues.

3.1 Effect of Manf on glucose metabolism

Increasing evidence suggests that if endoplasmic reticulum stress is not resolved, the UPR transitions from an adaptive (A-UPR) response to a prolonged unresolved UPR, which ultimately results in enhanced inflammatory signaling and autophagy, and apoptosis (50, 51). This is the main cause of β-cell malfunction and death in T2D (52). In T2DM, β-cells are subjected to local environmental parameters, including glycolipotoxicity and inflammatory cytokines, which results in impaired insulin synthesis and increased free fatty acid production, as well as unresolved cell endoplasmic reticulum stress and β-cell death (53, 54). The stimulation of the UPR, which is connected to the buildup of lipid metabolites, is also connected, in a pathological manner, with IR in specific tissues (54). Of interest, most of the literature suggests that the IRE1/XBP1 and ATF6 pathways are involved in the key function of Manf in attenuating the negative modulation of UPR by endoplasmic reticulum stress (45, 55–57). The latest research demonstrated that hepatocyte-derived MANF plays a crucial role in increasing insulin sensitivity and that the systemic injection of MANF protein greatly enhanced insulin sensitivity in mice exhibiting obesity (58). Besides insulin sensitivity, Manf promotes insulin secretion by maintaining the number of pancreaticβ-cell. Ablation of MANF in mouse embryos, both overall and in the pancreas, leads to early onset and severe diabetes mellitus. This is because in Manf -/- mice, phosphorylation of eIF2α inhibits the translation of cyclin D1 and the cell cycle is subsequently arrested in the G1 and G2/M phases, ultimately leading to reduced β-cell proliferation and increased apoptosis (59, 60). MANF overexpression promotes the growth of primary β-cells in humans and mice having diabetes, as well as protection of people and mouse β-cells from the death induced by endoplasmic reticulum stress in β-cells to some extent (61–63). The protective and proliferative effects of MANF on β-cells were correlated with the suppression of NF-κB signaling pathway and amelioration of endoplasmic reticulum stress as well as blocking BH3-only proteins BIM-dependent triggering of mitochondrial apoptotic pathway (64). Chen et al. found that MANF can interact with the DNA binding domain of p65 through its C-terminal SAP-like structural domain and is a key target gene for inhibiting NF-κB signaling pathway (65). Later, Yagi et al. reported that Neuroplastin (NPTN) is a plasma membrane receptor for MANF. The binding of MANF to NPTN attenuates the inflammatory reaction and cell death by inhibiting the NF-κB signaling pathway (66). Another study showed that MANF attenuates endoplasmic reticulum stress by suppressing the IRE1-caspase 12-caspase 3 cell death pathway and has a protective effect against pancreatic alveolar cell injury (67). These results are potential mechanisms for the protective and proliferative influences of MANF on β-cells, which perform important implications for the modulation of insulin production and improvement of glucose metabolism. Notably, Montaser et al. identified the MANF pure-hybrid loss-of-function mutation as a novel gene causing diabetes and neurodevelopmental disorders in children (68). In conclusion, these data further support that MANF performs a crucial part that helps pancreatic β-cell s to survive and proliferate and hence could provide a possible therapy for T2DM patients.

3.2 Effect of Manf on lipid metabolism

Manf differs from any known nerve growth factor in that its N-terminal structural domain is saposin-like lipid conjugation domain (69). SAPLIPs (saposin-like proteins) are a family of lipid-interacting proteins that vary in size and activity and have a wide variety of cellular functions (70). Bai et al. proposed that Manf binds to lipid sulfolipids, which are called 3-O-sulfogalactosyl ceramides, a lipid that exists in the outer leaflet of serum and cell membranes (71).Thus, Manf can bind lipids. Notably, Sousa-Victor et al. identified Manf as a stress response protein that is released and has immunomodulatory effects, as well as being part of an evolutionarily conserved system and a controller of the hepatic

metabolic homeostasis in particular (72). Manf heterozygous mice showed an inflammatory phenotype in multiple tissues, and hepatocellular steatosis and fibrosis, besides developing hepatic bone disease at a faster rate than control mice (72). Overexpression of Manf was able to rescue HepG2 cells from the steatosis that was caused by free fatty acids (FFAs). This was accomplished by inhibiting the synthesis and uptake of fatty acids, as well as suppressing the synthesis of cholesterol. Thus, Manf inhibited lipid deposition in HepG2 human hepatocytes (73). Furthermore, an increase in the levels of the autophagy markers LC3-II and Atg5 was responsible for the attenuation of hepatic steatosis in mice that had Manf overexpressed, which is liver-specific. In addition to this, Manf was responsible for a rise in the phosphorylation of Stat3 as well as its nuclear localization (74). Therefore, Manf influences the metabolism of hepatic lipids by controlling autophagy. The overexpression of genes linked to lipid metabolism, in particular G0/G1 Switch gene 2 (G0S2), appears to be associated with the negative effect of diminished Manf expression in the liver. G0S2 is an important modulator of lipid metabolism and act as a suppressor of lipolysis. It was demonstrated that knocking down Manf causes higher levels of G0S2, which in turn causes hepatic steatosis as well as a pro-inflammatory state in macrophages (72, 75). Knockdown of Manf resulted in an increase in the production of TNF-α, IL-1α, and IL-6 (76). In addition, the increased expression of Manf, which is liver-specific prevented obesity in mice caused by a high-fat diet and accelerated browning of white adipocytes through activating the P38 MAPK pathway. Elevating the expression of key lipolytic proteins (phosphorylated hormone-sensitive lipase (HSL) and adipose triglyceride lipase (ATGL)) is how Manf impedes the expression of M1-type macrophage polarization indicators in mouse eWAT. This assists in decreasing adipose inflammation and improving insulin sensitivity and lipid deposition in high-fat-fed mice (58). In conclusion, these data further support that MANF can improve lipid metabolism in T2DM by a down-modulating inflammatory reaction and lipid deposition in the liver and adipose tissues.

3.3 Effect of Manf on energy metabolism

MANF influences food intake and energy balance by regulating hypothalamic insulin signaling, suggesting that MANF-mediated neuronal activity plays an important role in maintaining energy homeostasis. Furthermore, MANF is enriched in different nuclei of the mouse hypothalamus and critically regulates energy intake, but energy expenditure seems to be unaffected (77). It has been shown that high levels of MANF expression in the rat hypothalamus persist into adulthood (78), raising the possibility that MANF plays an important role in the mature hypothalamus. It is known that ER stress in the hypothalamus leads to leptin resistance and hyperphagia (79), whereas MANF overexpression in Hypothalamic pro-opimelanocortin (POMC) attenuates ER stress and leads to increased thermogenesis in the BAT by improving leptin signaling in the hypothalamus and regulating sympathetic innervation and activity in it (80). These results suggest that MANF overexpression in the hypothalamic nucleus leads to severe hyperphagia and obesity. However, MANF can properly regulate energy homeostasis through POMC neurons. Furthermore, MANF appears to have multifaceted and cell type-specific functions, as recombinant human MANF was recently found to promote corneal epithelial wound healing and nerve regeneration in diabetic patients by attenuating hyperglycemia-induced endoplasmic reticulum stress through the Akt signaling pathway (81). MANF may be a useful therapeutic modality in the treatment of diabetic keratopathy (DK). A recent study showed that strong expression of MANF was also observed in the mouse pituitary, thyroid, and adrenal glands, all tissues involved in the neuroendocrine axis, and important for the regulation of feeding, stress, growth, and development. Interestingly, compared to wild-type mice, MANF-deficient mice have smaller anterior pituitary lobes and reduced numbers of cells producing growth hormone (GH) (82). GH has also been described as a diabetogenic agent with the ability to reduce insulin sensitivity (83). In the brain, GH activates the expression of AgRP neurons, increasing food intake while decreasing energy expenditure (84, 85). These results suggest that MANF plays an essential role in highly hormone-secreting cells within the hypothalamic-pituitary-thyroid/adrenal/gonadal axis and that proper regulation of MANF expression in the brain and other endocrine organs is vital to meet metabolic demands.

4 Leap2

Liver expresses antimicrobial peptide 2 (Leap2), a bicyclic cationic polypeptide (86). As a feeding-induced hepatokine, it is highly expressed in the liver, and its release is inhibited by stopping feeding and returns to baseline levels after subsequent feeding (87). Ghrelin is a hormone that stimulates hunger and is released by the stomach. Ghrelin's function is modulated by binding to the growth hormone secretagogue receptor (GHSR) (88). The ghrelin-GHSR system is implicated in a wide range of biological activities, including the enhancement of growth hormone (GH) production, enhanced hunger and food consumption, control of glucose homeostasis, and cardiovascular function (89–92). According to the most recent findings, Leap2 and Ghrelin are paired with other factors in a competing way with GHSR (93). Thus, Leap2 is a competitive antagonist of GHSR, rather than a non-competitive antagonist as previously reported (94). Leap2 prevents the principal activities of Ghrelin *in vivo*, such as food consumption, GH release, and the control of survivable blood glucose levels throughout periods of calorie reduction or fasting. On the other hand, inhibiting Leap2 has the effect of amplifying the actions of ghrelin (94, 95). As a result, it appears that Leap2 may perform a significant part in metabolic illnesses by acting as a modulator of the ghrelin-GHSR system. In addition, patients who have T2DM had lower serum levels of ghrelin and higher serum levels of Leap2. It's possible that the interaction between Ghrelin and Leap2 performs a significant part in the progression of T2DM. There is some speculation that the ghrelin-Leap2 axis could be a viable therapeutic target for T2D (96). In conclusion, we came to the view that Leap2 has the possibility to be an applicable treatment for the control of T2DM. This is because the ghrelin-GHSR system,

which modulates energy metabolism in the brain, also modulates glucose metabolism in pancreatic tissue.

4.1 Effect of Leap2 on energy metabolism

Furthermore, the N-terminal region alone delivers binding and activation to the Leap2 receptor. Leap2 and its N-terminal part were discovered to act as an inverse agonist of GHSR and also a competing antagonist of Ghrelin-induced phosphatidylinositol production and calcium mobility. Both inverse agonists and antagonists act on the agonist but interact with the receptor in different ways. The inverse agonist binds to the same receptor as the agonist but brings about the opposite response to the agonist, while the antagonist binds to the receptor and disrupts the interaction and function of the agonist and counter-agonist at the receptor (97). LEAP2 is both an inverse agonist of GHSR, which downregulates the constitutive activity of GHSR, and a competitive antagonist, which impairs gastrin-induced activation of GHSR. Thus, Leap2 exerts its inhibitory effect on the ghrelin-GHSR system through its N-terminal region (95). On the one hand, Ghrelin signals are transmitted *via* the vagus nerve to the hypothalamus, which is the modulatory center of nutritional behavior (90). Islam et al. declared that intracerebroventricular (i.c.v.) injection of Leap2 into mice was shown to suppress central Ghrelin function, such as hypothalamus nucleus Fos expression, promoted feeding, elevated blood glucose, and lowered body temperature. However, intraperitoneal (i.p.) leap2 administration showed no reduction in neuropeptide Y (NPY)-induced food consumption or des-acyl ghrelin-induced inhibition in body temperature, demonstrating that Leap2's suppressing activity is specific to the GHSR (98). In contrast, GHSR was strongly expressed in the arcuate nucleus of the hypothalamus (ARC), the dorsal medial nucleus of the hypothalamus (DMH), the ventral medial nucleus of the hypothalamus (VMH), and the lateral hypothalamic nucleus (LH) (99). GHSR governs essential physiological activities such as hunger, neuroendocrine axis, autonomic nervous system activities, and sophisticated mental processes like reward-related attitudes (100). Therefore, the primary function of GHSR is the modulation of neuronal activities (101). The voltage-gated calcium channel 2.2 (Cav2.2) is a prominent GHSR target in neurons. Heterologous expression systems and membrane clamp recordings suggest that the N-terminal region of Leap2 binds GHSR, thereby impairing the ghrelin-dependent (GQ protein signaling) and ghrelin-independent modes of GHSR action (Gi/o protein activation) on the suppression of Cav2.2 currents (102). In addition, the N-terminal region of Leap2 also affects the inhibitory modulation of Cav2.2 currents by the heterodimer of GHSR-the dopamine 2 receptor (D2R)-and its coupling to G proteins (103). Cornejo et al. declared that intracerebroventricular (i.c.v.) injection of C57BL/6J mice with an N-terminal Leap2 fragment diminished overeating in mice on a high-fat diet (104). Taken together, The N-terminus of Leap2 inhibits the ghrelin-GHSR pathway in the central nervous system. Contrarily, the elimination of the LEAP2 gene raised weight gain, food consumption, lean body mass, and liver adipose tissues in HFD-fed female rats. This is a result of less energy consumption, decreased physical exercise, and increased food consumption (105). Furthermore, the Ghrelin-AMPK-SREBP1 pathway may modulate the expression level of Leap2 in the liver. Through the hepatic-gastric-brain axis (98), Leap2 may impact eating and energy balance. Latest research demonstrated that Leap2 also mediates the outcomes of food consumption and energy metabolism through the endogenous cannabinoid system (eCBome)-gut microbiome (mBIome) axis (106). In summary, LEAP2 can improve diabetes by inhibiting brain intake-related energy metabolism.

4.2 Effect of Leap2 on glucose metabolism

One of the main features of Ghrelin and LEAP-2 is that they have opposite effects on GH secretion (107). A recent study using two animal models of GH deficiency found a significant inhibitory effect of LEAP2 on Ghrelin-induced food intake but no change in glucose levels. This suggests that the opposite effect between LEAP-2 and Ghrelin is not dependent on GH levels. The effect of LEAP2 on glucose levels was only observed in obese animals, which may be due to the fact that obese animals exhibit a state of hyperglycemia and insulin resistance, and therefore have a higher setting to trigger a counter-regulatory response to prevent hypoglycemia after LEAP-2 administration (108).On the one hand, GHSR increased expression in peptide cells of people and mice pancreatic islets (109). On the other hand, many studies have shown that both endogenous and exogenous Ghrelin can inhibit insulin production in mice, rats, and humans (110–113). Bayle et al. confronted isolated islets of Langerhans from rat pancreas to glucose with or without LEAP2 and ghrelin, and showed by measuring insulin production that Leap2 exerts modulation of insulin by blocking the insulin-inhibitory effect of Ghrelin (114). Similarly, M'Kadmi et al. demonstrated that N-terminal Leap2 $_{21\text{-}12}$ blocked the inhibitory effect of Ghrelin on insulin secretion in rat pancreatic islet cells cultured *in vitro* (95). Furthermore, overexpression of Leap2 in mice reduced blood glucose levels (94). Thus, circulating levels of Leap2 may influence glycemic control by blocking Ghrelin function to modulate insulin secretion. Recent studies have found that Leap2 $_{38\text{-}47}$ exhibits insulin-promoting properties in cultured human pancreatic islet cells. The insulin-promoting properties are consistent with the LEAP2 fragment (Leap2 $_{38\text{-}47}$) acting as a reverse Ghrelin receptor agonist (115). These impacts of Ghrelin are mediated at least in part by direct GHSR interactions that are differentially localized in α-cells, β-cells, δ-cells secreting growth inhibitory hormone (SST), and γ-cells of the pancreas expressing pancreatic polypeptide (pp) (109). *In vitro*, pharmacological and genetic inhibition of islet-derived ghrelin significantly enhances glucose-induced insulin response. In mice with modest obesity brought on by a high-fat diet, ghrelin deprivation increased insulin release and prevented decreased glucose tolerance (116). Leap2 thereby inhibits the insulin-suppressing and glucose-increasing actions of the ghrelin-GHSR pathway and might offer a therapeutic application for the control of T2DM.

5 Pcsk9

Pcsk9 (proprotein convertase subtilisin/kexin), has just come to light as one of the most important hepatokines, which induces the breakdown of hepatic low-density lipoprotein receptor (LDL-R) *via* the ribosomal/lysosomal pathway, thereby increasing circulating low-density lipoprotein cholesterol (LDL-C) levels (117, 118). It is expressed to a much lesser extent in the pancreas, adipose cells, gut, and kidney than it is in the liver, which has a high expression of it (119). According to the findings of a clinical investigation, a high-fructose diet elevated plasma Pcsk9 concentrations by 28% in healthy subjects and by 34% in the progeny of patients with T2DM who were more likely to be insulin resistant (120). As a result, decreasing plasma levels of PCSK9 presents itself as an intriguing possible treatment target for dyslipidemia in diabetic patients. Interestingly, feeding induced an increase in hepatic PCSK9 levels. Due to the increased insulin levels during feeding, it leads to the activation of Pcsk9 transcription by SREBP-1c (121). Given this characteristic, Pcsk9 is also referred to as feeding-induced hepatokine. Thus, we suggest that feeding-induced hepatokine Pcsk9 plays a role in T2DM. In this chapter, we describe in detail the impact of feeding-induced hepatokine Pcsk9 on the development of T2DM by acting on adipose tissue and the pancreas to improve glucolipid metabolism.

5.1 Effect of Pcsk9 on lipid metabolism

WAT malfunction and IR are thought to contribute significantly in the development of T2D, which delays clearance of triglyceride-rich lipoproteins (TRL), promotes elevated plasma TG and NEFA and flow to other peripheral tissues, leading to apoB overproduction, systemic lipotoxicity, inflammation, IR, and hyperinsulinemia (122–124). Upregulation of LDL-R uptake is associated with abnormal adipocyte metabolic function and risk of diabetes mellitus. Subjects who had normal cholesterol levels but had lower plasma levels of PCSK9 and higher levels of LDL-R and CD36 on the surface of their WAT also exhibited higher levels of WAT NLRP3 inflammasome activity and T2D-related hazard indicators (125). It's possible that this is because LDL causes a reduction in adipocyte activity. Consistently, native LDL reduced WAT function and inhibited preadipocyte differentiation and function in mice (126). Others have reported that oxidized low-density lipoprotein (OxLDL) inhibits adipocyte differentiation (127). however, this effect is dependent on CD36 (a native scavenger receptor for VLDL and LDL, oxLDL, and NEFA) (128, 129). Of note, NLRP3/IL-1β inflammatory pathway stimulation promotes WAT malfunction and T2D and is controlled by LDL-R and CD36. It was revealed that oxLDL in CD36-internalized macrophages (130) and oxLDL and native LDL in endothelial cells (131) enhance the NLRP3 inflammasome, resulting in the release of the pro-inflammatory cytokine white IL-1β, which impedes insulin signaling in multiple cells, including adipocytes, β-cells, and hepatocytes (132, 133). Demers et al. declared that Pcsk9 stimulates the breakdown of CD36 in the acidic compartment behind the endoplasmic reticulum through a proteasome-sensitive mechanism that contributes to reducing the uptake of fatty acids and the deposition of triglyceride in tissues (134). Furthermore, Pcsk9 also limits visceral adipogenesis by degrading adipose tissue VLDL-R and LDL-R (135, 136). In some populations, elevated Plasma apolipoprotein B (apo B) plasma counts can predict the incidence of T2D 3-10 years prior to the onset of T2D independently of traditional risk factors (137, 138). ApoB plasma level indicates the quantity of small, dense LDL particles (137). Higher plasma apoB/pcsk9 levels are related with risk indicators for WAT malfunction and T2D, including postprandial hypertriglyceridemia, IR, hyperinsulinemia, and increased plasma interleukin 1 receptor antagonist (IL-1ra), according to multiple research (139, 140). Recently, we discovered that this ratio was indeed linked to high expression of LDL-R and CD36 on the WAT surface as well as WAT malfunction, inflammation, and IR (141). Therefore, Pcsk9 may be beneficial in improving WAT malfunction, inflammation, and IR, thereby reducing the hazard of T2DM.

5.2 Impact of Pcsk9 on glucose metabolism

In pancreatic β-cells, cholesterol is an integral part of the cell membrane and is involved in controlling the physical properties of the cell membrane, thus influencing the distribution and the functionality of membrane proteins, as well as the formation and fusion of vesicles (142). Whereas cholesterol accumulation is mainly *via* LDL-R, Cholesterol overload in β-cell s is a mechanism that limits or destroys glucose-stimulated insulin secretion (GSIS) (143), and it is related to any genetic or pharmacological treatment that raises LDL-R expression. It is believed that factors that influence the homeostasis of cellular cholesterol metabolism can have an effect on the beta-cell activity as well as the development of diabetes (144). In contrast, Roehrich et al. showed that human lipoproteins play an important role in modulating the survivability of β-cells. Purified human VLDL and LDL induced increased apoptosis and decreased insulin transcript levels. Conversely, HDL effectively counteracts cell death through mechanisms such as stimulation of Akt/protein kinase B (Akt/protein kinase B) and inhibition of caspase-3 cleavage (145). These findings point to the possibility that changes in lipoproteins are linked to the beta-cell malfunction that is seen throughout the advancement of T2DM. Similarly, Cnop et al. found a series of lipid abnormalities in individuals having T2DM that associated with the accumulation of cholesterol and fatty acids in pancreatic β-cell s and may lead to islet degeneration (146). It has been shown that Pcsk9 reduces LDL-R, which in turn reduces cholesterol accumulation in pancreatic β-cells and promotes increased glucose-dependent insulin secretion (GSIS) (147). Furthermore, Mbikay et al. reported that male mice with Pcsk9 deletion above four months old had more LDL-R while having lesser insulin in their pancreas and showed hypoinsulinemia, hyperglycemia, and glucose intolerance (148). Thus, Pcsk9 prevents islet degeneration and promotes insulin secretion by

limiting pancreatic β-cell cholesterol accumulation. Notably, circulating/liver-derived (the primary target of monoclonal antibodies) does not affect the β-cell function and insulin secretion (147). Ramin et al. found that neither exogenous PCSK9, Alirocumab, nor PCSK9 silencing significantly affected glucose-stimulated insulin secretion (GSIS) from pancreatic β-cells (149). Similarly, Peyot et al. demonstrated that Pcsk9 deficiency did not have any toxic effects on β-cell activity and glucose homeostasis in either the whole-body KO or βKO mouse models (150). According to these findings, anti-PCSK9 medications, which primarily target circulating Pcsk9, have only a minimal influence on the malfunction of β-cell s and the prevalence of diabetes. In conclusion, Pcsk9 improves T2DM by limiting pancreatic β-cell cholesterol overload, maintaining glucose metabolic homeostasis, and preventing β-cell malfunction.

6 Clinical consideration of feeding-induced hepatokines in T2DM

Plasma levels of adropin are lower in individuals diagnosed with T2DM, particularly those who are obese or overweight (151). Recently, Adropin has also been increasingly studied in relation to diabetes-related complications. In addition to its function as a marker of malfunctional endothelium cells, adropin also has a preventative effect on the occurrence and advancement of cardiovascular diseases (26, 27, 152). Elevated plasma adropin concentrations in male individuals with T2DM patients and those showing obesity who were treated with liraglutide can partially explain the cardiovascular benefits and protective effects (153). Adropin, as a potential anti-inflammatory factor (154), emerges as a potential biomarker for predicting the development of MAFLD in patients with T2DM (155) and diabetic kidney disease (DKD) (156). In addition, the therapeutic potential of adropin for T2DM is demonstrated by its effects on the activity of various elements of the endocrine system, including the adrenal cortex. It has been shown that adropin inhibits steroidogenesis and secretion of adrenocorticotropic hormones (e.g., cortisol and aldosterone) in HAC15 cells by binding to the GPR19 receptor and activating the TGF-β-dependent pathway (157). Cortisol, a glucocorticoid that raises blood sugar and reduces insulin secretion (158), has been shown in a clinical study to increase insulin resistance in patients with T2D when the HPA axis loses its ability to lower cortisol levels during hyperglycemia (159). Similarly, aldosterone has been associated with glucose intolerance and insulin resistance, and drugs related to mineralocorticoid receptor (MRs) antagonists have been used to improve insulin resistance and endothelial dysfunction (160). Furthermore, the CNS effects of adropin inhibition of drinking water were also associated with the expression of GPR19 receptors (161). Perhaps adropin plays an important act in the control of water content in the body by modulating the CNS, with a pivotal role in preventing the intake of additional fluids. This could have a positive effect in relieving renal load in patients with diabetic nephropathy. The current study did not identify the adropin receptor(s), and clarifying the receptor(s) for adropin has potential implications for the treatment of type 2 diabetes. Some researchers have suggested that the biological effects of adropin are obtained by direct binding to the G protein-coupled receptor GPR19 (22, 161, 162), but the study by Foster et al. failed to confirm that adropin interacts with GPR19 (163). However, it has been shown that Adropin is a meningeal-binding protein that interacts with NB-3. Adropin may be important for NB-3 recruitment, concentration, and Notch1 receptor binding, which in turn contributes to cerebellar development (164). In addition, adropin may exert its physiological effects by acting directly on neurons in the PVN (165). PVN is a key autonomic control center that plays an important role in the regulation of fluid balance (166), energy homeostasis (167), and cardiovascular regulation (168). These findings further solidify that Adropin has an endocrine function as a hepatokine and provide the framework needed to link its peripheral effects to its role in the central nervous system.

Recent studies suggest that MANF performs a crucial part in food consumption as well as energy homeostasis (169) and its involvement in the modulation of metabolic disorders. Multiple clinical research declared that there is an association between T2DM and circulating MANF levels. Serum MANF levels were elevated in newly diagnosed prediabetic and T2DM patients than in non-diabetic controls (170), while circulating MANF levels were significantly diminished in T2DM patients (49). This is because early in patients with T2DM, IR in the liver, skeletal muscle, and adipose tissue causes endoplasmic reticulum stress in these tissues, inducing MANF expression. The compensatory increase in MANF may act as a protective mechanism against endoplasmic reticulum stress-induced cellular damage against disease progression, but as the disease progresses, accompanied by prolonged glucotoxicity and/or lipotoxicity, MANF expression decreases, thereby exacerbating the illness. Additionally, the negative correlation of MANF with FBG and HbA1c was confirmed by the results (49). Therefore, MANF may be a new therapeutic candidate to protect the organism from lipotoxicity and glucotoxicity-induced endoplasmic reticulum stress.

Through its interaction with the growth hormone secretagogue receptor (GHSR), the hormone ghrelin is able to control not only the amount of food that is consumed but also the level of glucose in the blood (171). A recently discovered endogenous ligand of the GHSR is known as Leap2 (172). The reduction in serum ghrelin levels and the elevation of Leap2 levels in individuals with type 2 diabetes may represent a physiological compensation as a response to a positive energy balance to maintain a normal energy balance. Lowering the Ghrelin/Leap2 ratio in individuals with T2DM may lower the overactivation of the GHSR in obese patients, which in turn may restore normal energy homeostasis (96). This viewpoint is reinforced by a paper that showed improvements in obesity and diabetes when levels of acyl ghrelin were reduced, levels of Leap2 were increased, or GHSR activity was blocked (173). A recent clinical study has shown that exogenous LEAP2 reduces postprandial glucose and suppresses appetite in healthy men, and these effects may be mediated through the GHSR (174). Thus the discovery of the endogenous inverse agonist LEAP2 may reveal potential therapeutic targets for gastric hunger-related diseases, including type 2 diabetes and obesity, as it interacts with gastric hunger and is expressed at elevated levels after RYGB surgery (115). Notably, many gastrin/gastrinase targeted drugs, such as anti-gastrin L-RNA inducers (anti-gastrin vaccines), GHSR antagonists, GHSR inverse agonists, GOAT inhibitors, cyclized

deacetyl-gastrin analogs, none of which have entered late-stage clinical trials for the treatment of obesity or type 2 diabetes due to uncertainty about their safety and/or efficacy in humans (175–178). Therefore, further studies are needed to confirm the safety of LEAP-2 based compounds.

PCSK9, an endogenous suppressor of the LDLR pathway, works by guiding the breakdown of LDLR to the lysosome (179). PCSK9 is thought to be a factor suggesting increased cardiovascular risk in T2DM (180). Therefore, commercially available PCSK9 inhibitors can lower circulating LDL-C, thereby treating dyslipidemia in T2DM (181, 182). Recent studies have shown that Patients who have prodromal diabetes but not yet T2DM lack plasma PCSK9 levels that can forecast their likelihood of developing T2DM (183). Circulating levels of PCSK9 are linked to dyslipidemia in T2DM, which we suggest is due to its unique physiological functions related to lipid metabolism, but its beneficial effects on metabolic organs cannot be ignored, and anti-PCSK9 treatments that focus on circulating PCSK9 have a minimal effect on the organs that are being targeted. To date, up to nine PCSK9 inhibition strategies have been or are being developed to either block it's binding to LDLR or prevent its maturation, secretion, or synthesis (184). These therapies include the use of anti-PCSK9 monoclonal antibodies (e.g., two FDA-approved drugs: alirocumab and evolocumab) (185), antisense oligonucleotides (ASO), small interfering RNAs (siRNAs), vaccines and small molecules (186). In patients with type 2 diabetes and hypercholesterolemia or mixed dyslipidemia treated with statins, PCSK9 inhibitors significantly reduced LDL-C, non-HDL-C and apoB levels. In addition, favorable changes were observed in postprandial levels of celiac disease, VLDL-C, and LDL-C (187, 188). Therefore, inhibition of PCSK9 is a promising new way to improve dyslipidemia in patients with T2DM to prevent cardiovascular disease. It should be noted, however, that completed clinical trials have not shown adverse effects of PCSK9 inhibitors on the risk of diabetes, but the safety of the inhibitors should be validated in long-term randomized trials (189).

7 Conclusion and future perspective

The liver is a vital organ in the body's reaction to alterations in nutritional condition because it performs a crucial part in glucose and lipid metabolism. This review summarizes the crosstalk of some feeding-induced hepatokines Adropin, Manf, Leap2, and Pcsk9 in the liver and extrahepatic tissues such as brain, adipose, heart, and pancreatic tissues, and by targeting these feeding-induced hepatokines is expected to be a possible therapy for T2DM to help in control and treatment.

Many recent studies have demonstrated the high sensitivity of the liver to metabolic changes during fasting and refeeding, and here we discuss the important role played by other hepatokines regulated during the feeding-fasting-refeeding cycle concerning energy and glucolipid metabolism. For instance, refeeding signals during intermittent fasting (IF) induce the liver to produce a release of pregnancy band protein (PZP), and circulating PZP binds to GRP78 on the cell surface *via* the p38 MAPK-ATF2 signaling pathway, increasing UCP1 expression in BAT. PZP acts as a key hepatokine regulating IF, triggering energy homeostasis *via* the liver/BAT axis (190). Another feeding-induced hepatokine Tsukushi (TSK), is also involved in regulating energy metabolism through the liver/BAT axis. TSK ablation enhances thermogenic gene expression in BAT and suppresses obesity-associated inflammation in the liver and adipose tissue. Meanwhile, TSK acts as a metabolic signal from the liver, balancing the activation of hypothalamic melanocortin circuits during feeding (191). Angptl8 is a key regulator of the liver clock in response to food. angptl8 is regulated by nutritional and hormonal factors, and feeding induces an increase in its levels (192). It has been reported that ANGPTL8 not only induces the expression of brown adipocyte markers (193) but also promotes subcutaneous white adipose tissue (SAT) browning under acute and chronic hypothermic conditions (194). Fibroblast growth factor 21 (FGF21) is known to be a hepatokine induced by fasting and is being pursued as a therapeutic target for diabetes and obesity due to its rapid and effective action in improving insulin sensitivity (195). However, several studies have also demonstrated that FGF21 maintains a presence and functional role even during feeding. The expression of the FGF21 gene is paradoxically regulated by fasting and feeding signals. On the one hand, two fasting signals, including PPARa and glucagon-PKA, increase the expression of FGF21 gene. On the other hand, glucose and xylitol, which are feeding signals, also induced FGF21 expression through ChREBP activation (196). Overall, expression of the human FGF21 gene is paradoxically independently regulated by fasting and feeding signals. These regulatory mechanisms suggest that FGF21 increases in response to nutritional crises, including starvation and overfeeding. Therefore, FGF21 levels are likely to be useful markers for determining our nutritional status. Additionally, recent studies have identified a novel fasting-induced hepatokine orosomucoid (ORM) 2 as a key regulator of hepatic *de novo* lipogenesis (DNL) production. ORM2 plays an important role in inhibiting lipogenesis and maintaining hepatic and systemic lipid homeostasis. Therefore, ORM2 and its analogs may provide a potential pharmacological treatment for dyslipidemia (197). All in all, most of the regulatory responses to diet initially occur in the liver, and hepatokines play a key role in maintaining nutritional homeostasis by regulating the metabolism of other organs as signal messengers from the liver.

Given the interaction between these feeding-induced hepatokines and multiple organs, further *in vivo* experiments are needed to investigate their relationship with glucolipid metabolism, energy homeostasis, and inflammation, thus presenting novel approaches for the clinical management of diabetes in the years to come. Important areas of future research include (1) understanding how preclinical evidence of feeding-induced hepatokines translates to human studies, (2) determining the mechanisms by which feeding-induced hepatokines and other secreted factors integrate to modulate metabolism through interorgan interactions, (3) determining the pathology of these hepatokines in the development of diabetes physiology, will help to improve the prevention and even the treatment of this disease.

We hope that this will systematize the knowledge of feeding-induced hepatokines and help establish new lines of research regarding their role in metabolic organs.

Author contributions

X-HX and Z-ZL contributed to the design of this article. Three authors R-BC, Q-YW and Y-YW conducted a literature search and research selection in Pubmed. R-BC and Q-YW wrote the manuscript and made final revisions. The work was done under the supervision of X-HX and Z-ZL. All authors contributed to the article and approved the submitted version.

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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/fendo.2023.1094458/full#supplementary-material

References

1. Cho NH, Shaw JE, Karuranga S, Huang Y, da Rocha Fernandes JD, Ohlrogge AW, et al. IDF diabetes atlas: Global estimates of diabetes prevalence for 2017 and projections for 2045. *Diabetes Res Clin Pract* (2018) 138:271–81. doi: 10.1016/j.diabres.2018.02.023

2. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, et al. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: Results from the international diabetes federation diabetes atlas, 9(th) edition. *Diabetes Res Clin Pract* (2019) 157:107843. doi: 10.1016/j.diabres.2019.107843

3. Neuenschwander M, Ballon A, Weber KS, Norat T, Aune D, Schwingshackl L, et al. Role of diet in type 2 diabetes incidence: Umbrella review of meta-analyses of prospective observational studies. *BMJ* (2019) 366:l2368. doi: 10.1136/bmj.l2368

4. Maggioni AP, Caterson ID, Urso R, Coutinho W, Finer N, Van Gaal L, et al. Relation between weight loss and causes of death in patients with cardiovascular disease: finding from the SCOUT trial. *J Cardiovasc Med (Hagerstown)* (2017) 18 (3):144–51. doi: 10.2459/JCM.0000000000000492

5. Luo SX, Huang J, Li Q, Mohammad H, Lee CY, Krishna K, et al. Regulation of feeding by somatostatin neurons in the tuberal nucleus. *Science* (2018) 361(6397):76–81. doi: 10.1126/science.aar4983

6. Katsumura S, Siddiqui N, Goldsmith MR, Cheah JH, Fujikawa T, Minegishi G, et al. Deadenylase-dependent mRNA decay of GDF15 and FGF21 orchestrates food intake and energy expenditure. *Cell Metab* (2022) 34(4):564–80 e8. doi: 10.1016/j.cmet.2022.03.005

7. Jiang S, Young JL, Wang K, Qian Y, Cai L. Diabeticinduced alterations in hepatic glucose and lipid metabolism: The role of type 1 and type 2 diabetes mellitus (Review). *Mol Med Rep* (2020) 22(2):603–11. doi: 10.3892/mmr.2020.11175

8. Athyros VG, Doumas M, Imprialos KP, Stavropoulos K, Georgianou E, Katsimardou A, et al. Diabetes and lipid metabolism. *Hormones (Athens)* (2018) 17 (1):61–7. doi: 10.1007/s42000-018-0014-8

9. Smati S, Regnier M, Fougeray T, Polizzi A, Fougerat A, Lasserre F, et al. Regulation of hepatokine gene expression in response to fasting and feeding: Influence of PPAR-alpha and insulin-dependent signalling in hepatocytes. *Diabetes Metab* (2020) 46(2):129–36. doi: 10.1016/j.diabet.2019.05.005

10. Priest C, Tontonoz P. Inter-organ cross-talk in metabolic syndrome. *Nat Metab* (2019) 1(12):1177–88. doi: 10.1038/s42255-019-0145-5

11. Kumar KG, Trevaskis JL, Lam DD, Sutton GM, Koza RA, Chouljenko VN, et al. Identification of adropin as a secreted factor linking dietary macronutrient intake with energy homeostasis and lipid metabolism. *Cell Metab* (2008) 8(6):468–81. doi: 10.1016/j.cmet.2008.10.011

12. Stevens JR, Kearney ML, St-Onge MP, Stanhope KL, Havel PJ, Kanaley JA, et al. Inverse association between carbohydrate consumption and plasma adropin concentrations in humans. *Obes (Silver Spring)* (2016) 24(8):1731–40. doi: 10.1002/oby.21557

13. Ganesh Kumar K, Zhang J, Gao S, Rossi J, McGuinness OP, Halem HH, et al. Adropin deficiency is associated with increased adiposity and insulin resistance. *Obes (Silver Spring)* (2012) 20(7):1394–402. doi: 10.1038/oby.2012.31

14. Ghoshal S, Stevens JR, Billon C, Girardet C, Sitaula S, Leon AS, et al. Adropin: An endocrine link between the biological clock and cholesterol homeostasis. *Mol Metab* (2018) 8:51–64. doi: 10.1016/j.molmet.2017.12.002

15. Kohsaka A, Laposky AD, Ramsey KM, Estrada C, Joshu C, Kobayashi Y, et al. High-fat diet disrupts behavioral and molecular circadian rhythms in mice. *Cell Metab* (2007) 6(5):414–21. doi: 10.1016/j.cmet.2007.09.006

16. Es-Haghi A, Al-Abyadh T, Mehrad-Majd H. The clinical value of serum adropin level in early detection of diabetic nephropathy. *Kidney Blood Press Res* (2021) 46 (6):734–40. doi: 10.1159/000519173

17. Wei W, Liu H, Qiu X, Zhang J, Huang J, Chen H, et al. The association between serum adropin and carotid atherosclerosis in patients with type 2 diabetes mellitus: a crosssectional study. *Diabetol Metab Syndr* (2022) 14(1):27. doi: 10.1186/s13098-022-00796-y

18. Li S, Sun J, Hu W, Liu Y, Lin D, Duan H, et al. The association of serum and vitreous adropin concentrations with diabetic retinopathy. *Ann Clin Biochem* (2019) 56 (2):253–8. doi: 10.1177/0004563218820359

19. Buchanan J, Mazumder PK, Hu P, Chakrabarti G, Roberts MW, Yun UJ, et al. Reduced cardiac efficiency and altered substrate metabolism precedes the onset of hyperglycemia and contractile dysfunction in two mouse models of insulin resistance and obesity. *Endocrinology* (2005) 146(12):5341–9. doi: 10.1210/en.2005-0938

20. Lopaschuk GD, Ussher JR, Folmes CD, Jaswal JS, Stanley WC. Myocardial fatty acid metabolism in health and disease. *Physiol Rev* (2010) 90(1):207–58. doi: 10.1152/physrev.00015.2009

21. Altamimi TR, Gao S, Karwi QG, Fukushima A, Rawat S, Wagg CS, et al. Adropin regulates cardiac energy metabolism and improves cardiac function and efficiency. *Metabolism* (2019) 98:37–48. doi: 10.1016/j.metabol.2019.06.005

22. Thapa D, Stoner MW, Zhang M, Xie B, Manning JR, Guimaraes D, et al. Adropin regulates pyruvate dehydrogenase in cardiac cells *via* a novel GPCR-MAPK-PDK4 signaling pathway. *Redox Biol* (2018) 18:25–32. doi: 10.1016/j.redox.2018.06.003

23. Thapa D, Xie B, Zhang M, Stoner MW, Manning JR, Huckestein BR, et al. Adropin treatment restores cardiac glucose oxidation in pre-diabetic obese mice. *J Mol Cell Cardiol* (2019) 129:174–8. doi: 10.1016/j.yjmcc.2019.02.012

24. Gao S, McMillan RP, Jacas J, Zhu Q, Li X, Kumar GK, et al. Regulation of substrate oxidation preferences in muscle by the peptide hormone adropin. *Diabetes* (2014) 63(10):3242–52. doi: 10.2337/db14-0388

25. Gao S, McMillan RP, Zhu Q, Lopaschuk GD, Hulver MW, Butler AA. Therapeutic effects of adropin on glucose tolerance and substrate utilization in diet-induced obese mice with insulin resistance. *Mol Metab* (2015) 4(4):310–24. doi: 10.1016/j.molmet.2015.01.005

26. Topuz M, Celik A, Aslantas T, Demir AK, Aydin S, Aydin S. Plasma adropin levels predict endothelial dysfunction like flow-mediated dilatation in patients with type 2 diabetes mellitus. *J Investig Med* (2013) 61(8):1161–4. doi: 10.2310/JIM.0000000000000003

27. Lovren F, Pan Y, Quan A, Singh KK, Shukla PC, Gupta M, et al. Adropin is a novel regulator of endothelial function. *Circulation* (2010) 122($^{11\ Suppl}$):S185–92. doi: 10.1161/CIRCULATIONAHA.109.931782

28. Kwon OS, Andtbacka RHI, Hyngstrom JR, Richardson RS. Vasodilatory function in human skeletal muscle feed arteries with advancing age: the role of adropin. *J Physiol* (2019) 597(7):1791–804. doi: 10.1113/JP277410

29. Lian W, Gu X, Qin Y, Zheng X. Elevated plasma levels of adropin in heart failure patients. *Intern Med* (2011) 50(15):1523–7. doi: 10.2169/internalmedicine.50.5163

30. Yosaee S, Soltani S, Sekhavati E, Jazayeri S. Adropin- a novel biomarker of heart disease: A systematic review article. *Iran J Public Health* (2016) 45(12):1568–76.

31. Mushala BAS, Scott I. Adropin: a hepatokine modulator of vascular function and cardiac fuel metabolism. *Am J Physiol Heart Circ Physiol* (2021) 320(1):H238–H44. doi: 10.1152/ajpheart.00449.2020

32. Sato K, Yamashita T, Shirai R, Shibata K, Okano T, Yamaguchi M, et al. Adropin contributes to anti-atherosclerosis by suppressing monocyte-endothelial cell adhesion and smooth muscle cell proliferation. *Int J Mol Sci* (2018) 19(5):1293. doi: 10.3390/ijms19051293

33. Wu L, Fang J, Chen L, Zhao Z, Luo Y, Lin C, et al. Low serum adropin is associated with coronary atherosclerosis in type 2 diabetic and non-diabetic patients. *Clin Chem Lab Med* (2014) 52(5):751–8. doi: 10.1515/cclm-2013-0844

34. Zhao LP, You T, Chan SP, Chen JC, Xu WT. Adropin is associated with hyperhomocysteine and coronary atherosclerosis. *Exp Ther Med* (2016) 11(3):1065–70. doi: 10.3892/etm.2015.2954

35. Celik A, Balin M, Kobat MA, Erdem K, Baydas A, Bulut M, et al. Deficiency of a new protein associated with cardiac syndrome x; called adropin. *Cardiovasc Ther* (2013) 31(3):174–8. doi: 10.1111/1755-5922.12025

36. Jasaszwili M, Wojciechowicz T, Billert M, Strowski MZ, Nowak KW, Skrzypski M. Effects of adropin on proliferation and differentiation of 3T3-L1 cells and rat primary preadipocytes. *Mol Cell Endocrinol* (2019) 496:110532. doi: 10.1016/j.mce.2019.110532

37. Jasaszwili M, Wojciechowicz T, Strowski MZ, Nowak KW, Skrzypski M. Adropin stimulates proliferation but suppresses differentiation in rat primary brown preadipocytes. *Arch Biochem Biophys* (2020) 692:108536. doi: 10.1016/j.abb.2020.108536

38. Jasaszwili M, Pruszynska-Oszmalek E, Wojciechowicz T, Strowski MZ, Nowak KW, Skrzypski M. Adropin slightly modulates lipolysis, lipogenesis and expression of adipokines but not glucose uptake in rodent adipocytes. *Genes (Basel)* (2021) 12(6):914. doi: 10.3390/genes12060914

39. Zhang S, Chen Q, Lin X, Chen M, Liu Q. A review of adropin as the medium of dialogue between energy regulation and immune regulation. *Oxid Med Cell Longev* (2020) 2020:3947806. doi: 10.1155/2020/3947806

40. Chen S, Zeng K, Liu QC, Guo Z, Zhang S, Chen XR, et al. Adropin deficiency worsens HFD-induced metabolic defects. *Cell Death Dis* (2017) 8(8):e3008. doi: 10.1038/cddis.2017.362

41. Gyllenhammer LE, Lam J, Alderete TL, Allayee H, Akbari O, Katkhouda N, et al. Lower omental t-regulatory cell count is associated with higher fasting glucose and lower beta-cell function in adults with obesity. *Obes (Silver Spring)* (2016) 24(6):1274–82. doi: 10.1002/oby.21507

42. Billert M, Jasaszwili M, Strowski M, Nowak KW, Skrzypski M. Adropin suppresses insulin expression and secretion in INS-1E cells and rat pancreatic islets. *J Physiol Pharmacol* (2020) 71(1). doi: 10.26402/jpp.2020.1.09

43. Jntti M, Harvey BK. Trophic activities of endoplasmic reticulum proteins CDNF and MANF. *Cell Tissue Res* (2020) 382(1):83–100. doi: 10.1007/s00441-020-03263-0

44. Glembotski CC, Thuerauf DJ, Huang C, Vekich JA, Gottlieb RA, Doroudgar S. Mesencephalic astrocyte-derived neurotrophic factor protects the heart from ischemic damage and is selectively secreted upon sarco/endoplasmic reticulum calcium depletion. *J Biol Chem* (2012) 287(31):25893–904. doi: 10.1074/jbc.M112.356345

45. Tadimalla A, Belmont PJ, Thuerauf DJ, Glassy MS, Martindale JJ, Gude N, et al. Mesencephalic astrocyte-derived neurotrophic factor is an ischemia-inducible secreted endoplasmic reticulum stress response protein in the heart. *Circ Res* (2008) 103(11):1249–58. doi: 10.1161/CIRCRESAHA.108.180679

46. Apostolou A, Shen Y, Liang Y, Luo J, Fang S. Armet, a UPR-upregulated protein, inhibits cell proliferation and ER stress-induced cell death. *Exp Cell Res* (2008) 314(13):2454–67. doi: 10.1016/j.yexcr.2008.05.001

47. Wang M, Kaufman RJ. Protein misfolding in the endoplasmic reticulum as a conduit to human disease. *Nature* (2016) 529(7586):326–35. doi: 10.1038/nature17041

48. Walter P, Ron D. The unfolded protein response: From stress pathway to homeostatic regulation. *Science* (2011) 334(6059):1081–6. doi: 10.1126/science.1209038

49. Wang C, Peng JJ, Miao H, Liu DF, Zhang LL. Decreased plasma MANF levels are associated with type 2 diabetes. *BioMed Environ Sci* (2021) 34(3):236–40. doi: 10.3967/bes2021.030

50. Hetz C, Papa FR. The unfolded protein response and cell fate control. *Mol Cell* (2018) 69(2):169–81. doi: 10.1016/j.molcel.2017.06.017

51. Tabas I, Ron D. Integrating the mechanisms of apoptosis induced by endoplasmic reticulum stress. *Nat Cell Biol* (2011) 13(3):184–90. doi: 10.1038/ncb0311-184

52. Ohta Y, Taguchi A, Matsumura T, Nakabayashi H, Akiyama M, Yamamoto K, et al. Clock gene dysregulation induced by chronic ER stress disrupts beta-cell function. *EBioMedicine* (2017) 18:146–56. doi: 10.1016/j.ebiom.2017.03.040

53. Cnop M, Ladriere L, Igoillo-Esteve M, Moura RF, Cunha DA. Causes and cures for endoplasmic reticulum stress in lipotoxic beta-cell dysfunction. *Diabetes Obes Metab* (2010) 12(Suppl 20):76–82. doi: 10.1111/j.1463-1326.2010.01279.x

54. Cnop M, Foufelle F, Velloso LA. Endoplasmic reticulum stress, obesity and diabetes. *Trends Mol Med* (2012) 18(1):59–68. doi: 10.1016/j.molmed.2011.07.010

55. Pakarinen E, Danilova T, Voikar V, Chmielarz P, Piepponen P, Airavaara M, et al. MANF ablation causes prolonged activation of the UPR without neurodegeneration in the mouse midbrain dopamine system. *eNeuro* (2020) 7(1):ENEURO.0477-19.2019. doi: 10.1523/ENEURO.0477-19.2019

56. Xu S, Di Z, He Y, Wang R, Ma Y, Sun R, et al. Mesencephalic astrocyte-derived neurotrophic factor (MANF) protects against abeta toxicity *via* attenuating abeta-induced endoplasmic reticulum stress. *J Neuroinflamm* (2019) 16(1):35. doi: 10.1186/s12974-019-1429-0

57. Wang D, Hou C, Cao Y, Cheng Q, Zhang L, Li H, et al. XBP1 activation enhances MANF expression *via* binding to endoplasmic reticulum stress response elements within MANF promoter region in hepatitis b. *Int J Biochem Cell Biol* (2018) 99:140–6. doi: 10.1016/j.biocel.2018.04.007

58. Wu T, Liu Q, Li Y, Li H, Chen L, Yang X, et al. Feeding-induced hepatokine, manf, ameliorates diet-induced obesity by promoting adipose browning *via* p38 MAPK pathway. *J Exp Med* (2021) 218(6):e20201203. doi: 10.1084/jem.20201203

59. Bourougaa K, Naski N, Boularan C, Mlynarczyk C, Candeias MM, Marullo S, et al. Endoplasmic reticulum stress induces G2 cell-cycle arrest *via* mRNA translation of the p53 isoform p53/47. *Mol Cell* (2010) 38(1):78–88. doi: 10.1016/j.molcel.2010.01.041

60. Brewer JW, Diehl JA. PERK mediates cell-cycle exit during the mammalian unfolded protein response. *Proc Natl Acad Sci U.S.A.* (2000) 97(23):12625–30. doi: 10.1073/pnas.220247197

61. Danilova T, Belevich I, Li H, Palm E, Jokitalo E, Otonkoski T, et al. MANF is required for the postnatal expansion and maintenance of pancreatic beta-cell mass in mice. *Diabetes* (2019) 68(1):66–80. doi: 10.2337/db17-1149

62. Hakonen E, Chandra V, Fogarty CL, Yu NY, Ustinov J, Katayama S, et al. MANF protects human pancreatic beta cells against stress-induced cell death. *Diabetologia* (2018) 61(10):2202–14. doi: 10.1007/s00125-018-4687-y

63. Lindahl M, Danilova T, Palm E, Lindholm P, Voikar V, Hakonen E, et al. MANF is indispensable for the proliferation and survival of pancreatic beta cells. *Cell Rep* (2014) 7(2):366–75. doi: 10.1016/j.celrep.2014.03.023

64. Cunha DA, Cito M, Grieco FA, Cosentino C, Danilova T, Ladriere L, et al. Pancreatic beta-cell protection from inflammatory stress by the endoplasmic reticulum proteins thrombospondin 1 and mesencephalic astrocyte-derived neutrotrophic factor (MANF). *J Biol Chem* (2017) 292(36):14977–88. doi: 10.1074/jbc.M116.769877

65. Chen L, Feng L, Wang X, Du J, Chen Y, Yang W, et al. Mesencephalic astrocyte-derived neurotrophic factor is involved in inflammation by negatively regulating the NF-kappaB pathway. *Sci Rep* (2015) 5:8133. doi: 10.1038/srep08133

66. Yagi T, Asada R, Kanekura K, Eesmaa A, Lindahl M, Saarma M, et al. Neuroplastin modulates anti-inflammatory effects of MANF. *iScience* (2020) 23(12):101810. doi: 10.1016/j.isci.2020.101810

67. Wu H, Li H, Wen W, Wang Y, Xu H, Xu M, et al. MANF protects pancreatic acinar cells against alcohol-induced endoplasmic reticulum stress and cellular injury. *J Hepatobiliary Pancreat Sci* (2021) 28(10):883–92. doi: 10.1002/jhbp.928

68. Montaser H, Patel KA, Balboa D, Ibrahim H, Lithovius V, Naatanen A, et al. Loss of MANF causes childhood-onset syndromic diabetes due to increased endoplasmic reticulum stress. *Diabetes* (2021) 70(4):1006–18. doi: 10.2337/db20-1174

69. Parkash V, Lindholm P, Peranen J, Kalkkinen N, Oksanen E, Saarma M, et al. The structure of the conserved neurotrophic factors MANF and CDNF explains why they are bifunctional. *Protein Eng Des Sel* (2009) 22(4):233–41. doi: 10.1093/protein/gzn080

70. Bruhn H. A short guided tour through functional and structural features of saposin-like proteins. *Biochem J* (2005) 389(Pt 2):249–57. doi: 10.1042/BJ20050051

71. Bai M, Vozdek R, Hnizda A, Jiang C, Wang B, Kuchar L, et al. Conserved roles of c. elegans and human MANFs in sulfatide binding and cytoprotection. *Nat Commun* (2018) 9(1):897. doi: 10.1038/s41467-018-03355-0

72. Sousa-Victor P, Neves J, Cedron-Craft W, Ventura PB, Liao CY, Riley RR, et al. MANF regulates metabolic and immune homeostasis in ageing and protects against liver damage. *Nat Metab* (2019) 1(2):276–90. doi: 10.1038/s42255-018-0023-6

73. He M, Wang C, Long XH, Peng JJ, Liu DF, Yang GY, et al. Mesencephalic astrocyte-derived neurotrophic factor ameliorates steatosis in HepG2 cells by regulating hepatic lipid metabolism. *World J Gastroenterol* (2020) 26(10):1029–41. doi: 10.3748/wjg.v26.i10.1029

74. Zhang G, Liu Q, Li Y, Huang C, Zhou J, Zhao Y, et al. Mesencephalic astrocyte-derived neurotrophic factor alleviates alcohol induced hepatic steatosis *via* activating Stat3-mediated autophagy. *Biochem Biophys Res Commun* (2021) 550:197–203. doi: 10.1016/j.bbrc.2021.02.123

75. Zhang X, Heckmann BL, Campbell LE, Liu J. G0S2: A small giant controller of lipolysis and adipose-liver fatty acid flux. *Biochim Biophys Acta Mol Cell Biol Lipids* (2017) 1862(10 Pt B):1146–54. doi: 10.1016/j.bbalip.2017.06.007

76. Liu J, Wu Z, Han D, Wei C, Liang Y, Jiang T, et al. Mesencephalic astrocyte-derived neurotrophic factor inhibits liver cancer through small ubiquitin-related modifier (SUMO)ylation-related suppression of NF-kappaB/Snail signaling pathway and epithelial-mesenchymal transition. *Hepatology* (2020) 71(4):1262–78. doi: 10.1002/hep.30917

77. Yang S, Yang H, Chang R, Yin P, Yang Y, Yang W, et al. MANF regulates hypothalamic control of food intake and body weight. *Nat Commun* (2017) 8(1):579. doi: 10.1038/s41467-017-00750-x

78. Wang H, Ke Z, Alimov A, Xu M, Frank JA, Fang S, et al. Spatiotemporal expression of MANF in the developing rat brain. *PloS One* (2014) 9(2):e90433. doi: 10.1371/journal.pone.0090433

79. Cakir I, Nillni EA. Endoplasmic reticulum stress, the hypothalamus, and energy balance. *Trends Endocrinol Metab* (2019) 30(3):163–76. doi: 10.1016/j.tem.2019.01.002

80. Tang Q, Liu Q, Li J, Yan J, Jing X, Zhang J, et al. MANF in POMC neurons promotes brown adipose tissue thermogenesis and protects against diet-induced obesity. *Diabetes* (2022) 71(11):2344–59. doi: 10.2337/db21-1128

81. Wang X, Li W, Zhou Q, Li J, Wang X, Zhang J, et al. MANF promotes diabetic corneal epithelial wound healing and nerve regeneration by attenuating hyperglycemia-induced endoplasmic reticulum stress. *Diabetes* (2020) 69(6):1264–78. doi: 10.2337/db19-0835

82. Danilova T, Galli E, Pakarinen E, Palm E, Lindholm P, Saarma M, et al. Mesencephalic astrocyte-derived neurotrophic factor (MANF) is highly expressed in mouse tissues with metabolic function. *Front Endocrinol (Lausanne)* (2019) 10:765. doi: 10.3389/fendo.2019.00765

83. Bartke A, Sun LY, Longo V. Somatotropic signaling: trade-offs between growth, reproductive development, and longevity. *Physiol Rev* (2013) 93(2):571–98. doi: 10.1152/physrev.00006.2012

84. Furigo IC, Teixeira PDS, de Souza GO, Couto GCL, Romero GG, Perello M, et al. Growth hormone regulates neuroendocrine responses to weight loss *via* AgRP neurons. *Nat Commun* (2019) 10(1):662. doi: 10.1038/s41467-019-08607-1

85. Zhong C, Song Y, Wang Y, Zhang T, Duan M, Li Y, et al. Increased food intake in growth hormone-transgenic common carp (Cyprinus carpio l.) may be mediated by upregulating agouti-related protein (AgRP). *Gen Comp Endocrinol* (2013) 192:81–8. doi: 10.1016/j.ygcen.2013.03.024

86. Krause A, Sillard R, Kleemeier B, Kluver E, Maronde E, Conejo-Garcia JR, et al. Isolation and biochemical characterization of LEAP-2, a novel blood peptide expressed in the liver. *Protein Sci* (2003) 12(1):143–52. doi: 10.1110/ps.0213603

87. Mani BK, Puzziferri N, He Z, Rodriguez JA, Osborne-Lawrence S, Metzger NP, et al. LEAP2 changes with body mass and food intake in humans and mice. *J Clin Invest* (2019) 129(9):3909–23. doi: 10.1172/JCI125332

88. Kojima M, Hosoda H, Date Y, Nakazato M, Matsuo H, Kangawa K. Ghrelin is a growth-hormone-releasing acylated peptide from stomach. *Nature* (1999) 402 (6762):656–60. doi: 10.1038/45230

89. Zhao TJ, Liang G, Li RL, Xie X, Sleeman MW, Murphy AJ, et al. Ghrelin O-acyltransferase (GOAT) is essential for growth hormone-mediated survival of calorie-restricted mice. *Proc Natl Acad Sci U.S.A.* (2010) 107(16):7467–72. doi: 10.1073/pnas.1002271107

90. Yanagi S, Sato T, Kangawa K, Nakazato M. The homeostatic force of ghrelin. *Cell Metab* (2018) 27(4):786–804. doi: 10.1016/j.cmet.2018.02.008

91. Mani BK, Zigman JM. Ghrelin as a survival hormone. *Trends Endocrinol Metab* (2017) 28(12):843–54. doi: 10.1016/j.tem.2017.10.001

92. McFarlane MR, Brown MS, Goldstein JL, Zhao TJ. Induced ablation of ghrelin cells in adult mice does not decrease food intake, body weight, or response to high-fat diet. *Cell Metab* (2014) 20(1):54–60. doi: 10.1016/j.cmet.2014.04.007

93. Wang JH, Li HZ, Shao XX, Nie WH, Liu YL, Xu ZG, et al. Identifying the binding mechanism of LEAP2 to receptor GHSR1a. *FEBS J* (2019) 286(7):1332–45. doi: 10.1111/febs.14763

94. Ge X, Yang H, Bednarek MA, Galon-Tilleman H, Chen P, Chen M, et al. LEAP2 is an endogenous antagonist of the ghrelin receptor. *Cell Metab* (2018) 27(2):461–9 e6. doi: 10.1016/j.cmet.2017.10.016

95. M'Kadmi C, Cabral A, Barrile F, Giribaldi J, Cantel S, Damian M, et al. N-terminal liver-expressed antimicrobial peptide 2 (LEAP2) region exhibits inverse agonist activity toward the ghrelin receptor. *J Med Chem* (2019) 62(2):965–73. doi: 10.1021/acs.jmedchem.8b01644

96. Li J, Huang P, Xiong J, Liang X, Li M, Ke H, et al. Serum levels of ghrelin and LEAP2 in patients with type 2 diabetes mellitus: correlation with circulating glucose and lipids. *Endocr Connect* (2022) 11(5):e220012. doi: 10.1530/EC-22-0012

97. Schalla MA, Stengel A. Pharmacological modulation of ghrelin to induce weight loss: Successes and challenges. *Curr Diabetes Rep* (2019) 19(10):102. doi: 10.1007/s11892-019-1211-9

98. Islam MN, Mita Y, Maruyama K, Tanida R, Zhang W, Sakoda H, et al. Liver-expressed antimicrobial peptide 2 antagonizes the effect of ghrelin in rodents. *J Endocrinol* (2020) 244(1):13–23. doi: 10.1530/JOE-19-0102

99. Zigman JM, Jones JE, Lee CE, Saper CB, Elmquist JK. Expression of ghrelin receptor mRNA in the rat and the mouse brain. *J Comp Neurol* (2006) 494(3):528–48. doi: 10.1002/cne.20823

100. Cornejo MP, Mustafa ER, Barrile F, Cassano D, De Francesco PN, Raingo J, et al. The intriguing ligand-dependent and ligand-independent actions of the growth hormone secretagogue receptor on reward-related behaviors. *Neurosci Biobehav Rev* (2021) 120:401–16. doi: 10.1016/j.neubiorev.2020.10.017

101. Lee JH, Lin L, Xu P, Saito K, Wei Q, Meadows AG, et al. Neuronal deletion of ghrelin receptor almost completely prevents diet-induced obesity. *Diabetes* (2016) 65 (8):2169–78. doi: 10.2337/db15-1587

102. Lopez Soto EJ, Agosti F, Cabral A, Mustafa ER, Damonte VM, Gandini MA, et al. Constitutive and ghrelin-dependent GHSR1a activation impairs CaV2.1 and CaV2.2 currents in hypothalamic neurons. *J Gen Physiol* (2015) 146(3):205–19. doi: 10.1085/jgp.201511383

103. Mustafa ER, Cordisco Gonzalez S, Damian M, Cantel S, Denoyelle S, Wagner R, et al. LEAP2 impairs the capability of the growth hormone secretagogue receptor to regulate the dopamine 2 receptor signaling. *Front Pharmacol* (2021) 12:712437. doi: 10.3389/fphar.2021.712437

104. Cornejo MP, Castrogiovanni D, Schioth HB, Reynaldo M, Marie J, Fehrentz JA, et al. Growth hormone secretagogue receptor signalling affects high-fat intake independently of plasma levels of ghrelin and LEAP2, in a 4-day binge eating model. *J Neuroendocrinol* (2019) 31(10):e12785. doi: 10.1111/jne.12785

105. Shankar K, Metzger NP, Singh O, Mani BK, Osborne-Lawrence S, Varshney S, et al. LEAP2 deletion in mice enhances ghrelin's actions as an orexigen and growth hormone secretagogue. *Mol Metab* (2021) 53:101327. doi: 10.1016/j.molmet.2021.101327

106. Shen M, Manca C, Suriano F, Nallabelli N, Pechereau F, Allam-Ndoul B, et al. Three of a kind: Control of the expression of liver-expressed antimicrobial peptide 2 (LEAP2) by the endocannabinoidome and the gut microbiome. *Molecules* (2021) 27 (1):1. doi: 10.3390/molecules27010001

107. Al-Massadi O, Muller T, Tschop M, Dieguez C, Nogueiras R. Ghrelin and LEAP-2: Rivals in energy metabolism. *Trends Pharmacol Sci* (2018) 39(8):685–94. doi: 10.1016/j.tips.2018.06.004

108. Lugilde J, Casado S, Beiroa D, Cunarro J, Garcia-Lavandeira M, Alvarez CV, et al. LEAP-2 counteracts ghrelin-induced food intake in a nutrient, growth hormone and age independent manner. *Cells* (2022) 11(3):324. doi: 10.3390/cells11030324

109. Gupta D, Dowsett GKC, Mani BK, Shankar K, Osborne-Lawrence S, Metzger NP, et al. High coexpression of the ghrelin and LEAP2 receptor GHSR with pancreatic polypeptide in mouse and human islets. *Endocrinology* (2021) 162(10):bqab148. doi: 10.1210/endocr/bqab148

110. Dezaki K, Hosoda H, Kakei M, Hashiguchi S, Watanabe M, Kangawa K, et al. Endogenous ghrelin in pancreatic islets restricts insulin release by attenuating Ca2+ signaling in beta-cells: Implication in the glycemic control in rodents. *Diabetes* (2004) 53(12):3142–51. doi: 10.2337/diabetes.53.12.3142

111. Egido EM, Rodriguez-Gallardo J, Silvestre RA, Marco J. Inhibitory effect of ghrelin on insulin and pancreatic somatostatin secretion. *Eur J Endocrinol* (2002) 146 (2):241–4. doi: 10.1530/eje.0.1460241

112. Colombo M, Gregersen S, Xiao J, Hermansen K. Effects of ghrelin and other neuropeptides (CART, MCH, orexin a and b, and GLP-1) on the release of insulin from isolated rat islets. *Pancreas* (2003) 27(2):161–6. doi: 10.1097/00006676-200308000-00009

113. Reimer MK, Pacini G, Ahren B. Dose-dependent inhibition by ghrelin of insulin secretion in the mouse. *Endocrinology* (2003) 144(3):916–21. doi: 10.1210/en.2002-220819

114. Bayle M, Peraldi-Roux S, Gautheron G, Cros G, Oiry C, Neasta J. Liver-expressed antimicrobial peptide 2 antagonizes the insulinostatic effect of ghrelin in rat isolated pancreatic islets. *Fundam Clin Pharmacol* (2022) 36(2):375–7. doi: 10.1111/fcp.12722

115. Hagemann CA, Zhang C, Hansen HH, Jorsal T, Rigbolt KTG, Madsen MR, et al. Identification and metabolic profiling of a novel human gut-derived LEAP2 fragment. *J Clin Endocrinol Metab* (2021) 106(2):e966–e81. doi: 10.1210/clinem/dgaa803

116. Dezaki K, Sone H, Yada T. Ghrelin is a physiological regulator of insulin release in pancreatic islets and glucose homeostasis. *Pharmacol Ther* (2008) 118(2):239–49. doi: 10.1016/j.pharmthera.2008.02.008

117. Horton JD, Cohen JC, Hobbs HH. Molecular biology of PCSK9: its role in LDL metabolism. *Trends Biochem Sci* (2007) 32(2):71–7. doi: 10.1016/j.tibs.2006.12.008

118. Stoekenbroek RM, Lambert G, Cariou B, Hovingh GK. Inhibiting PCSK9 - biology beyond LDL control. *Nat Rev Endocrinol* (2018) 15(1):52–62. doi: 10.1038/s41574-018-0110-5

119. Cariou B, Si-Tayeb K, Le May C. Role of PCSK9 beyond liver involvement. *Curr Opin Lipidol* (2015) 26(3):155–61. doi: 10.1097/MOL.0000000000000180

120. Cariou B, Langhi C, Le Bras M, Bortolotti M, Le KA, Theytaz F, et al. Plasma PCSK9 concentrations during an oral fat load and after short term high-fat, high-fat high-protein and high-fructose diets. *Nutr Metab (Lond)* (2013) 10(1):4. doi: 10.1186/1743-7075-10-4

121. Costet P, Cariou B, Lambert G, Lalanne F, Lardeux B, Jarnoux AL, et al. Hepatic PCSK9 expression is regulated by nutritional status *via* insulin and sterol

regulatory element-binding protein 1c. *J Biol Chem* (2006) 281(10):6211–8. doi: 10.1074/jbc.M508582200

122. Lamantia V, Bissonnette S, Wassef H, Cyr Y, Baass A, Dufour R, et al. ApoB-lipoproteins and dysfunctional white adipose tissue: Relation to risk factors for type 2 diabetes in humans. *J Clin Lipidol* (2017) 11(1):34–45 e2. doi: 10.1016/j.jacl.2016.09.013

123. Faraj M. LDL, LDL receptors, and PCSK9 as modulators of the risk for type 2 diabetes: A focus on white adipose tissue. *J BioMed Res* (2020) 34(4):251–9. doi: 10.7555/JBR.34.20190124

124. Gastaldelli A, Gaggini M, DeFronzo RA. Role of adipose tissue insulin resistance in the natural history of type 2 diabetes: Results from the San Antonio metabolism study. *Diabetes* (2017) 66(4):815–22. doi: 10.2337/db16-1167

125. Cyr Y, Lamantia V, Bissonnette S, Burnette M, Besse-Patin A, Demers A, et al. Lower plasma PCSK9 in normocholesterolemic subjects is associated with upregulated adipose tissue surface-expression of LDLR and CD36 and NLRP3 inflammasome. *Physiol Rep* (2021) 9(3):e14721. doi: 10.14814/phy2.14721

126. Bissonnette S, Salem H, Wassef H, Saint-Pierre N, Tardif A, Baass A, et al. Low density lipoprotein delays clearance of triglyceride-rich lipoprotein by human subcutaneous adipose tissue. *J Lipid Res* (2013) 54(5):1466–76. doi: 10.1194/jlr.P023176

127. Masella R, Vari R, D'Archivio M, Santangelo C, Scazzocchio B, Maggiorella MT, et al. Oxidised LDL modulate adipogenesis in 3T3-L1 preadipocytes by affecting the balance between cell proliferation and differentiation. *FEBS Lett* (2006) 580 (10):2421–9. doi: 10.1016/j.febslet.2006.03.068

128. Calvo D, Gomez-Coronado D, Suarez Y, Lasuncion MA, Vega MA. Human CD36 is a high affinity receptor for the native lipoproteins HDL, LDL, and VLDL. *J Lipid Res* (1998) 39(4):777–88. doi: 10.1016/S0022-2275(20)32566-9

129. Kuniyasu A, Hayashi S, Nakayama H. Adipocytes recognize and degrade oxidized low density lipoprotein through CD36. *Biochem Biophys Res Commun* (2002) 295(2):319–23. doi: 10.1016/s0006-291x(02)00666-6

130. Sheedy FJ, Grebe A, Rayner KJ, Kalantari P, Ramkhelawon B, Carpenter SB, et al. CD36 coordinates NLRP3 inflammasome activation by facilitating intracellular nucleation of soluble ligands into particulate ligands in sterile inflammation. *Nat Immunol* (2013) 14(8):812–20. doi: 10.1038/ni.2639

131. Rampanelli E, Orso E, Ochodnicky P, Liebisch G, Bakker PJ, Claessen N, et al. Metabolic injury-induced NLRP3 inflammasome activation dampens phospholipid degradation. *Sci Rep* (2017) 7(1):2861. doi: 10.1038/s41598-017-01994-9

132. Skeldon AM, Faraj M, Saleh M. Caspases and inflammasomes in metabolic inflammation. *Immunol Cell Biol* (2014) 92(4):304–13. doi: 10.1038/icb.2014.5

133. Stienstra R, Joosten LA, Koenen T, van Tits B, van Diepen JA, van den Berg SA, et al. The inflammasome-mediated caspase-1 activation controls adipocyte differentiation and insulin sensitivity. *Cell Metab* (2010) 12(6):593–605. doi: 10.1016/j.cmet.2010.11.011

134. Demers A, Samami S, Lauzier B, Des Rosiers C, Ngo Sock ET, Ong H, et al. PCSK9 induces CD36 degradation and affects long-chain fatty acid uptake and triglyceride metabolism in adipocytes and in mouse liver. *Arterioscler Thromb Vasc Biol* (2015) 35(12):2517–25. doi: 10.1161/ATVBAHA.115.306032

135. Roubtsova A, Munkonda MN, Awan Z, Marcinkiewicz J, Chamberland A, Lazure C, et al. Circulating proprotein convertase subtilisin/kexin 9 (PCSK9) regulates VLDLR protein and triglyceride accumulation in visceral adipose tissue. *Arterioscler Thromb Vasc Biol* (2011) 31(4):785–91. doi: 10.1161/ATVBAHA.110.220988

136. Baragetti A, Balzarotti G, Grigore L, Pellegatta F, Guerrini U, Pisano G, et al. PCSK9 deficiency results in increased ectopic fat accumulation in experimental models and in humans. *Eur J Prev Cardiol* (2017) 24(17):1870–7. doi: 10.1177/2047487317724342

137. Onat A, Can G, Hergenc G, Yazici M, Karabulut A, Albayrak S. Serum apolipoprotein b predicts dyslipidemia, metabolic syndrome and, in women, hypertension and diabetes, independent of markers of central obesity and inflammation. *Int J Obes (Lond)* (2007) 31(7):1119–25. doi: 10.1038/sj.ijo.0803552

138. Hwang YC, Ahn HY, Park SW, Park CY. Apolipoprotein b and non-HDL cholesterol are more powerful predictors for incident type 2 diabetes than fasting glucose or glycated hemoglobin in subjects with normal glucose tolerance: A 3.3-year retrospective longitudinal study. *Acta Diabetol* (2014) 51(6):941–6. doi: 10.1007/s00592-014-0587-x

139. Wassef H, Bissonnette S, Saint-Pierre N, Lamantia V, Cyr Y, Chretien M, et al. The apoB-to-PCSK9 ratio: A new index for metabolic risk in humans. *J Clin Lipidol* (2015) 9(5):664–75. doi: 10.1016/j.jacl.2015.06.012

140. Bissonnette S, Saint-Pierre N, Lamantia V, Cyr Y, Wassef H, Faraj M. Plasma IL-1Ra: Linking hyperapoB to risk factors for type 2 diabetes independent of obesity in humans. *Nutr Diabetes* (2015) 5:e180. doi: 10.1038/nutd.2015.30

141. Cyr Y, Bissonnette S, Lamantia V, Wassef H, Loizon E, Ngo Sock ET, et al. White adipose tissue surface expression of LDLR and CD36 is associated with risk factors for type 2 diabetes in adults with obesity. *Obes (Silver Spring)* (2020) 28 (12):2357–67. doi: 10.1002/oby.22985

142. Churchward MA, Coorssen JR. Cholesterol, regulated exocytosis and the physiological fusion machine. *Biochem J* (2009) 423(1):1–14. doi: 10.1042/BJ20090969

143. Hao M, Head WS, Gunawardana SC, Hasty AH, Piston DW. Direct effect of cholesterol on insulin secretion: A novel mechanism for pancreatic beta-cell dysfunction. *Diabetes* (2007) 56(9):2328–38. doi: 10.2337/db07-0056

144. Perego C, Da Dalt L, Pirillo A, Galli A, Catapano AL, Norata GD. Cholesterol metabolism, pancreatic beta-cell function and diabetes. *Biochim Biophys Acta Mol Basis Dis* (2019) 1865(9):2149–56. doi: 10.1016/j.bbadis.2019.04.012

145. Roehrich ME, Mooser V, Lenain V, Herz J, Nimpf J, Azhar S, et al. Insulin-secreting beta-cell dysfunction induced by human lipoproteins. *J Biol Chem* (2003) 278 (20):18368–75. doi: 10.1074/jbc.M300102200

146. Cnop M. Fatty acids and glucolipotoxicity in the pathogenesis of type 2 diabetes. *Biochem Soc Trans* (2008) 36(Pt 3):348–52. doi: 10.1042/BST0360348

147. Da Dalt L, Ruscica M, Bonacina F, Balzarotti G, Dhyani A, Di Cairano E, et al. PCSK9 deficiency reduces insulin secretion and promotes glucose intolerance: the role of the low-density lipoprotein receptor. *Eur Heart J* (2019) 40(4):357–68. doi: 10.1093/eurheartj/ehy357

148. Mbikay M, Sirois F, Mayne J, Wang GS, Chen A, Dewpura T, et al. PCSK9-deficient mice exhibit impaired glucose tolerance and pancreatic islet abnormalities. *FEBS Lett* (2010) 584(4):701–6. doi: 10.1016/j.febslet.2009.12.018

149. Ramin-Mangata S, Thedrez A, Nativel B, Diotel N, Blanchard V, Wargny M, et al. Effects of proprotein convertase subtilisin kexin type 9 modulation in human pancreatic beta cells function. *Atherosclerosis* (2021) 326:47–55. doi: 10.1016/j.atherosclerosis.2021.03.044

150. Peyot ML, Roubtsova A, Lussier R, Chamberland A, Essalmani R, Murthy Madiraju SR, et al. Substantial PCSK9 inactivation in beta-cells does not modify glucose homeostasis or insulin secretion in mice. *Biochim Biophys Acta Mol Cell Biol Lipids* (2021) 1866(8):158968. doi: 10.1016/j.bbalip.2021.158968

151. Zang H, Jiang F, Cheng X, Xu H, Hu X. Serum adropin levels are decreased in Chinese type 2 diabetic patients and negatively correlated with body mass index. *Endocr J* (2018) 65(7):685–91. doi: 10.1507/endocrj.EJ18-0060

152. Li L, Xie W, Zheng XL, Yin WD, Tang CK. A novel peptide adropin in cardiovascular diseases. *Clin Chim Acta* (2016) 453:107–13. doi: 10.1016/j.cca.2015.12.010

153. Ticinovic Kurir T, Milicevic T, Novak A, Vilovic M, Bozic J. Adropin - potential link in cardiovascular protection for obese Male type 2 diabetes mellitus patients treated with liraglutide. *Acta Clin Croat* (2020) 59(2):344–50. doi: 10.20471/acc.2020.59.02.19

154. Akcilar R, Kocak FE, Simsek H, Akcilar A, Bayat Z, Ece E, et al. Antidiabetic and hypolipidemic effects of adropinin streoptozotocin-induced type 2 diabetic rats. *Bratisl Lek Listy* (2016) 117(2):100–5. doi: 10.4149/bll_2016_020

155. Li N, Xie G, Zhou B, Qu A, Meng H, Liu J, et al. Serum adropin as a potential biomarker for predicting the development of type 2 diabetes mellitus in individuals with metabolic dysfunction-associated fatty liver disease. *Front Physiol* (2021) 12:696163. doi: 10.3389/fphys.2021.696163

156. Li B, Tian X, Guo S, Zhang M, Li J, Zhai N, et al. Pentraxin-3 and adropin as inflammatory markers of early renal damage in type 2 diabetes patients. *Int Urol Nephrol* (2020) 52(11):2145–52. doi: 10.1007/s11255-020-02568-x

157. Stelcer E, Milecka P, Komarowska H, Jopek K, Tyczewska M, Szyszka M, et al. Adropin stimulates proliferation and inhibits adrenocortical steroidogenesis in the human adrenal carcinoma (HAC15) cell line. *Front Endocrinol (Lausanne)* (2020) 11:561370. doi: 10.3389/fendo.2020.561370

158. Sunena, Mishra DN. Stress etiology of type 2 diabetes. *Curr Diabetes Rev* (2022) 18(9):e240222201413. doi: 10.2174/1573399818666220224140934

159. Gumus Balikcioglu P, Balikcioglu M, Soros A, Chalew S. The 24-hour average concentration of cortisol is elevated in obese African-American youth with type 2 diabetes. *J Diabetes Complicat* (2021) 35(7):107933. doi: 10.1016/j.jdiacomp.2021.107933

160. Zavatta G, Casadio E, Rinaldi E, Pagotto U, Pasquali R, Vicennati V. Aldosterone and type 2 diabetes mellitus. *Horm Mol Biol Clin Investig* (2016) 26 (1):53–9. doi: 10.1515/hmbci-2015-0065

161. Stein LM, Yosten GL, Samson WK. Adropin acts in brain to inhibit water drinking: Potential interaction with the orphan G protein-coupled receptor, GPR19. *Am J Physiol Regul Integr Comp Physiol* (2016) 310(6):R476–80. doi: 10.1152/ajpregu.00511.2015

162. Rao A, Herr DR. G Protein-coupled receptor GPR19 regulates e-cadherin expression and invasion of breast cancer cells. *Biochim Biophys Acta Mol Cell Res* (2017) 1864(7):1318–27. doi: 10.1016/j.bbamcr.2017.05.001

163. Foster SR, Hauser AS, Vedel L, Strachan RT, Huang XP, Gavin AC, et al. Discovery of human signaling systems: Pairing peptides to G protein-coupled receptors. *Cell* (2019) 179(4):895–908 e21. doi: 10.1016/j.cell.2019.10.010

164. Wong CM, Wang Y, Lee JT, Huang Z, Wu D, Xu A, et al. Adropin is a brain membrane-bound protein regulating physical activity *via* the NB-3/Notch signaling pathway in mice. *J Biol Chem* (2014) 289(37):25976–86. doi: 10.1074/jbc.M114.576058

165. Loewen SP, Ferguson AV. Adropin acts in the rat paraventricular nucleus to influence neuronal excitability. *Am J Physiol Regul Integr Comp Physiol* (2017) 312(4): R511–R9. doi: 10.1152/ajpregu.00517.2016

166. Swanson LW, Sawchenko PE. Paraventricular nucleus: A site for the integration of neuroendocrine and autonomic mechanisms. *Neuroendocrinology* (1980) 31(6):410–7. doi: 10.1159/000123111

167. Sutton AK, Myers MGJr., Olson DP. The role of PVH circuits in leptin action and energy balance. *Annu Rev Physiol* (2016) 78:207–21. doi: 10.1146/annurev-physiol-021115-105347

168. Amorim ED, Peras VR, de Andrade O, Martins-Pinge MC. Functional evidence of paraventricular nucleus involvement in cardiovascular and autonomic modulation in response to acute microgravity (head-down tilt) in unanesthetized rats. *J Neurosci Res* (2015) 93(8):1305–12. doi: 10.1002/jnr.23586

169. Yang S, Li S, Li XJ. MANF: A new player in the control of energy homeostasis, and beyond. *Front Physiol* (2018) 9:1725. doi: 10.3389/fphys.2018.01725

170. Wu T, Zhang F, Yang Q, Zhang Y, Liu Q, Jiang W, et al. Circulating mesencephalic astrocyte-derived neurotrophic factor is increased in newly diagnosed prediabetic and diabetic patients, and is associated with insulin resistance. *Endocr J* (2017) 64(4):403–10. doi: 10.1507/endocrj.EJ16-0472

171. Sun Y, Wang P, Zheng H, Smith RG. Ghrelin stimulation of growth hormone release and appetite is mediated through the growth hormone secretagogue receptor. *Proc Natl Acad Sci U.S.A.* (2004) 101(13):4679–84. doi: 10.1073/pnas.0305930101

172. Xiao X, Bi M, Jiao Q, Chen X, Du X, Jiang H. A new understanding of GHSR1a–independent of ghrelin activation. *Ageing Res Rev* (2020) 64:101187. doi: 10.1016/j.arr.2020.101187

173. Gupta D, Ogden SB, Shankar K, Varshney S, Zigman JM. "A LEAP 2 conclusions? targeting the ghrelin system to treat obesity and diabetes". *Mol Metab* (2021) 46:101128. doi: 10.1016/j.molmet.2020.101128

174. Hagemann CA, Jensen MS, Holm S, Gasbjerg LS, Byberg S, Skov-Jeppesen K, et al. LEAP2 reduces postprandial glucose excursions and ad libitum food intake in healthy men. *Cell Rep Med* (2022) 3(4):100582. doi: 10.1016/j.xcrm.2022.100582

175. Zorrilla EP, Iwasaki S, Moss JA, Chang J, Otsuji J, Inoue K, et al. Vaccination against weight gain. *Proc Natl Acad Sci U.S.A.* (2006) 103(35):13226–31. doi: 10.1073/pnas.0605376103

176. Holst B, Cygankiewicz A, Jensen TH, Ankersen M, Schwartz TW. High constitutive signaling of the ghrelin receptor–identification of a potent inverse agonist. *Mol Endocrinol* (2003) 17(11):2201–10. doi: 10.1210/me.2003-0069

177. Allas S, Delale T, Ngo N, Julien M, Sahakian P, Ritter J, et al. Safety, tolerability, pharmacokinetics and pharmacodynamics of AZP-531, a first-in-class analogue of unacylated ghrelin, in healthy and overweight/obese subjects and subjects with type 2 diabetes. *Diabetes Obes Metab* (2016) 18(9):868–74. doi: 10.1111/dom.12675

178. Abizaid A, Hougland JL. Ghrelin signaling: GOAT and GHS-R1a take a LEAP in complexity. *Trends Endocrinol Metab* (2020) 31(2):107–17. doi: 10.1016/j.tem.2019.09.006

179. Lambert G, Sjouke B, Choque B, Kastelein JJ, Hovingh GK. The PCSK9 decade. *J Lipid Res* (2012) 53(12):2515–24. doi: 10.1194/jlr.R026658

180. Tomlinson B, Patil NG, Fok M, Lam CWK. Managing dyslipidemia in patients with type 2 diabetes. *Expert Opin Pharmacother* (2021) 22(16):2221–34. doi: 10.1080/14656566.2021.1912734

181. Colhoun HM, Leiter LA, Muller-Wieland D, Cariou B, Ray KK, Tinahones FJ, et al. Effect of alirocumab on individuals with type 2 diabetes, high triglycerides, and low high-density lipoprotein cholesterol. *Cardiovasc Diabetol* (2020) 19(1):14. doi: 10.1186/s12933-020-0991-1

182. Lorenzatti AJ, Monsalvo ML, Lopez JAG, Wang H, Rosenson RS. Effects of evolocumab in individuals with type 2 diabetes with and without atherogenic dyslipidemia: An analysis from BANTING and BERSON. *Cardiovasc Diabetol* (2021) 20(1):94. doi: 10.1186/s12933-021-01287-6

183. Ramin-Mangata S, Wargny M, Pichelin M, Le May C, Thedrez A, Blanchard V, et al. Circulating PCSK9 levels are not associated with the conversion to type 2 diabetes. *Atherosclerosis* (2020) 293:49–56. doi: 10.1016/j.atherosclerosis.2019.11.027

184. Xu S, Luo S, Zhu Z, Xu J. Small molecules as inhibitors of PCSK9: Current status and future challenges. *Eur J Med Chem* (2019) 162:212–33. doi: 10.1016/j.ejmech.2018.11.011

185. Warden BA, Fazio S, Shapiro MD. The PCSK9 revolution: Current status, controversies, and future directions. *Trends Cardiovasc Med* (2020) 30(3):179–85. doi: 10.1016/j.tcm.2019.05.007

186. Nishikido T, Ray KK. Non-antibody approaches to proprotein convertase subtilisin kexin 9 inhibition: siRNA, antisense oligonucleotides, adnectins, vaccination, and new attempts at small-molecule inhibitors based on new discoveries. *Front Cardiovasc Med* (2018) 5:199. doi: 10.3389/fcvm.2018.00199

187. Ray KK, Del Prato S, Muller-Wieland D, Cariou B, Colhoun HM, Tinahones FJ, et al. Alirocumab therapy in individuals with type 2 diabetes mellitus and atherosclerotic cardiovascular disease: Analysis of the ODYSSEY DM-DYSLIPIDEMIA and DM-INSULIN studies. *Cardiovasc Diabetol* (2019) 18(1):149. doi: 10.1186/s12933-019-0951-9

188. Rosenson RS, Daviglus ML, Handelsman Y, Pozzilli P, Bays H, Monsalvo ML, et al. Efficacy and safety of evolocumab in individuals with type 2 diabetes mellitus: Primary results of the randomised controlled BANTING study. *Diabetologia* (2019) 62(6):948–58. doi: 10.1007/s00125-019-4856-7

189. Filippatos TD, Filippas-Ntekouan S, Pappa E, Panagiotopoulou T, Tsimihodimos V, Elisaf MS. PCSK9 and carbohydrate metabolism: A double-edged sword. *World J Diabetes* (2017) 8(7):311–6. doi: 10.4239/wjd.v8.i7.311

190. Lin J, Jiang X, Dong M, Liu X, Shen Q, Huang Y, et al. Hepatokine pregnancy zone protein governs the diet-induced thermogenesis through activating brown adipose tissue. *Adv Sci (Weinh)* (2021) 8(21):e2101991. doi: 10.1002/advs.202101991

191. Wang Q, Zhang P, Cakir I, Mi L, Cone RD, Lin JD. Deletion of the feeding-induced hepatokine TSK ameliorates the melanocortin obesity syndrome. *Diabetes* (2021) 70(9):2081–91. doi: 10.2337/db21-0161

192. Chen S, Feng M, Zhang S, Dong Z, Wang Y, Zhang W, et al. Angptl8 mediates food-driven resetting of hepatic circadian clock in mice. *Nat Commun* (2019) 10(1):3518. doi: 10.1038/s41467-019-11513-1

193. Martinez-Perez B, Ejarque M, Gutierrez C, Nunez-Roa C, Roche K, Vila-Bedmar R, et al. Angiopoietin-like protein 8 (ANGPTL8) in pregnancy: A brown adipose tissue-derived endocrine factor with a potential role in fetal growth. *Transl Res* (2016) 178:1–12. doi: 10.1016/j.trsl.2016.06.012

194. Arefanian H, Al-Khairi I, Khalaf NA, Cherian P, Kavalakatt S, Madhu D, et al. Increased expression level of ANGPTL8 in white adipose tissue under acute and chronic cold treatment. *Lipids Health Dis* (2021) 20(1):117. doi: 10.1186/s12944-021-01547-0

195. Markan KR, Naber MC, Ameka MK, Anderegg MD, Mangelsdorf DJ, Kliewer SA, et al. Circulating FGF21 is liver derived and enhances glucose uptake during refeeding and overfeeding. *Diabetes* (2014) 63(12):4057–63. doi: 10.2337/db14-0595

196. Uebanso T, Taketani Y, Yamamoto H, Amo K, Ominami H, Arai H, et al. Paradoxical regulation of human FGF21 by both fasting and feeding signals: Is FGF21 a nutritional adaptation factor? *PloS One* (2011) 6(8):e22976. doi: 10.1371/journal.pone.0022976

197. Zhou B, Luo Y, Ji N, Hu C, Lu Y. Orosomucoid 2 maintains hepatic lipid homeostasis through suppression of *de novo* lipogenesis. *Nat Metab* (2022) 4(9):1185–201. doi: 10.1038/s42255-022-00627-4

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Laughter yoga as an enjoyable therapeutic approach for glycemic control in individuals with type 2 diabetes: A randomized controlled trial

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Background: Laughter has been reported to have various health benefits. However, data on the long-term effects of laughter interventions on diabetes are limited. This study aimed to investigate whether laughter yoga can improve glycemic control among individuals with type 2 diabetes.

Methods: In a single-center, randomized controlled trial, 42 participants with type 2 diabetes were randomly assigned to either the intervention or the control group. The intervention consisted of a 12-week laughter yoga program. Hemoglobin A1c (HbA1c), body weight, waist circumference, psychological factors, and sleep duration were evaluated at baseline and week 12.

Results: Intention-to-treat analysis showed that participants in the laughter yoga group experienced significant improvements in HbA1c levels (between-group difference: −0.31%; 95% CI −0.54, −0.09) and positive affect scores (between-group difference: 0.62 points; 95% CI 0.003, 1.23). Sleep duration tended to increase in the laughter yoga group with a between-group difference of 0.4 hours (95% CI −0.05, 0.86; P = 0.080). The mean attendance rate for laughter yoga program was high (92.9%).

Conclusions: A 12-week laughter yoga program is feasible for individuals with type 2 diabetes and improves glycemic control. These findings suggest that having fun could be a self-care intervention. Further studies with larger numbers of participants are warranted to better evaluate the effects of laughter yoga.

Clinical trial registration: http://www.chinadrugtrials.org.cn, identifier UMIN000047164.

KEYWORDS

type 2 diabetes, laughter, laughter yoga, glycemic control, self-care, positive affect

1 Introduction

Diabetes is a chronic condition that requires lifelong self-management for metabolic control, such as healthy eating, regular physical activity, and self-monitoring of the blood glucose levels, as well as taking medication as prescribed (1, 2). Previous studies have shown that good glycemic control maintenance is important for reducing the risk of diabetes-related complications and mortality (3–5). However, it has been reported that about 40-60% of individuals with type 2 diabetes have not achieved good glycemic control in all regions of the world (6). For example, about 60% of individuals with type 2 diabetes in Japan and about half of those in the US, have not achieved the HbA1c < 7% glycemic target (7, 8). Furthermore, a study has found that despite greater utilization of newly developed glucose-lowering medications, concurrent improvements in overall glycemic control were not shown (9). These findings suggest that existing therapeutic options may not be sufficient for maintaining good glycemic control and investigating new complementary therapeutic approaches would be needed.

Previous studies have also found that individuals with type 2 diabetes had poor adherence to exercise and a healthy diet (10, 11). It would be difficult to sustain lifestyle changes (12), and efforts toward lifestyle modifications have the potential to cause emotional distress. On the other hand, it has been suggested that positive affect experienced during health behaviors facilitates long-term adherence to the behaviors (13). Previous studies have shown that people are more likely to intend to engage in a health behavior and to actually engage in it, when the behavior is seen as enjoyable (14, 15). A systematic review has found that pleasant affect experienced during exercise predicts future physical activity (16). Thus, we expected that enjoyable activities, which boost positive psychological states, would be useful as self-care behavior in individuals with type 2 diabetes.

In this study, we focused on laughter as an enjoyable diabetes self-care intervention. Laughter has been demonstrated to have various health benefits, such as reducing stress (17), enhancing the activities of natural killer cells (17–19), suppressing allergic reactions (20), and inhibiting increases in postprandial blood glucose levels (21). Prospective cohort studies demonstrated that a low frequency of laughter is associated with increased risks of functional disabilities, all-cause mortality, and cardiovascular diseases (22, 23). Additionally, laughter interventions have a positive effect on depression and the well-being of older adults (24).

We previously reported that the combination of laughter and exercise decreased hemoglobin A1c (HbA1c) levels among older adults (25). In that study, we combined watching comedy programs with a general exercise program to increase the participants' enjoyment and enhance their motivation to attend the intervention. However, making all participants laugh by watching the same comedy programs is difficult. Different individuals have different preferences. In contrast, self-induced laughter, including laughter yoga, which combines simulated laughter with deep breathing, does not require humorous stimuli (26). Additionally, a study that examined the potential benefits of laughter-inducing therapies has suggested that simulated laughter not caused by humor or other stimuli is more effective than spontaneous laughter triggered by humorous stimuli (27). Therefore, we conducted a laughter yoga intervention in this study.

Laughter yoga is now used in many countries (28). Several studies have shown that it improves depressive symptoms (29), decrease the levels of stress hormones (30, 31), and improves heart rate variability (32). Additionally, a study has reported that laughter yoga inhibits increases in postprandial blood glucose levels in individuals with type 2 diabetes (33). However, that study has shown the effect of a single 30-min session of laughter yoga only, and to the best of our knowledge, little is known about the long-term effects of laughter yoga on diabetes.

Therefore, this 12-week randomized controlled trial investigated the effects of laughter yoga on glycemic control and psychological well-being in individuals with type 2 diabetes.

2 Materials and methods

2.1 Study design and participants

This study was a single-center, two-group, randomized controlled trial designed to evaluate the effects of laughter yoga on individuals with type 2 diabetes. The intervention was conducted at Osaka University from October 2015 to December 2015. The study protocol was approved by the Scientific Ethics Committee of Fukushima Medical University (no. 2028). The Declaration of Helsinki was followed, and reporting in this article is aligned with the Consolidated Standards of Reporting Trials standards. All participants provided oral and written informed consent. This trial was registered at the University Hospital Medical Information Network Clinical Trials Registry (no. UMIN000047164).

Participants were recruited at the Diabetes Center of Osaka University Hospital, between August 2015 and September 2015. Three diabetologists evaluated their outpatients for study eligibility and approached patients meeting the eligibility criteria. Eligible patients received a flyer and the study was explained. Patients were recruited in order of their visit until the number of participants reached the target sample size. The inclusion criteria were outpatients aged 40 years or older with type 2 diabetes, HbA1c levels ranging from 6.1% to 7.9%, and changes in HbA1c levels < 1.0% during the last 3 months before baseline measurements. The glycemic control target is set at HbA1c <7.0% in Japan and modification of treatment (including intensive pharmacotherapy or insulin treatment) is needed when HbA1c level exceeded 8% (34). In addition, it would be difficult to determine whether the change in HbA1c level is due to the laughter yoga intervention or other factors in patients with originally large HbA1c fluctuations (unstable diabetes) before the laughter yoga intervention. Therefore, we included patients with HbA1c level changes of <1.0% during the last 3 months before baseline measurements (with stable treatment status). Patients with active coronary heart disease or stroke were excluded to reduce the risk of adverse events due to the intervention (e.g., a cardiac event during physical activity). Additionally, patients who could participate in light to moderate–intensity exercises were

included, and those with other vascular complications and severe illness were excluded.

Before randomization, the participants were informed that similar laughter yoga sessions would be held after the study period for those who were allocated to the control group to reduce the reporting bias.

2.2 Intervention

All participants continued to receive standard therapy for diabetes as they received before the study began. The control group continued the standard therapy and the intervention group received the standard therapy plus the laughter yoga program. Standard therapy included taking oral hypoglycemic medications, receiving advice from the doctor in charge of dietary modifications and physical activity in accordance with the "Treatment Guide for Diabetes" in Japan (35). Three participants received insulin therapy (one participant in the intervention group and two in the control group); the other participants received oral hypoglycemic agents. The control group was instructed to spend the study period as usual.

The participants in the intervention group received laughter yoga program once a week during the first 4 weeks and then every other week during the last 8 weeks. In total, eight sessions over 12 weeks were provided. The duration of laughter yoga intervention in previous studies ranged from 4 weeks to 8 weeks (28). However, a systematic review assessing the effects of yoga intervention on cardiovascular disease risk factors reported that the effects were most prominent in randomized controlled trials with 12 weeks of intervention duration (36). Therefore, we considered 12 weeks of intervention as appropriate. Every session began with a lecture of approximately 30 min on laughter and health, followed by a 60-min laughter yoga session. The laughter yoga session was based on the standardized laughter yoga program and the mini-lecture was added. The purpose of the lecture was to relax the participants and create a friendly atmosphere before the laughter yoga. Laughter yoga sessions were group-based interventions and guided by certified laughter yoga trainers from the Japan Laughter Yoga Association. All sessions were delivered by the same laughter yoga trainers to unify the contents of the intervention.

Laughter yoga is a kind of exercise consisting of deep breathing and voluntary laughter in a sitting or standing position. Each laughter yoga session consisted of warm up exercises, deep-breathing exercise, laughter exercise, and calming activity. At the beginning, the participants were asked to clap their hands along with saying the phrase "Ho, Ho, Ha, Ha, Ha," as a warm up exercises. Then, deep breathing with laughter were performed. Subsequently, the participants were asked to participate in voluntary laughing imaging in a variety of situations, including 5-min break. For example, when doing the "milkshake laughter," participants were asked to imagine that they have a glass of milk in their right hand and a glass of their favorite fruit juice in their left hand. They pretended to pour the milk from one glass into the other and pour it back into the first glass (to mix them). Then, they pretended to drink the milkshake, with a laugh. Finally, the participants were asked to close their eyes and relax.

2.3 Outcomes

The primary outcome was changes in HbA1c levels from the baseline to the 12-week follow-up measurement. Exploratory outcomes included changes in body weight, waist circumference, body mass index (BMI), positive affect, negative affect, subjective stress, and sleep duration from baseline to the 12-week follow-up. Physical examination and self-administered questionnaire were assessed on weeks 0 and 12.

HbA1c levels were measured in capillary whole blood, collected by finger prick, using the COBAS b101 point-of-care system (Roche Diagnostics International Ltd, Rotkreuz, Switzerland). Body weights were measured using a UC-322 weighing scale (A&D Co. Ltd., Tokyo, Japan), and BMIs were calculated as follows: weight (kg)/height squared (m^2). Waist circumference was measured halfway between the lower border of the ribs and the iliac crest using a measuring tape.

The following data were obtained *via* a self-administered questionnaire: height; lifestyle factors, such as smoking and drinking habits, sleep duration, and physical activity; and psychological factors. Positive and negative affects were assessed using the Japanese version of the 15-item Geriatric Depression Scale (GDS-15) (37). Most study participants were at least 65 years old. Thus, the GDS-15 was used for the assessment of psychological status. The GDS-15 is widely used to screen depression among older adults. Although the factor structure of the GDS-15 varies across study populations and language groups, a meta-analysis showed that the positive mood factor, including five items (Are you basically satisfied with your life? Are you in good spirits most of the time? Do you feel happy most of the time? Do you think it is wonderful to be alive now? and Do you feel full of energy)? is the most similar across languages (38). In this study, we used the 5 items as an indicator of positive affect and another 10 items as an indicator of negative affect. In a previous study, the Cronbach's alpha value was 0.72 for the 5 positive affect items and 0.82 for the 10 negative affect items in community-dwelling older adults in Japan (39). All GDS-15 items were assessed in a "yes/no" format. We calculated the sum of the presence of the positive affect and that of the negative affect (yes = 1, no = 0), respectively. In this study, positive (0–5) and negative (0–10) affect summary scores were created. Subjective stress was assessed using a single item ("Do you feel stressed at work or in daily life?") with four response options: very much = 4, quite a lot = 3, a little = 2, and not at all = 1. In addition, we took attendance in every session and calculated the attendance rate.

2.4 Sample size calculation

A previous study reporting that laughter can decrease HbA1c levels in older adults without diabetes (25) revealed a 0.19% difference in HbA1c changes between the intervention and control groups. A meta-analysis reported that low to moderate–intensity resistance exercises reduced HbA1c levels by 0.23% in individuals with type 2 diabetes (40). Therefore, we assumed a 0.2% difference in the mean decrease in HbA1c levels between the intervention and control groups (standard deviation of 0.2). A

sample size of 44 participants (22 in each group) was sufficient to detect the difference of 0.2% between groups using a two-tailed t-test of the difference between means with 90% power and a 5% significant level. The required sample size was 48, considering a dropout rate of 10%.

2.5 Randomization

After baseline measurements were completed, the participants were stratified according to sex and randomly allocated to either the intervention (laughter yoga program and standard therapy) or control (standard therapy only) group in a 1:1 ratio according to a computer-generated sequence. All randomization was carried out by a researcher who was not involved with participant enrollment. Participants were informed of their group assignment after consent and baseline measurements. Outcome assessors were blinded to group allocation.

2.6 Statistical analysis

Data were analyzed according to an intention to treat principle, with the baseline value carried forward for missing data. The differences in baseline characteristics between the two groups were tested using independent samples *t* tests for continuous variables and chi-square tests for categorical variables. Mann-Whitney U tests were used when the continuous variables were not normally distributed. The changes in measurements between the baseline and the 12-week follow-up in both groups were compared using paired-samples *t* tests. Unadjusted differences of changes from baseline to the 12-week follow-up between the two groups were analyzed using the independent samples *t* tests. Differences in changes between the two groups adjusting for age, BMI and each dependent variable value at baseline were analyzed using analysis of covariance. As a further check, per-protocol analyses excluding dropouts were conducted. Attendance rates for laughter yoga program were calculated by dividing the number of sessions attended by the number of sessions prescribed (41). All analyses were conducted using Statistical Package for the Social Sciences (IBM Corporation, Armonk, NY, USA). *P*-values of less than 0.05 were used to denote statistical significance.

3 Results

Forty-five eligible participants were enrolled in the study, and 42 agreed to participate in this study and underwent baseline measurements. Twenty-one participants were assigned to the laughter yoga group and 21 participants were assigned to the control group (Figure 1). One person in each group dropped out because of personal reasons. Both groups had high retention rates of 95%. The mean attendance rate for the laughter yoga program was 92.9%. No serious adverse events occurred. No changes in the content of the medical treatment occurred in either group during the study period.

The baseline characteristics of the 42 participants stratified by groups are shown in Table 1. No significant differences in baseline characteristics were observed between the two groups. The

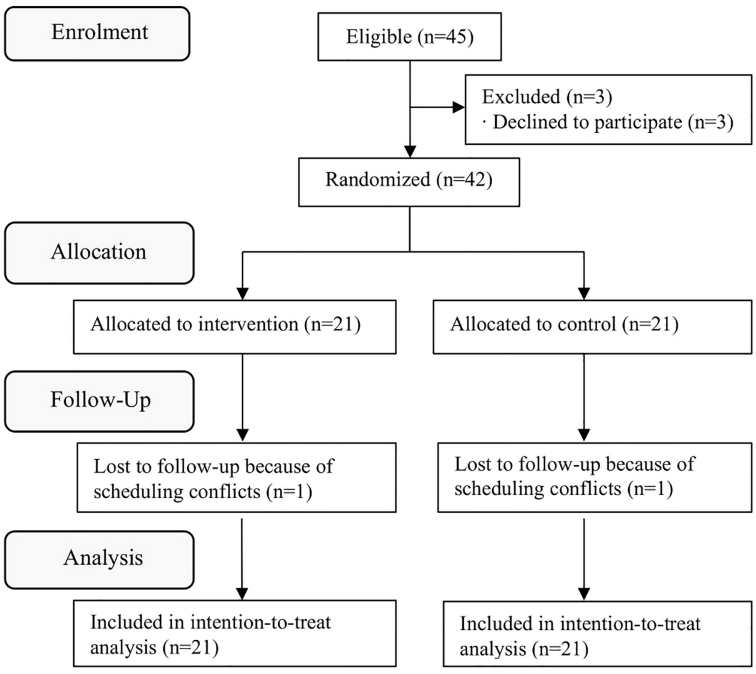

FIGURE 1
Consolidated Standards of Reporting Trials (CONSORT) diagram of study participants.

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TABLE 1 Baseline characteristics of the participants.

	Laughter yoga group (n = 21)	Control group (n = 21)	*p*-value
Age, years	71.8 (6.4)	70.6 (8.2)	0.641
Women, n (%)	14 (66.7)	14 (66.7)	1.000
Body weight, kg	58.1 (9.2)	62.2 (9.7)	0.165
BMI, kg/m^2	23.4 (3.2)	24.7 (2.8)	0.156
Waist circumference, cm	87.5 (9.3)	91.9 (8.3)	0.116
HbA1c, %	7.07 (0.7)	7.19 (0.7)	0.580
Positive affect	3.4 (1.6)	3.9 (1.3)	0.454
Negative affect	2.1 (1.6)	2.0 (1.7)	0.745
Subjective stress	1.9 (0.7)	2.2 (0.9)	0.320
Sleep duration, hours	6.0 (0.9)	6.2 (1.3)	0.542
Exercise, ≥2 days/week, n (%)	14 (66.7)	10 (47.6)	0.350
Current alcohol drinking, n (%)	3 (14.3)	8 (38.1)	0.160
Current smoking, n (%)	0 (0)	0 (0)	1.000
Number of family members	3.2 (1.6)	3.0 (1.4)	0.731

BMI, body mass index; HbA1c, hemoglobin A1c. Data are mean (SD) unless otherwise indicated.

Cronbach's alpha value was 0.70 for the positive affect items and 0.51 for the negative affect items.

Table 2 presents the changes from baseline to 12th week follow-up. The mean HbA1c levels changed from 7.07% to 6.82% in the laughter yoga group and from 7.19% to 7.26% in the control group. The unadjusted difference between the two groups was statistically significant (−0.31%; 95% confidence interval [CI]: −0.54% to −0.09%; P = 0.008). The adjusted difference (adjustment for age, BMI and HbA1c level at baseline) was also significant (P = 0.002). The positive affect score significantly increased in the laughter yoga group (between-group difference: 0.62 points; 95% CI 0.003 to 1.23; P = 0.049), although the adjusted between-group difference (adjustment for age, BMI and positive affect score at baseline) was not statistically significant (P = 0.139). A trend toward an increase in sleep duration was observed in the laughter yoga group with a between-group difference of 0.4 hours (unadjusted: 95% CI −0.05 to 0.86; P = 0.080), and the adjusted difference was marginally significant (P = 0.054). No significant differences in body weight, BMI, waist circumference, negative affect, and subjective stress were found between groups. Per-protocol analyses excluding dropouts and missing data in the follow-up questionnaire revealed a very similar pattern of results for all outcomes (Table 3).

At baseline and 12-week follow-up, 14 (66.7%) and 19 (90.5%) individuals in the laughter yoga group, and 10 (47.6%) and 8 (38.1%) individuals in the control group reported having exercise at least twice a week, respectively. The number of individuals with exercise habits increased in the laughter yoga group but with no statistical significance. However, the adjusted between-group difference of HbA1c remained statistically significant after adding the change in exercise habits as a covariate. The mean change in HbA1c was 0% in the five participants who reported increased exercise habits and −0.32% in the 16 participants without exercise habit changes. Additionally, the number of individuals who reported eating until full and skipping breakfast was not significantly different between the two groups at baseline and 12-week follow-up (data not shown). The number of individuals who reported eating until full and skipping breakfast remained from baseline to 12-week follow-up in both groups.

4 Discussion

The results of this study showed that laughter yoga program for 12 weeks decreased HbA1c levels in individuals with type 2 diabetes. Additionally, the high attendance rate suggests that the program is feasible for the participants. To the best of our knowledge, this is the first randomized controlled trial that has evaluated the long-term effects of laughter yoga on glycemic control in individuals with type 2 diabetes.

The findings of the current study are consistent with previous studies demonstrating that laughter by watching a comedy show for 40 min (21) or a single 30-min session of laughter yoga (33) inhibits the increase in postprandial glucose levels in individuals with type 2 diabetes. The findings are also consistent with our previous study showing that the combination of laughter by watching comedy shows and exercise for 10 weeks decreased HbA1c levels among older adults (25).

The mean HbA1c levels changed from 7.07% to 6.82% in the laughter yoga group. Recent study has shown that maintaining HbA1c levels at <7% over 5 years is associated with significant reductions in the odds of being diagnosed with diabetes-related complications (3). Another study found that the secondary

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TABLE 2 Physiological and psychological changes during the intervention in the laughter yoga and control groups.

	Baseline		12 weeks		Δ0-12 weeks	*p*-value[a]	Between-group difference (95% CI)	*p*-value[b]	*p*-value[c]
	Mean (SD)		Mean (SD)		(95% CI)				
HbA1c (%)									
Laughter yoga (n = 21)	7.07	(0.7)	6.82	(0.6)	−0.24 (−0.39, −0.09)	0.004	−0.31 (−0.54, −0.09)	0.008	0.002
Control (n = 21)	7.19	(0.7)	7.26	(0.7)	0.07 (−0.11, 0.24)	0.410			
Body weight (kg)									
Laughter yoga (n = 21)	58.1	(9.2)	58.2	(9.3)	0.15 (−0.23, 0.54)	0.421	−0.36 (−0.88, 0.16)	0.166	0.164
Control (n = 21)	62.2	(9.7)	62.7	(9.5)	0.51 (0.15, 0.88)	0.009			
BMI (kg/m^2)									
Laughter yoga (n = 21)	23.4	(3.2)	23.5	(3.2)	0.06 (−0.10, 0.22)	0.447	−0.16 (−0.38, 0.06)	0.146	0.134
Control (n = 21)	24.7	(2.8)	25.0	(2.8)	0.22 (0.06, 0.37)	0.010			
Waist circumference (cm)									
Laughter yoga (n = 21)	87.5	(9.3)	86.7	(8.8)	−0.82 (−2.17, 0.52)	0.217	−1.04 (−3.07, 0.97)	0.302	0.141
Control (n = 21)	91.9	(8.3)	92.1	(8.2)	0.22 (−1.37, 1.82)	0.773			
Positive affect									
Laughter yoga (n = 21)	3.4	(1.6)	4.1	(1.2)	0.62 (0.11, 1.13)	0.020	0.62 (0.003, 1.23)	0.049	0.139
Control (n = 21)	3.9	(1.3)	3.9	(1.0)	0.00 (−0.38, 0.38)	1.000			
Negative affect									
Laughter yoga (n = 21)	2.1	(1.6)	2.2	(1.8)	0.05 (−0.54, 0.63)	0.867	−0.05 (−0.74, 0.64)	0.890	0.786
Control (n = 21)	2.0	(1.7)	2.1	(1.6)	0.10 (−0.31, 0.50)	0.629			
Subjective stress									
Laughter yoga (n = 21)	1.9	(0.7)	2.1	(1.0)	0.14 (−0.16, 0.44)	0.329	0.34 (−0.11, 0.77)	0.133	0.339
Control (n = 21)	2.2	(0.9)	2.0	(0.6)	−0.19 (−0.53, 0.15)	0.258			
Sleep duration (hours)									
Laughter yoga (n = 21)	6.0	(0.9)	6.3	(1.0)	0.33 (−0.04, 0.70)	0.074	0.40 (−0.05, 0.86)	0.080	0.054
Control (n = 21)	6.2	(1.3)	6.1	(1.4)	−0.07 (−0.40, 0.22)	0.614			

BMI, body mass index; HbA1c, hemoglobin A1c; SD, standard deviation; 95% CI, 95% confidence interval.
[a]P for comparing the difference in outcomes before and after the intervention using paired-samples t test.
[b]P for unadjusted between-group difference in changes in outcomes over 12 weeks using independent samples t test.
[c]P for adjusted between-group difference (adjustment for age, BMI and each dependent variable value at baseline) using analysis of covariance.

structure of hemoglobin in individuals with good glycemic control (HbA1c < 7.0%) was not significantly altered although elevated HbA1c levels contribute to hemoglobin structural modifications, which are associated with pathological complications in type 2 diabetes mellitus (42). A previous meta-analysis reported that every 1% increase in HbA1c is associated with a 15% increase in the hazard of all-cause mortality, a 25% increase in cardiovascular disease mortality, a 17% increase in cardiovascular diseases, and an 11% increase in stroke, and suggested a positive dose-response trend between HbA1c levels and cardiovascular outcomes in people with type 2 diabetes (5). A recent study in the USA also demonstrated that a 1% reduction in HbA1c is associated with a 13% reduction in diabetes-related total healthcare costs, resulting in an annual cost savings of $736 (43). Thus, changes in HbA1c in this study might be clinically significant in patients with type 2 diabetes.

A meta-analysis found that, in individuals with type 2 diabetes, low-to-moderate-intensity resistance exercise reduced HbA1c levels by 0.23%, and high-intensity resistance exercise reduced HbA1c levels by 0.61% (40). The 0.24% reduction in HbA1c levels in this study is similar to the effects of low-to-moderate-intensity resistance exercise. Another meta-analysis reported a 0.33% reduction in HbA1c levels in a psychological treatment group compared with a control group, and greater improvements in participants with higher baseline HbA1c levels (44). Similarly, a systematic review of healthcare interventions reported a 0.34% reduction in HbA1c levels, and subgroup analysis showed that populations with baseline HbA1c levels > 9.5% exhibited more reduction in HbA1c (0.58%) than populations with baseline HbA1c levels < 9.5% (0.17%) (45). Considering that the average baseline HbA1c level in our study

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TABLE 3 Physiological and psychological changes during the intervention in the laughter yoga and control groups using per-protocol analyses.

	Baseline		12 weeks		Δ0-12 weeks	*p*-value[a]	Between-group difference (95% CI)	*p*-value[b]	*p*-value[c]
	Mean (SD)		Mean (SD)		(95% CI)				
HbA1c (%)									
Laughter yoga (n = 20)	7.12	(0.7)	6.86	(0.6)	−0.26 (−0.42, −0.09)	0.004	−0.33 (−0.57, −0.09)	0.008	0.003
Control (n = 20)	7.20	(0.7)	7.27	(0.7)	0.08 (−0.11, 0.26)	0.410			
Body weight (kg)									
Laughter yoga (n = 20)	58.3	(9.4)	58.5	(9.5)	0.16 (−0.25, 0.57)	0.422	−0.38 (−0.92, 0.16)	0.166	0.175
Control (n = 20)	62.5	(9.8)	63.0	(9.7)	0.54 (0.15, 0.93)	0.009			
BMI (kg/m^2)									
Laughter yoga (n = 20)	23.4	(3.3)	23.5	(3.3)	0.06 (−0.11, 0.23)	0.455	−0.17 (−0.39, 0.06)	0.145	0.143
Control (n = 20)	24.9	(2.7)	25.2	(2.7)	0.23 (0.06, 0.39)	0.009			
Waist circumference (cm)									
Laughter yoga (n = 20)	87.4	(9.5)	86.6	(9.0)	−0.87 (−2.28, 0.55)	0.217	−1.10 (−3.23, 1.03)	0.302	0.133
Control (n = 20)	92.5	(8.1)	92.7	(8.0)	0.24 (−1.45, 1.92)	0.774			
Positive affect									
Laughter yoga (n = 19)	3.3	(1.7)	4.0	(1.2)	0.68 (0.13, 1.24)	0.019	0.68 (0.01, 1.36)	0.048	0.246
Control (n = 19)	4.0	(1.3)	4.0	(1.0)	0.00 (−0.43, 0.43)	1.000			
Negative affect									
Laughter yoga (n = 19)	2.3	(1.6)	2.3	(1.8)	0.05 (−0.60, 0.71)	0.867	−0.05 (−0.82, 0.71)	0.890	0.935
Control (n = 19)	1.9	(1.7)	2.0	(1.7)	0.11 (−0.35, 0.56)	0.630			
Subjective stress									
Laughter yoga (n = 18)	1.8	(0.7)	2.0	(1.0)	0.17 (−0.19, 0.52)	0.331	0.40 (−0.13, 0.93)	0.132	0.415
Control (n = 17)	2.2	(0.9)	1.9	(0.6)	−0.24 (−0.66, 0.19)	0.260			
Sleep duration (hours)									
Laughter yoga (n = 18)	6.0	(1.0)	6.4	(1.0)	0.39 (−0.04, 0.82)	0.074	0.47 (−0.06, 1.00)	0.080	0.058
Control (n = 18)	6.3	(1.4)	6.2	(1.5)	−0.08 (−0.43, 0.26)	0.616			

BMI, body mass index; HbA1c, hemoglobin A1c; SD, standard deviation; 95% CI, 95% confidence interval.
[a]P for comparing the difference in outcomes before and after the intervention using paired-samples t test.
[b]P for unadjusted between-group difference in changes in outcomes over 12 weeks using independent samples t test.
[c]P for adjusted between-group difference (adjustment for age, BMI and each dependent variable value at baseline) using analysis of covariance.

was 7.1%, laughter yoga may be effective as an adjunctive therapy for the management of type 2 diabetes.

Although the mechanisms remain unclear, there are several possibilities for the effect of laughter yoga on diabetes. First, it has been reported that laughter upregulates genes related to natural killer cell activity in individuals with type 2 diabetes, which may ameliorate glucose intolerance (18). Second, laughter could influence glycemic control through the effects of positive affect that accompanies laughter. Positive psychological constructs such as positive affect, optimism and self-efficacy have been suggested to increase adherence to health behaviors (46), which may benefit individuals with type 2 diabetes. A longitudinal cohort study has also reported that positive affect such as enjoyment of life predicts lower risk of mortality in older adults with diabetes (47). In the present study, positive affect significantly increased from baseline to the 12-week follow-up in the laughter yoga group, which might be beneficial for glycemic control. In addition, laughter might have a stress-buffering effect. It has been reported that diabetes-related distress predicts poor glycemic control and poor medication adherence (48). Laughter yoga could attenuate cortisol stress response (30), which might buffer against the negative impact of stress. Third, increased energy expenditure during laughter might be beneficial for glycemic control. A study has suggested that 10–15 min of voiced laughter (by viewing a humorous film) could increase energy expenditure by 10–40 kcal (49). Laughter yoga may increase energy expenditure more than laughter during watching a humorous film, although no studies have examined energy expenditure during laughter yoga.

It has been reported that when people experience positive affect during a specific behavior, they are more likely to continue that

behavior (14). The positive affect experienced through laughter yoga may have led to the high attendance rate in the laughter yoga program. Although most health behaviors are difficult to sustain, continuing laughter yoga as a habit may be relatively easy.

In this study, sleep duration tended to improve. This finding is consistent with previous studies reporting that laughter has favorable effects on sleep quality or insomnia. One study has shown that 1 h of laughter therapy once a week for 4 weeks improves insomnia and sleep quality among older individuals (50). Another study has reported that a 30-min laughter yoga session twice weekly for 8 weeks improves sleep quality in patients undergoing hemodialysis (51). Additionally, a significant correlation was observed between changes in sleep duration and HbA1c levels in the laughter yoga group ($r = -0.47$; $P = 0.050$). It has been reported that short sleep durations (less than 4.5–6 h/night) are associated with increased HbA1c levels in individuals with type 2 diabetes (52). Thus, the increase in sleep duration in the laughter yoga group might be associated with better glycemic control.

Laughter yoga combines simulated laughter with yoga breathing techniques. The effects of laughter and the effects of yogic breathing are difficult to distinguish because laughter also consists of mixed patterns of expiration, inspiration, and interval pauses (53). It has been reported that voiced laughter causes a 10%–20% increase in energy expenditure and heart rate compared with resting values (49), and may activate sympathetic activity (54). In contrast, most yogic breathing practices result in a parasympathetic shift of autonomic nervous system activity (55). Laughter may have effects similar to exercise, and yogic breathing may enhance the effects of relaxation. However, this study did not measure the heart rate. Further studies are needed to assess the effects of laughter yoga on the autonomic nervous system.

This study conducted a 12-week laughter yoga intervention. In a systematic review of the effects of laughter-inducing interventions, the duration of most laughter yoga interventions ranged 4 to 8 weeks and one study conducted a 12-week laughter yoga intervention (28). Another systematic review of the effects of laughter yoga in older adults reported that the duration of interventions ranged from 4 to 6 weeks (26). To our best knowledge, little is known about the effects of longer-term laughter yoga interventions and it remains unclear which duration of intervention is most effective. It is possible that longer-term intervention is more effective. On the other hand, a systematic review on yoga for type 2 diabetes reported that the duration of yoga intervention ranged one week to 26 weeks (a median of 12 weeks) (56). Additionally, a meta-analysis assessing the effects of yoga intervention on cardiovascular disease risk factors reported that the effects were most prominent in randomized controlled trials with 12 weeks of intervention duration, and fewer effects were found in shorter or longer interventions (36). More studies with various duration of laughter yoga intervention including especially longer-term interventions are needed to examine the most effective intervention duration.

In this study, we did not assess the sustainability of the effects of the intervention after the study period. Further follow-up studies are needed to examine how long the effects of laughter yoga are maintained. A meta-analysis showed that improved physical activity through behavioral change interventions is generally not sustained after the intervention (57). Motivating participants to continue laughter yoga beyond intervention termination would be important (e.g., introducing laughter yoga class in the local community or recommending making a new laughter yoga group).

This study has several limitations. First, the number of participants was small, and the study was conducted in a single center. Multicenter studies with larger sample sizes are needed to better understand the intervention's efficacy and generalizability. The study participants were all Japanese living in an urban area and outpatients of the university hospital with relatively good glycemic control. Our findings may not apply to different populations, including other ethnic groups, those living in rural areas, and those with poorly controlled type 2 diabetes. Further studies are needed to evaluate the effects of laughter yoga on glycemic control in different populations. In contrast, the effects of laughter yoga on mental health have been reported worldwide (Asia, the Middle East, Australia, and United States) and in different clinical settings (28, 58). Therefore, this intervention could be applied to various populations. Second, changes in unknown psychosocial or lifestyle factors that were not assessed in this study might have affected glycemic outcomes. Additionally, group activity participation might have beneficial effects. Communicating with other participants and developing a sense of community could improve their motivation to attend the intervention. Third, diabetes-related distress was not assessed in this study. Further study is needed to assess distress using validated questionnaires. Fourth, positive and negative affect was measured using items from GDS-15. The Cronbach's Alpha value in this study was acceptable for the positive affect items, however the value was relatively low for the negative affect items. The Cronbach's Alpha value is influenced by the number of items, item inter-relatedness, and dimensionality, and a small number of items will underestimate the reliability (59). A study has suggested that a value of 0.50 is satisfactory when the items are fewer than 20 (60). However, using the ten items as one factor may not be suitable for this study population. Negative affect has various types. The factor structure of the GDS-15 has been reported to vary depending on the study population (38). For example, a study among community-dwelling older Japanese reported three factors: depressed mood, positive affect, and energy loss (61). Another study that included home-dwelling poststroke patients in Japan reported two factors: the positive affect and the depressed mood, including lack of energy (62). In addition, although the GDS-15 includes components of positive and negative affect, they might not represent pure positive and negative affect. Fifth, comorbid psychological conditions were not considered in the exclusion criteria. In terms of depression, no individuals received treatment for depression after confirming the medication status, and the baseline scores of the GDS-15 did not indicate depression in any participants. Sixth, sleep duration was self-reported, which might have affected the reliability of the data.

In conclusion, laughter yoga for 12 weeks decreased HbA1c levels in individuals with type 2 diabetes, and the program is feasible

with a high attendance rate. The importance of psychological well-being in individuals with diabetes has been gradually recognized, and positive psychological interventions have been recommended (63). We propose that having fun could be a self-care intervention. Although further studies with larger numbers of participants are warranted to better evaluate the beneficial effects, laughter yoga may be applied as an easy, enjoyable, and effective option for self-managing type 2 diabetes.

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 Scientific Ethics Committee of Fukushima Medical University. The patients/participants provided their written informed consent to participate in this study.

Author contributions

MH analyzed and interpreted data, and wrote the manuscript. MH, TO, and YW conducted the study and contributed to data acquisition. TO, EE, KS, HIm, NF, HN, NK, IS, and HIs contributed to the concept and design of the study and participated in critical revision of the manuscript. HN, NK and IS recruited participants and interpreted the diabetes-related outcome measures. MH and TO are the guarantors of this work and, as such, had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. All authors contributed to the article and approved the submitted version.

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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.

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References

1. Araki E, Goto A, Kondo T, Noda M, Noto H, Origasa H, et al. Japanese Clinical practice guideline for diabetes 2019. *J Diabetes Invest* (2020) 11(4):1020–76. doi: 10.1111/jdi.13306

2. DeFronzo RA, Ferrannini E, Groop L, Henry RR, Herman WH, Holst JJ, et al. Type 2 diabetes mellitus. *Nat Rev Dis Primers* (2015) 1:15019. doi: 10.1038/nrdp.2015.19

3. Boye KS, Thieu VT, Lage MJ, Miller H, Paczkowski R. The association between sustained HbA1c control and long-term complications among individuals with type 2 diabetes: A retrospective study. *Adv Ther* (2022) 39(5):2208–21. doi: 10.1007/s12325-022-02106-4

4. Gebregziabher M, Egede LE, Lynch CP, Echols C, Zhao Y. Effect of trajectories of glycemic control on mortality in type 2 diabetes: A semiparametric joint modeling approach. *Am J Epidemiol* (2010) 171(10):1090–8. doi: 10.1093/aje/kwq070

5. Zhang Y, Hu G, Yuan Z, Chen L. Glycosylated hemoglobin in relationship to cardiovascular outcomes and death in patients with type 2 diabetes: A systematic review and meta-analysis. *PloS One* (2012) 7(8):e42551. doi: 10.1371/journal.pone.0042551

6. Blonde L, Aschner P, Bailey C, Ji L, Leiter LA, Matthaei S, et al. Gaps and barriers in the control of blood glucose in people with type 2 diabetes. *Diabetes Vasc Dis Res* (2017) 14(3):172–83. doi: 10.1177/1479164116679775

7. Fang M, Wang D, Coresh J, Selvin E. Trends in diabetes treatment and control in U.S. adults, 1999-2018. *N Engl J Med* (2021) 384(23):2219–28. doi: 10.1056/NEJMsa2032271

8. Tajima N, Nishimura R, Izumi K, Hayashino Y, Origasa H, Noda M, et al. A large-scale, observational study to investigate the current status of diabetes complications and their prevention in Japan: Research outline and baseline data for type 2 diabetes-JDCP study 1. *Diabetol Int* (2015) 6:243–51. doi: 10.1007/s13340-015-0223-1

9. Lipska KJ, Yao X, Herrin J, McCoy RG, Ross JS, Steinman MA, et al. Trends in drug utilization, glycemic control, and rates of severe hypoglycemia, 2006-2013. *Diabetes Care* (2017) 40(4):468–75. doi: 10.2337/dc16-0985

10. Al-Salmi N CP, D'Souza MS. Diet adherence among adults with type 2 diabetes mellitus: A concept analysis. *Oman Med J* (2022) 37(2):e361. doi: 10.5001/omj.2021.69

11. da Rocha RB, Silva CS, Cardoso VS. Self-care in adults with type 2 diabetes mellitus: A systematic review. *Curr Diabetes Rev* (2020) 16(6):598–607. doi: 10.2174/1573399815666190702161849

12. Schmidt SK, Hemmestad L, MacDonald CS, Langberg H, Valentiner LS. Motivation and barriers to maintaining lifestyle changes in patients with type 2 diabetes after an intensive lifestyle intervention (The U-TURN trial): A longitudinal qualitative study. *Int J Environ Res Public Health* (2020) 17(20). doi: 10.3390/ijerph17207454

13. Van Cappellen P, Rice EL, Catalino LI, Fredrickson BL. Positive affective processes underlie positive health behaviour change. *Psychol Health* (2018) 33(1):77–97. doi: 10.1080/08870446.2017.1320798

14. Kiviniemi MT, Voss-Humke AM, Seifert AL. How do I feel about the behavior? the interplay of affective associations with behaviors and cognitive beliefs as influences on physical activity behavior. *Health psychology: Off J Division Health Psychology Am Psychol Assoc* (2007) 26(2):152–8. doi: 10.1037/0278-6133.26.2.152

15. Lawton R, Conner M, McEachan R. Desire or reason: Predicting health behaviors from affective and cognitive attitudes. *Health psychology: Off J Division Health Psychology Am Psychol Assoc* (2009) 28(1):56–65. doi: 10.1037/a0013424

16. Rhodes RE, Kates A. Can the affective response to exercise predict future motives and physical activity behavior? a systematic review of published evidence. *Ann Behav medicine: A Publ Soc Behav Med* (2015) 49(5):715–31. doi: 10.1007/s12160-015-9704-5

17. Bennett MP, Zeller JM, Rosenberg L, McCann J. The effect of mirthful laughter on stress and natural killer cell activity. *Altern therapies Health Med* (2003) 9(2):38–45.

18. Hayashi T, Tsujii S, Iburi T, Tamanaha T, Yamagami K, Ishibashi R, et al. Laughter up-regulates the genes related to NK cell activity in diabetes. *Biomed Res* (2007) 28(6):281–5. doi: 10.2220/biomedres.28.281

19. Takahashi K, Iwase M, Yamashita K, Tatsumoto Y, Ue H, Kuratsune H, et al. The elevation of natural killer cell activity induced by laughter in a crossover designed study. *Int J Mol Med* (2001) 8(6):645–50. doi: 10.3892/ijmm.8.6.645

20. Kimata H. Effect of humor on allergen-induced wheal reactions. *Jama* (2001) 285(6):738. doi: 10.1001/jama.285.6.738

21. Hayashi K, Hayashi T, Iwanaga S, Kawai K, Ishii H, Shoji S, et al. Laughter lowered the increase in postprandial blood glucose. *Diabetes Care* (2003) 26(5):1651–2. doi: 10.2337/diacare.26.5.1651

22. Sakurada K, Konta T, Watanabe M, Ishizawa K, Ueno Y, Yamashita H, et al. Associations of frequency of laughter with risk of all-cause mortality and cardiovascular disease incidence in a general population: Findings from the yamagata study. *J Epidemiol* (2020) 30(4):188–93. doi: 10.2188/jea.JE20180249

23. Tamada Y, Takeuchi K, Yamaguchi C, Saito M, Ohira T, Shirai K, et al. Does laughter predict onset of functional disability and mortality among older Japanese adults? the JAGES prospective cohort study. *J Epidemiol* (2021) 31(5):301–7. doi: 10.2188/jea.JE20200051

24. Gonot-Schoupinsky FN, Garip G. Laughter and humour interventions for well-being in older adults: A systematic review and intervention classification. *Complement Ther Med* (2018) 38:85–91. doi: 10.1016/j.ctim.2018.04.009

25. Hirosaki M, Ohira T, Kajiura M, Kiyama M, Kitamura A, Sato S, et al. Effects of a laughter and exercise program on physiological and psychological health among community-dwelling elderly in Japan: Randomized controlled trial. *Geriatr Gerontol Int* (2013) 13(1):152–60. doi: 10.1111/j.1447-0594.2012.00877.x

26. Kuru Alici N, Arikan Donmez A. A systematic review of the effect of laughter yoga on physical function and psychosocial outcomes in older adults. *Complement Ther Clin Pract* (2020) 41:101252. doi: 10.1016/j.ctcp.2020.101252

27. van der Wal CN, Kok RN. Laughter-inducing therapies: Systematic review and meta-analysis. *Soc Sci Med* (2019) 232:473–88. doi: 10.1016/j.socscimed.2019.02.018

28. Stiwi K, Rosendahl J. Efficacy of laughter-inducing interventions in patients with somatic or mental health problems: A systematic review and meta-analysis of randomized-controlled trials. *Complement Ther Clin Pract* (2022) 47:101552. doi: 10.1016/j.ctcp.2022.101552

29. Bressington D, Mui J, Yu C, Leung SF, Cheung K, Wu CST, et al. Feasibility of a group-based laughter yoga intervention as an adjunctive treatment for residual symptoms of depression, anxiety and stress in people with depression. *J Affect Disord* (2019) 248:42–51. doi: 10.1016/j.jad.2019.01.030

30. Meier M, Wirz L, Dickinson P, Pruessner JC. Laughter yoga reduces the cortisol response to acute stress in healthy individuals. *Stress* (2021) 24(1):44–52. doi: 10.1080/10253890.2020.1766018

31. Tanaka A, Tokuda N, Ichihara K. Psychological and physiological effects of laughter yoga sessions in Japan: A pilot study. *Nurs Health Sci* (2018) 20(3):304–12. doi: 10.1111/nhs.12562

32. Dolgoff-Kaspar R, Baldwin A, Johnson MS, Edling N, Sethi GK. Effect of laughter yoga on mood and heart rate variability in patients awaiting organ transplantation: A pilot study. *Altern Ther Health Med* (2012) 18(5):61–6.

33. Čokolič M, Herodež Š, Sternad S, Krebs S. The inhibitory effect of laughter yoga on the increase in postprandial blood glucose in type2 diabetic patients. *Diabetol Croat* (2013) 42(2):54–8.

34. Araki E, Haneda M, Kasuga M, Nishikawa T, Kondo T, Ueki K, et al. New glycemic targets for patients with diabetes from the Japan diabetes society. *Diabetol Int* (2016) 7(4):327–30. doi: 10.1007/s13340-016-0297-4

35. Japan Diabetes Society. *Treatment guide for diabetes 2014-2015*. Tokyo: Bunkodo (2014).

36. Cramer H, Lauche R, Haller H, Steckhan N, Michalsen A, Dobos G. Effects of yoga on cardiovascular disease risk factors: A systematic review and meta-analysis. *Int J Cardiol* (2014) 173(2):170–83. doi: 10.1016/j.ijcard.2014.02.017

37. Niino N, Imaizumi T, Kawakami N. A Japanese translation of the geriatric depression scale. *Clin Gerontologist* (1991) 10:85–7.

38. Kim G, DeCoster J, Huang CH, Bryant AN. A meta-analysis of the factor structure of the geriatric depression scale (GDS): The effects of language. *Int Psychogeriatr* (2013) 25(1):71–81. doi: 10.1017/S1041610212001421

39. Hirosaki M, Ishimoto Y, Kasahara Y, Konno A, Kimura Y, Fukutomi E, et al. Positive affect as a predictor of lower risk of functional decline in community-dwelling elderly in Japan. *Geriatr Gerontol Int* (2013) 13(4):1051–8. doi: 10.1111/ggi.12008

40. Liu Y, Ye W, Chen Q, Zhang Y, Kuo CH, Korivi M. Resistance exercise intensity is correlated with attenuation of HbA1c and insulin in patients with type 2 diabetes: A systematic review and meta-analysis. *Int J Environ Res Public Health* (2019) 16(1). doi: 10.3390/ijerph16010140

41. Witlox L, Velthuis MJ, Boer JH, Steins Bisschop CN, Wall EV, Meulen WJTMV, et al. Attendance and compliance with an exercise program during localized breast cancer treatment in a randomized controlled trial: The PACT study. *PloS One* (2019) 14(5):e0215517. doi: 10.1371/journal.pone.0215517

42. Ye S, Ruan P, Yong J, Shen H, Liao Z, Dong X. The impact of the HbA1c level of type 2 diabetics on the structure of haemoglobin. *Sci Rep* (2016) 6:33352. doi: 10.1038/srep33352

43. Lage MJ, Boye KS. The relationship between HbA1c reduction and healthcare costs among patients with type 2 diabetes: Evidence from a U.S. claims database. *Curr Med Res Opin* (2020) 36(9):1441–7. doi: 10.1080/03007995.2020.1787971

44. Winkley K, Upsher R, Stahl D, Pollard D, Kasera A, Brennan A, et al. Psychological interventions to improve self-management of type 1 and type 2 diabetes: A systematic review. *Health Technol Assess.* (2020) 24(28):1–232. doi: 10.3310/hta24280

45. Murphy ME, Byrne M, Galvin R, Boland F, Fahey T, Smith SM. Improving risk factor management for patients with poorly controlled type 2 diabetes: A systematic review of healthcare interventions in primary care and community settings. *BMJ Open* (2017) 7(8):e015135. doi: 10.1136/bmjopen-2016-015135

46. Massey CN, Feig EH, Duque-Serrano L, Huffman JC. Psychological well-being and type 2 diabetes. *Curr Res Diabetes Obes J* (2017) 4(4). doi: 10.19080/crdoj.2017.04.555641

47. Moskowitz JT, Epel ES, Acree M. Positive affect uniquely predicts lower risk of mortality in people with diabetes. *Health psychology: Off J Division Health Psychology Am psychol Assoc* (2008) 27(1S):S73–82. doi: 10.1037/0278-6133.27.1.S73

48. Aikens JE. Prospective associations between emotional distress and poor outcomes in type 2 diabetes. *Diabetes Care* (2012) 35(12):2472–8. doi: 10.2337/dc12-0181

49. Buchowski MS, Majchrzak KM, Blomquist K, Chen KY, Byrne DW, Bachorowski JA. Energy expenditure of genuine laughter. *Int J Obes* (2007) 31(1):131–7. doi: 10.1038/sj.ijo.0803353

50. Ko HJ, Youn CH. Effects of laughter therapy on depression, cognition and sleep among the community-dwelling elderly. *Geriatr Gerontol Int* (2011) 11(3):267–74. doi: 10.1111/j.1447-0594.2010.00680.x

51. Ozer Z, Ates S. Effects of laughter yoga on hemodialysis patients' plasma-beta endorphin levels, pain levels and sleep quality: A randomized controlled trial. *Complementary Ther Clin Pract* (2021) 43:101382. doi: 10.1016/j.ctcp.2021.101382

52. Lee SWH, Ng KY, Chin WK. The impact of sleep amount and sleep quality on glycemic control in type 2 diabetes: A systematic review and meta-analysis. *Sleep Med Rev* (2017) 31:91–101. doi: 10.1016/j.smrv.2016.02.001

53. Miller M, Fry WF. The effect of mirthful laughter on the human cardiovascular system. *Med Hypotheses* (2009) 73(5):636–9. doi: 10.1016/j.mehy.2009.02.044

54. Sakuragi S, Sugiyama Y, Takeuchi K. Effects of laughing and weeping on mood and heart rate variability. *J Physiol Anthropol Appl Hum Sci* (2002) 21(3):159–65. doi: 10.2114/jpa.21.159

55. Saoji AA, Raghavendra BR, Manjunath NK. Effects of yogic breath regulation: A narrative review of scientific evidence. *J Ayurveda Integr Med* (2019) 10(1):50–8. doi: 10.1016/j.jaim.2017.07.008

56. Thind H, Lantini R, Balletto BL, Donahue ML, Salmoirago-Blotcher E, Bock BC, et al. The effects of yoga among adults with type 2 diabetes: A systematic review and meta-analysis. *Prev Med* (2017) 105:116–26. doi: 10.1016/j.ypmed.2017.08.017

57. McEwan D, Rhodes RE, Beauchamp MR. What happens when the party is over?: Sustaining physical activity behaviors after intervention cessation. *Behav Med* (2022) 48(1):1–9. doi: 10.1080/08964289.2020.1750335

58. Bressington D, Yu C, Wong W, Ng TC, Chien WT. The effects of group-based laughter yoga interventions on mental health in adults: A systematic review. *J Psychiatr Ment Health Nurs* (2018) 25(8):517–27. doi: 10.1111/jpm.12491

59. Tavakol M, Dennick R. Making sense of cronbach's alpha. *Int J Med Educ* (2011) 2:53–5. doi: 10.5116/ijme.4dfb.8dfd

60. Dall'Oglio AM, Rossiello B, Coletti MF, Caselli MC, Rava L, di Ciommo V, et al. Developmental evaluation at age 4: Validity of an Italian parental questionnaire. *J Paediatr Child Health* (2010) 46(7-8):419–26. doi: 10.1111/j.1440-1754.2010.01748.x

61. Yatomi N. The factor structure and item characteristics of the GDS (Geriatric depression scale) short version in a Japanese elderly sample. *Jpn J Geriatr* (1994) 16:29–36.

62. Schreiner AS, Morimoto T, Asano H. Depressive symptoms among poststroke patients in Japan: Frequency distribution and factor structure of the GDS. *Int J Geriatr Psychiatry* (2001) 16(10):941–9. doi: 10.1002/gps.444

63. Huffman JC, DuBois CM, Millstein RA, Celano CM, Wexler D. Positive psychological interventions for patients with type 2 diabetes: Rationale, theoretical model, and intervention development. *J Diabetes Res* (2015) 2015:428349. doi: 10.1155/2015/428349

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Missed meal boluses and poorer glycemic control impact on neurocognitive function may be associated with white matter integrity in adolescents with type 1 diabetes

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Background: The notion that pediatric type 1 diabetes impacts brain function and structure early in life is of great concern. Neurological manifestations, including neurocognitive and behavioral symptoms, may be present from childhood, initially mild and undetectable in daily life. Despite intensive management and technological therapeutic interventions, most pediatric patients do not achieve glycemic control targets for HbA1c. One of the most common causes of such poor control and frequent transient hyperglycemic episodes may be lifestyle factors, including missed meal boluses.

Objective: The aim of this study was to assess the association between specific neurocognitive accomplishments—learning and memory, inhibition ability learning, and verbal and semantic memory—during meals with and without bolusing, correlated to diffusion tensor imaging measurements of major related tracts, and glycemic control in adolescents with type 1 diabetes compared with their healthy siblings of similar age.

Study design and methods: This is a case–control study of 12- to 18-year-old patients with type 1 diabetes (N = 17, 8 male patients, diabetes duration of 6.53 ± 4.1 years) and their healthy siblings (N = 13). All were hospitalized for 30 h for continuous glucose monitoring and repeated neurocognitive tests as a function of a missed or appropriate pre-meal bolus. This situation was mimicked by controlled, patient blinded manipulation of lunch pre-meal bolus administration to enable capillary glucose level of <180 mg/dl and to >240 mg/d 2 hours after similar meals, at a similar time. The diabetes team randomly and blindly manipulated post-lunch glucose levels by subcutaneous injection of either

rapid-acting insulin or 0.9% NaCl solution before lunch. A specific neurocognitive test battery was performed twice, after each manipulation, and its results were compared, along with additional neurocognitive tasks administered during hospitalization without insulin manipulation. Participants underwent brain imaging, including diffusion tensor imaging and tractography.

Results: A significant association was demonstrated between glycemic control and performance in the domains of executive functions, inhibition ability, learning and verbal memory, and semantic memory. Inhibition ability was specifically related to food management. Poorer glycemic control (>8.3%) was associated with a slower reaction time.

Conclusion: These findings highlight the potential impairment of brain networks responsible for learning, memory, and controlled reactivity to food in adolescents with type 1 diabetes whose glycemic control is poor.

KEYWORDS

type 1 diabetes, glycemic control, adolescents, brain domains, diffusion tension imaging, cognitive performance

Introduction

The notion that diabetes mellitus (DM), one of the most prevalent chronic conditions in youngsters, impacts brain function and structure is far from new (1, 2). The theory was first proposed in 1922 (3) and has intrigued many investigators since then, especially regarding its effect on the quality of life. Type 1 diabetes (T1D) remains incurable, the outcome of an autoimmune assault on the insulin-producing pancreatic beta cells in genetically susceptible children (4, 5). Despite intensive management and technological interventions in therapy, most pediatric T1D patients fail to achieve glycemic control goals (6, 7), mainly due to inaccurate, late, or a lack of meal boluses. This may lead to a poorer prognosis for long-term diabetic complications (8). Neurological manifestations, including neurocognitive and behavioral complications, may appear soon after disease onset during childhood and adolescence (9). Studies show that brain volume alterations are detectable in childhood and have long-term influences on adulthood (10, 11).

Although the association between T1D and neurocognitive impairment is well known, the debate today focuses on which abilities are affected, their onset according to disease acquisition, and the underlying mechanisms. The brains of children and adolescents undergo constant change as they re-modulate into adulthood. In line with these changes, personality and abilities are formed in parallel with the continuously redesigned micro and macro-structure of the brain. It is thus critical to understanding the full impact of T1D on the brain, in addition to the impact of glycemic control and, in particular, of glycemic excursions (12), especially in children and adolescents. Glucose is the main brain fuel. Its uptake in the brains of young children reaches adult levels at the age of 2 and is almost double that by 5 years of age, falling back to adult levels at approximately 10 years of age (13). Approximately 25% of total adult glucose consumption is used for brain metabolism (14). These figures suggest that the brain may be vulnerable to glycemic extremes, especially during the first two decades of development (13). Missed meal boluses are known to be both frequent and devastating in the long term in adolescents with T1D and poor glycemic control. Furthermore, a significant association has been demonstrated between frequently missed boluses in pediatric T1D patients with poor glycemic control, and complications (8, 15).

A study that combines the parameters of acute glucose excursions and chronic glycemic control with neurocognitive assessment is thus required. As reported in the review we performed when preparing the study protocol (1), more than 40 years of diabetes research have demonstrated brain alterations and an increased risk of cognitive decline in T1D (16). These findings can now be corroborated by exploring changes in neural network activity using methods such as diffusion tensor imaging (DTI). Conventional MR techniques, such as T1- and T2-based measurements, cannot provide detailed information about the integrity and location of white matter (WM) tracts. DTI provides unique biologically and clinically relevant information for the study of diabetes-related alterations in the integrity of neuronal pathways (17–24). Based on MR measurement of the speed of water diffusion in tissues, it enables the characterization of tissue composition, physical properties, and architectural organization (25). With tractography, WM pathways can be traced *in vivo*, permitting the study of the nature of damage to WM tracts.

The overarching hypothesis of the association between acute transient and long-term hyperglycemia, explored in the research described here, is an anticipated relationship between neurocognitive performance in adolescent patients with T1D, as influenced by diabetes glycemic control, and its association with the quantitative parameters of WM in specific major pathways.

Materials and methods

Study design

This report includes findings from a wider, case-control proof-of-concept study conducted at the Pediatric Neurology and Epilepsy Department Research Unit (PNRU) at the Shamir (Assaf Harofeh) Medical Centre (SHMC), Be'er Ya'akov, in collaboration with SHMC's Neuroradiology Institute and Pediatric Endocrinology and Diabetes Institute, the Hadassah Medical Center's Neurology Department, and Bar Ilan University's Gonda Multidisciplinary Brain Research Center. The study observed the Helsinki Declaration's ethical principles for biomedical research involving human patients, together with local and national regulations. Prior to enrollment, all participating institutions obtained approval from their ethics committees and Israel's Health Ministry's Helsinki Committee. The study, with its full protocol, is listed at www.clinicaltrials.gov (NCT02923323).

Study population

The study population comprised 12- to 18-year-old T1D patients who were being cared for at SHMC's Pediatric Endocrinology and Diabetes Mellitus Institute, along with their healthy siblings of similar age. Healthy siblings, sharing close genetic profiles and similar environments, were a natural control group. Inclusion criteria for the T1D group were a T1D diagnosis according to ADA criteria (26) and a basal-bolus regimen for more than 2 years. Exclusion criteria were more than one severe hypoglycemic event or more than one episode of diabetic ketoacidosis (DKA), other than at diagnosis. The exclusion criteria for all participants comprised a history of head injury, epileptic episodes, psychiatric medications, renal or liver function abnormalities, and language limitation. The study population was divided into three groups: healthy control siblings, T1D patients with good glycemic control, and T1D patients with poor glycemic control. Glycemic control clusters were defined as glycated hemoglobin (HbA1c) above 8.3% as poor control, and HbA1c ≤ 8.3% as good control according to the EXCHANGE study results, with a mean teenager HbA1c of 8.26% in a large population (27).

Out of the 31 adolescents recruited, 8 were in the better glycemic control group, 9 were in the poorer glycemic control group, and 13 healthy siblings comprised the control group. One T1D participant from the better-controlled group was excluded due to incidental abnormal MRI scans. Five participants (two with T1D and three controls) did not undergo MRI for technical reasons.

Study setup: Three sessions performed in 1–4 weeks

Session 1 comprised signing informed consent by parents and participants and obtaining medical histories, physical assessments, baseline cognitive and lingual readiness by parents and participants, and cognitive assessments of participants.

Session 2 was a 30-h stay at the PNRU for neurocognitive assessments while monitoring food intake, glucose levels, and insulin administration. A specific neurocognitive test battery was performed twice, each time 2 h after lunch—at glucose >240 mg/dl and glucose ≤180 mg/dl. The glucose level was randomly and blindly manipulated before lunchtime tasks: rapid-acting insulin was administered before lunch on one day, and an injection of 0.9% NaCl solution was given before a lunch bolus on another day. Additional tasks were performed without insulin manipulation during hospitalization.

Session 3 comprised brain imaging with a prior capillary glucose measurement to verify levels of 70–240 mg/dl.

Measurements

Clinical data were retrieved from medical files; they included demographic information :age, gender, and socioeconomic status by home address. The SEP (socioeconomic position) based on home address was analyzed according to the Israel Central Bureau of Statistics Characterization and Classification of Statistical Areas within Municipalities and Local Councils by Socio-Economic Level of the Population, 2015. The SEP index classifies neighborhoods and localities into clusters, with 1 being the lowest rating and 10 being the highest. It is an adjusted calculation of 14 variables that measure social and economic level in four domains—demographics, education, standard of living, and employment. (28), clinical data :diabetes duration, annual HbA1c based on 3 last annual measurements, and complications. Physical examination elicited weight, height, body mass index (BMI), (SDS were calculated by CDC 2000 growth charts) (29), and Tanner staging (30). Glycemic control was defined according to HbA1c (26).

Glucose level measurements

ISG was assessed using a blinded CGMS (Minimed Inc., Sylmar, CA). Capillary glucose was measured regularly before meals and 2 hours after, and prior to neurocognitive testing. Patients and the neurocognitive team were blinded to glucose levels measured prior to neurocognitive tests.

Neurocognitive and psychosocial measurements

Neurocognitive data included a designed battery of tasks specifically modified for food intake. This report refers to the following tasks:

1. The Word Selective Reminding Test (%) subset 3 of the Test of Memory and Learning (TOMAL-2) (31) measures the ability of learning and immediate verbal recall. The examiner reads off a list of words to the participant, who is encouraged to recall as many of them as possible, regardless of the order of recall. After each trial, the examiner reminds the participant of the forgotten words and reads out the word list again. The subtest ends when the participant

remembers every word on two consecutive trials, or after eight trials, regardless of memory proficiency (32).

2. The Day and Night Task—Emotional Stroop for Eating Disorders (EST-ED), which is specifically designed, modified, and computerized to evaluate response inhibition to emotional food-related stimuli. It had two general shapes, each presented in three versions—one signifying an edible item and two as controls. Six stimuli were presented: nonfood pictures (for example, the moon and sun), food pictures (for example, a sweet item, a banana, and a low-sugar item, such as an egg), and two natural pictures (for example, an umbrella and a flower) (Figure 1). All items were shown at random with neutral emotion. Participants were instructed to push one button in reaction to stimulations similar to the moon and another in reaction to those similar to the sun (33, 34). The task was specifically modified for the study. Dependent measures analyzed for the study's purposes were accuracy and response time in emotional Stroop response (sun–moon), emotional nonfood Stroop (umbrella–flower), and emotional food Stroop (banana–egg).
3. The Visual Update Task is a spatial–figural updating task that evaluates the executive function of updating and the monitoring of working memory representations often associated with the prefrontal cortex dorsolateral section. To succeed, participants must monitor and code relevant incoming information and correctly adjust items held in their working memory by replacing old information with that which is newer and more pertinent (35). Our modified computerized task consisted of differently colored house shapes presented in different positions within a flower-shaped frame. Depending on the trial's load level, two to five different colors were used. The colored houses were presented one at a time, with participants asked to keep track of the last position of each color. At recall, the differently colored houses that had been shown were presented again, one at a time within the frame. Participants responded by clicking the mouse in the area of the frame where the color had appeared (Figure 2).
4. The Object Recall Task is a computerized semantic memory task in which new objects with different casings are presented on two possible backgrounds: blue (calming arousal) and yellow (exciting arousal). The task, described in an unpublished thesis by Tamar Schwarz of Bar Ilan's Department of Psychology, is based on an fMRI paradigm of semantic object representation (36) that was modified for our study. Initially, a series of objects shown against a colored background were presented to the participant, who was asked to remember as many as possible (Figure 3). The objects were then presented again, in succession, without backgrounds, and the participant was required to indicate whether they had been previously presented and whether against a yellow or blue background. Semantic memory of visually presented recall objects involves the thalamus, pre-supplementary motor area, and several other somatic cortical regions (37). We analyzed responses for accuracy and response time.

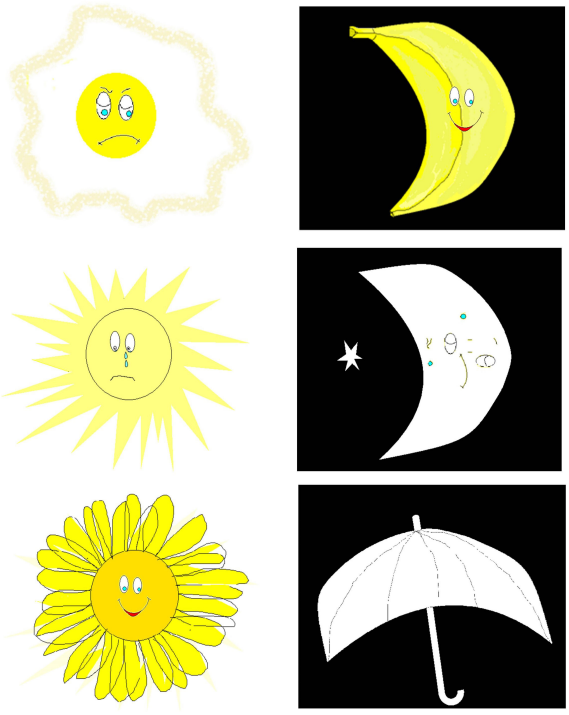

FIGURE 1
Day–night emotional Stroop stimuli.

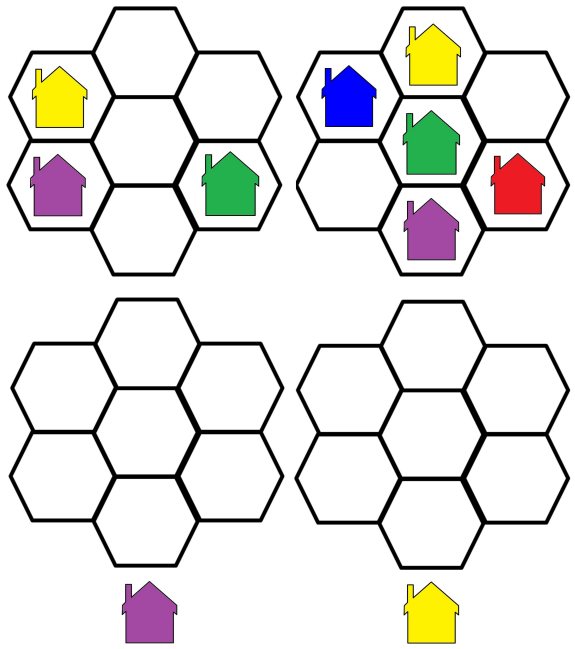

FIGURE 2
Visual Update Stimulus example.

Data were anonymized and coded by the neurocognitive team prior to analysis, according to clinical and glycemic measures.

Brain imaging measurements

Magnetic resonance imaging (MRI) scans were performed on a Siemens Medical Solutions 3.0-Tesla MRI scanner at SHMC. The T1 scans were used as an anatomical reference, while DTI was used to map neuronal tracts and evaluate brain WM properties. This was performed without sedation or contrast material.

MR data acquisition

T1 data were collected at a spatial resolution of 0.8 × 0.8 mm voxels, with 0.9-mm-thick axial slices covering the entire brain with no gaps. Repetition Time/Echo Time (TR/TE) = 2,000/2.41 ms, field of view (FOV) 137 = 245 mm, matrix size = 287 × 287. Scanning time was approximately 5 min.

DTI data acquisition protocol was as follows: *b*-value = 1,000 s/mm^2, along 30 non-collinear gradient directions (plus one b0 image), TR/TE = 15,000/91 ms, matrix size = 113 × 113, and a flip angle of 90°. Spatial resolution was 1.5 × 1.5 mm voxels, with 1.5-mm-thick axial slices with no gaps covering the entire cortex. Scanning time was about 9 min. Data were analyzed using MATLAB and C++-based software tools—SPM software (version 12, UCL, Queen Square Institute of Neurology, London, UK) and mrVista packages (http://white.stanford.edu/newlm/index.php/Software). This included correction for head movement and image artifacts, and the normalization and creation of a reference volume using a T1-weighted, AC-PC-aligned image. T1-weighted images were used for grey matter (GM) and WM volume assessment.

DTI data preprocessing

Using the mrDiffusion package from VISTA, the DTI data preprocessing pipeline had three additional steps:

1. Correct DTI data for eddy current and movement noise and align these to the anatomical reference.

FIGURE 3
Object Recall examples.

2. For each voxel in the scanned volume, fit a tensor model based on a Gaussian diffusion signal decay model and linear least-squares fits. Then, extract the three eigenvalues (Λ_1, Λ_2, and Λ_3) by tensor diagonalization and calculate the FA (an index that reflects the orientation of diffusion—mainly the uniform directionality of the tract, and it is high along well-defined pathways), AD (the rate of diffusion in the principal diffusion direction of the voxel), and RD (the rate of diffusion perpendicular to the principal diffusion direction of the voxel) according to the following equations:

Fractional anisotropy

$$= \sqrt{\frac{3}{2}} \frac{\sqrt{(\lambda 1 - \bar{\lambda})^2 + (\lambda 2 - \bar{\lambda})^2 + (\lambda 3 - \bar{\lambda})^2}}{\sqrt{\lambda 1^2 + \lambda 2^2 + \lambda 3^2}} \quad (1)$$

$$\text{Axial diffusivity} = \lambda_1 \quad (2)$$

$$\text{Radial diffusivity} = (\lambda_2 + \lambda_3)/2 \quad (3)$$

3. From the tensors created in the preprocessing procedure, create DTI maps (FA, RD, and AD) for each participant.

Fiber tracking and quantification

Automated fiber quantification (AFQ) (open source and freely available at https://github.com/yeatmanlab/AFQ) was used for the automated identification and quantification of cerebral WM pathways. This software package uses mrDiffusion functions and specially built functions and scripts executed with MATLAB. As first published in 2012, the AFQ automated fiber tract segmentation has proven to be equivalent to the time-consuming manual techniques that served as the gold standard (38). It is now widely used in clinical trials but has a particular advantage in this study as it was demonstrated in healthy children and adolescents.

First, tracing is initialized from the hemisphere mask: eight seed points are placed at equidistant locations in all voxels with an FA value greater than 0.3. Fiber tracts are estimated using a deterministic streamlined tracking algorithm (39) with a fourth-order Runge–Kutta path integration method. For tracking purposes, a continuous tensor field is estimated using trilinear interpolation of the tensor elements. Paths were tracked with a 1-mm step size; the stopping criterion was FA< 0.2 or tracking angle > 30°. The methodology and algorithms for the automated segmentation, tract cleaning, and tract quantification procedures are described elsewhere (38). AFQ was used to trace 20 tracts, including corpus callosum segments, corticospinal tract, thalamic radiation, cingulum cingulate, cingulum hippocampus, superior longitudinal fasciculus, inferior longitudinal fasciculus, inferior fronto-occipital fasciculus, uncinate, and the arcuate, and to segment the corpus callosum into eight cortical segments. Each of the 28 tracts was resampled to 100 equally spaced nodes, and diffusion properties were calculated for each node of each fiber (Figure 4). The mean and standard deviation were calculated for each diffusion property at each node of each tract for the healthy participants. A confidence interval was generated for each tract to quantify the similarity of each patient to the standard tract profile of the healthy group.

Callosal segmentation

The callosal segmentation of regions of interest (ROIs) deep in the WM was based on Huang (40) and Dougherty (41). This protocol was extended in a previous study by dividing the superior frontal segment into two parts, using the medial branching of the precentral sulcus as an anatomical landmark. Based on the cortical destinations of callosal fiber tracts, the corpus callosum was divided into eight segments: orbital, anterior frontal, superior frontal, motor (posterior frontal), superior parietal, posterior parietal, temporal, and occipital. A dual analysis using both ROI-based segmentation and selecting fiber segments was performed with the AFQ software. The original AFQ package was created for the segmentation of 20 major tracts, as defined by Mori et al. (42), with additional WM tracking functions added over the years. Bar Ilan's Neurolinguistics Lab in the Gonda Multidisciplinary Brain Research Center and Stanford's Wandell Lab in the Center for Cognitive and Neurobiological Imaging collaborated to add special functions for callosal segmentation to the AFQ software, based on the segmentation described previously. This automated callosal segmentation was validated in the Gonda Neurolinguistics Lab by comparing it with manual segmentation using Quench software (Vista Lab), by visual inspection of the created tracts, and by statistical comparison of the FA extracted using both methods. Callosal segments were clipped 5 mm to each side of the mid-sagittal plane to extract the quantitative properties of the callosal segments of the corpus callosum itself, uninfluenced by distant location. Thirty nodes (representing locations) were defined on every clipped segment to calculate the tract profile. The mean diffusivity parameters for all callosal segments were calculated for each patient with T1D and each healthy participant. FA, AD, and RD were further examined for each segment.

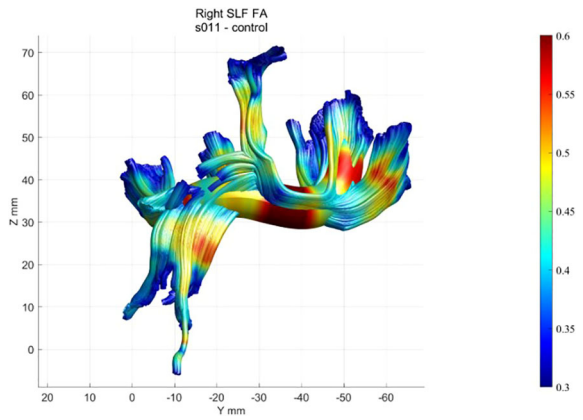

FIGURE 4
Example of a healthy participant's FA profile of the right SLF.

Outcome measures

The two primary outcome measures were: (a) difference in neurocognitive assessment scores between groups, according to glycemic control, and (b) difference in DTI parameters of AD, RD, and FA and corpus callosum (CC) segments between T1D and healthy groups, according to glycemic control. A secondary outcome measure was the correlation between associations with specific MRI tract alterations and neurocognitive performance scores.

Statistical analysis

All analyses were performed with MATLAB 2020a (© 1994-2020, The MathWorks, Inc.), with the assistance and guidance of a trained statistician.

Quantitative/numerical measures are presented as means ± SDs and min–max.

Qualitative/categorical measures are presented as percentages.

Participants' tasks with artifacts or missing data were excluded. After the performance was corrected and normalized, we ensured that no confounding variables, such as age or gender, influenced group differences. Correlations between cognitive performance, neurobehavioral outcome measures, and disease attributes, in addition to analysis of variance (ANOVA), were used to examine differences between T1D (distinguishing between glycemic clusters where relevant), healthy control variables, and neurobehavioral outcome measures. We considered the correlation to be significant when $R \geq |0.4|$ and the p-value< 0.05.

Voxel-based analysis (VBA) is a statistical method that detects differences in brain regions on a voxel-by-voxel basis (43) on segmented GM probability maps and DTI maps (FA, RD, AD, and volume). A two-sample t-test was used to compare the T1D and healthy groups.

Statistics of AFQ tract profiles: t-tests were calculated pointwise along each tract for diffusion properties. Given the high degree of correlation between nearby points, the Bonferroni correction was too conservative (38), and permutation-based multiple comparison correction (44) was used to adjust p-values given the structure of the data. This proved significant and resulted in a corrected p-value of<0.05.

The correlation between the locations of significant tract group differences and neurocognitive and neurobehavioral task performance was analyzed with MATLAB R2020a. Pearson and Spearman's correlations were employed using a p-value threshold of 0.05. For Pearson correlations, $r > |0.6|$ was applied, and for Spearman correlations, $r > |0.5|$.

Results

The study population comprised 30 adolescents. Of the 17 participants in the T1D group, 8 were boys (mean age, 14.7 ± 1.68 years). Of the 13 participants in the healthy group, 8 were boys (mean age, 14.6 ± 1.73 years). Table 1 shows their clinical and demographic parameters. Parental and participant baseline neurocognitive and language assessments were within normal ranges, with no significant differences within families.

Neurocognitive performance

One-way ANOVA analysis revealed significant group and cluster differences in cognitive function.

TABLE 1 Statistics: Clinical and demographic characteristics of the study population.

Group	T1D well-controlled group	T1D poorly controlled group	Healthy group	p-value
Number	8	9	13	NA
Gender (boys: girls)	3:5	5:4	8:5	0.45
Age†	14.5 ± 1.91	14.9 ± 1.54	14.64 ± 1.73	0.97
Puberty†	3.4 ± 1.51	3.7 ± 1.32	3.54 ± 1.71	0.99
T1D duration†	5.8 ± 2.83	7.2 ± 5.05	NA	NA
HbA_{1c}%††	7.6 ± 0.59	9.5 ± 1.03	NA	NA
HbA_{1c} (mmol/mol)	70.6		NA	NA
SEP cluster	6.80 ± 2.11		6.46 ± 1.81	0.66
SEP index	0.72 ± 0.92		0.59 ± 0.75	0.70
Height SDS	−0.16 ± 0.89		−0.40 ± 1.38	0.56
BMI SDS	0.10 ± 0.67		−0.19 ± 0.77	0.27

†Mean ± standard deviation.

††HbA_{1c} calculated from the last three visits during the past year at session 1.

The SEP (socioeconomic position) based on home address was analyzed according to the Israel Central Bureau of Statistics Characterization and Classification of Statistical Areas within Municipalities and Local Councils by Socio-Economic Level of the Population, 2015. The SEP index classifies neighborhoods and localities into clusters, with 1 being the lowest rating and 10 being the highest. It is an adjusted calculation of 14 variables that measure social and economic level in four domains—demographics, education, standard of living, and employment.

NA, not applicable.

Memory and learning

The performance of the TOMAL Word Selective Reminding task differed by group ($p = 0.03$) and by glycemic control ($p = 0.02$) and was associated with ISG ($r = 0.54$, $p = 0.002$). On average, lower HbA1c and lower ISG were associated with better scores.

Inhibition ability

Both group differences ($p = 0.001$) and glycemic control differences ($p = 0.004$) were found in the Stroop response time, which was associated with a higher ISG ($r = 0.65$, $p = 0.003$). Stroop non-food and food response times differed between groups ($p = 0.04$ and $p = 0.02$, respectively). Stroop food response time implied a significant difference between glycemic control clusters ($p = 0.05$). The relationship was such that better glycemic control and lower ISG were associated with better inhibition ability. On average, the response time of the T1D group was significantly higher than that of the control group.

Executive function performance

We found a significant difference in Visual Update response time by group ($p = 0.01$) and by glycemic control ($p = 0.04$): poorer glycemic control mean response time was 1,463 ms, whereas better glycemic control mean response time was 1,437 ms.

Semantic memory

On the New Object Recall Task (Figure 5), we observed significant difference in response time by groups (p=0.02), and by glycemic control (p=0.03). The response time in remembering whether the object had been presented was 2,943.2 msec among those with T1D, compared with a response time of 2,278.2 msec in the healthy control group. Poorer glycemic control associated with longer time needed to recall the object.

Anatomical brain differences between groups

Using VBA, the MRI data of each participant were analyzed and compared between groups. A *t*-test and multiple corrections on all diffusivity measures (FA, AD, and RD) in every tract of the corpus callosum segments and major WM bundles revealed significant differences between groups. FA was significantly lower in the T1D group, mainly in the superior longitudinal fasciculus (SLF) (0.25 ± 0.03 vs. 0.29 ± 0.02. $p = 0.0001$) and corona radiata (CR) (0.32 ± 0.05 vs. 0.38 ± 0.03. $p = 0.0002$). Lower AD was also observed in the T1D group ($p< 0.005$).

GM density measured by the GM probability index was higher in the T1D group in the Broca and Wernicke regions, connected by the WM fiber tracts mentioned above (SLF and CR) (Figure 6).

Comparison of diffusion properties in major brain segments

Diffusivity coefficients of corpus callosum clipped segments

The ANOVA on callosal segment diffusion properties revealed a significant difference between the T1D group and the healthy group in the superior frontal callosal segment centers FA ($p = 0.02$) and RD ($p = 0.03$). FA was significantly lower, and RD was significantly higher in the T1D group (Figure 7).

Diffusivity coefficients of major WM brain tracts

Using the AFQ framework for quantifying diffusion measurements at multiple locations along the trajectory of major WM tracts, a diffusion measurement "tract profile" was created at anatomically equivalent locations along T1D and

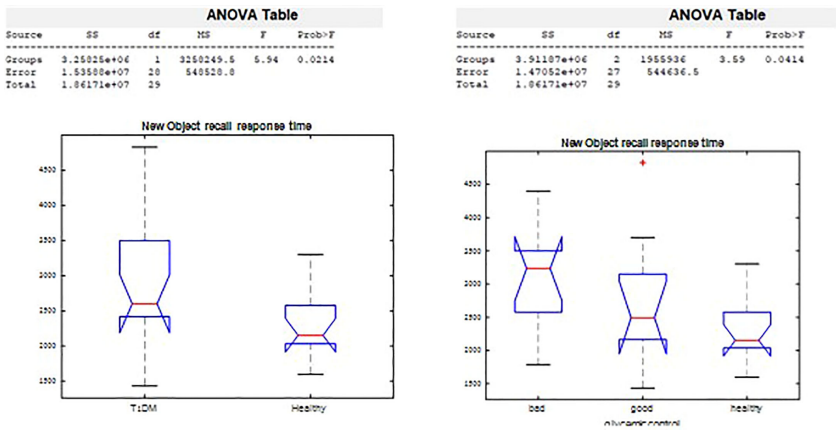

FIGURE 5
New Object Recall significant differences.

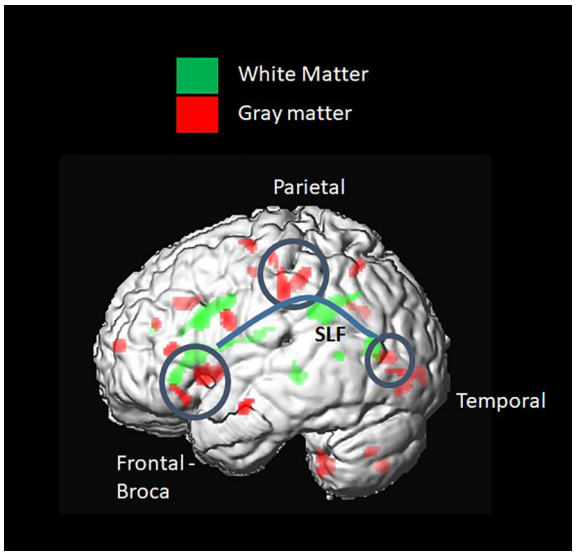

FIGURE 6
Voxel-based group comparison revealed a higher grey matter (GM) index in the type 1 diabetes (T1D) group. This included brain regions connected by white-matter (WM) fiber tracts, such as the SLF and corona radiata. A higher GM probability index was found in the frontal inferior triangular and operculum regions, known as Wernicke, and in parietal regions, such as the inferior parietal and postcentral gyrus.

healthy brain trajectories. Many locations along the observed tracts revealed group differences and were later associated with neurocognitive performance. A threshold of at least five locations (5%) with significant differences along T1D and healthy trajectories was applied to reveal anatomical group differences and to mark specific bundles. Of the eight fiber groups, the most apparent group differences in mean FA were seen in the superior frontal corpus callosum ($p< 0.005$) and the posterior parietal corpus callosum ($p< 0.005$) (Figure 8). Of the nine fiber groups, the most apparent group differences in mean AD were observed in the right superior longitudinal fasciculus (SLF) ($p< 0.001$), left uncinate fasciculus ($p< 0.04$), superior parietal corpus callosum ($p< 0.005$), forceps minor ($p< 0.005$), and anterior frontal corpus callosum ($p< 0.005$). Of the eight fiber groups, the most apparent differences in mean RD were found in the posterior parietal corpus callosum ($p< 0.001$), left arcuate ($p< 0.003$), left cingulum hippocampus ($p< 0.02$) and left SLF ($p< 0.03$). The differences along the tracts were in both directions, but most of the differences were in favor of the control group, suggesting denser and more coherent bundles.

Correlations between brain diffusion measures and cognitive performance

Right SLF quantitative diffusion parameters associated with Stroop response time

Stroop, Stroop food, and Stroop non-food response times correlated with the mean AD of the dorsal part of the right SLF (most significant $r = -0.77$ $p = 0.005$). The relationships were such that lower inhibitory ability was negatively associated with tract coherence.

Integrity of the superior frontal segment of the corpus callosum correlated with inhibition ability

Higher mean FA of the superior frontal segment of the corpus callosum, which connects the hemispheres, was significantly negatively correlated (most significant $r = -i.77$, $p = 0.006$) with emotional food Stroop response time. The relationship was such that higher bundle integrity was associated with a better ability to inhibit the emotional response to a food-related stimulus.

Posterior parietal corpus callosum density associated with verbal memory

Using Spearman correlation, performance on Word Selective Reminding (%) correlated with the mean RD of the left posterior parietal segment of the corpus callosum connecting the hemispheres (most significant $r = -0.63$ $p = 0.0008$). The relationship was such that higher bundle integrity was associated with better performance.

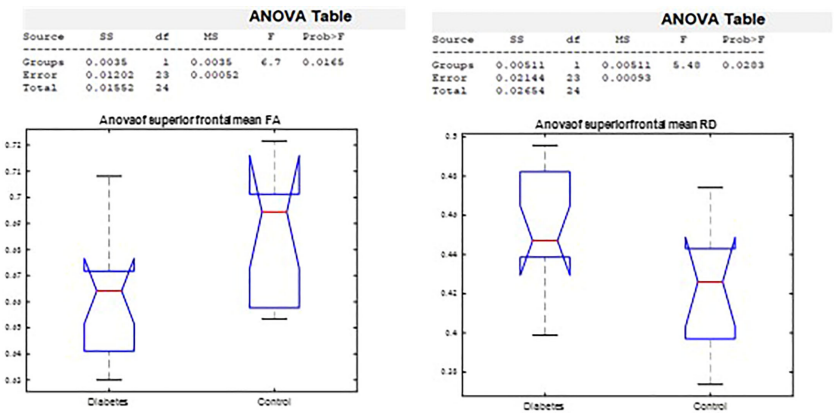

FIGURE 7
Analysis of variance by group in the superior frontal FA and RD corpus callosum segment center.

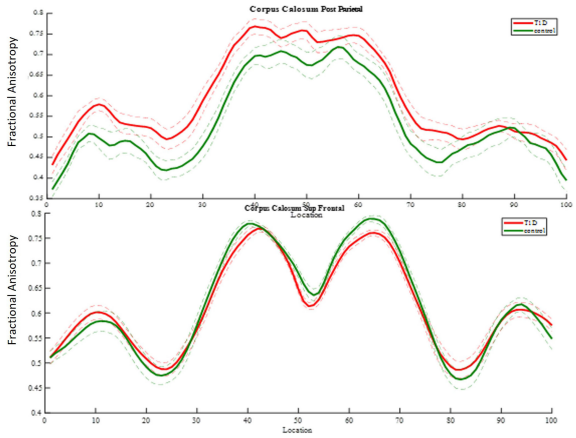

FIGURE 8
FA group comparison plots. Control group and diabetes mean FA ± 1 SD is plotted along 100 nodes on tract center. The upper graph shows significantly higher FA values in the diabetes group in 32 locations of the posterior parietal callosal segment. The panel beneath it shows significantly lower FA values in the diabetes group in 12 locations of the superior frontal callosal segment.

Discussion

A significant association was found between glycemic control and performance in the domains of EF, inhibition ability, semantic memory, and learning and verbal memory. Inhibition ability was found to be specifically related to food management. Poorer glycemic control (>8.3%) and greater glycemic excursions following a missed bolus meal were associated with longer reaction times in the cognitive tasks tested and poorer performance in learning and verbal memory. These findings highlight the impairment of brain networks responsible for learning, memory, and controlled food responsiveness in adolescents with T1D.

Neurobehavioral functions

The hypothesis tested here was that faulty glycemic control in adolescents with T1D, caused by inaccurate, late, or lack of meal boluses (15), may decrease performance even without previous risk factors such as a significant episode of DKA or hypoglycemic seizure (8).

A recent review (16) indicates that cognitive declines in young people with T1D are characterized by overall lower cognitive performance and moderately lower memory, attention, and EF. This is mostly found with early diabetes onset. It took 2 years from diabetes onset to show a moderate decline in these domains, which were still present after 6 and 12 years (45, 46). Collectively, the early age of diabetes onset, higher HbA1c, hypoglycemic events, and DKA around onset were all major contributors to cognitive decline. These factors may cause an initial "strike" to the brain, which later adapts to fluctuating glucose levels (16). Function during blinded CGMS and food-related assignments was not, however, assessed among adolescents.

Our study reveals group differences in inhibition abilities and executive function, with specific differences in the executive control of processing food stimuli. Poorer glycemic control and higher glycemic excursions were associated with poorer cognitive processing, learning, memory, and executive performance, supporting our hypothesized relationship between glycemic control and cognitive performance.

Brain structure

DTI tractography is considered a sound method for examining delicate differences in brain matter coherence, and we used this to evaluate the association between T1D and brain microstructure properties. The literature contains a variety of methods and study populations, all with inconclusive results concerning T1D and healthy populations regarding GM and WM volume and WM integrity (21, 22, 47). Associations between WM integrity and cognitive performance are sparse. The uniqueness of our study is its in-depth analysis, achieved by using many attributes and large-scale acquired data and performed in genetically and socioeconomically similar populations of adolescents. Several interesting findings and new insights into all these aspects pave the way for future, more focused studies.

As in other recent research, our results show significant differences in brain matter integrity between healthy adolescents and those with T1D. WM and GM differences were found in cognition-related brain structures, such as the SLF (Figure 9) and the corona radiata (CR), and in the GM structures that these tracts connect. Significant group differences were observed in the integrity of several tracts.

Most of our findings concern higher FA, higher AD, or lower RD in the healthy group compared with those with T1D. Most tracts examined were connected to frontal lobe segments or to fibers that connect frontal areas with other lobes, mainly temporal.

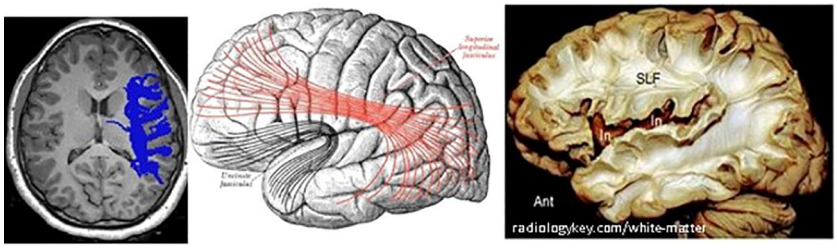

FIGURE 9
Right SLF white matter. Left: axial view of one diabetes patient (c016), right SLF pathway as traced by AFQ–Center: diagram showing the lateral surface of the left cerebral hemisphere. SLF is at the center, in red. Right: dissection of the human brain from the lateral aspect by the Klingler technique. Resecting the superficial digitations exposes the superior longitudinal fasciculus (SLF) superior to the sylvian fissure and insula (In).

Some significant differences were found, however, in the opposite direction. Significantly higher FA and restricted RD in the T1D group compared with healthy subjects were detected in several locations along the posterior parietal segment of the corpus callosum. This may suggest that its fiber bundles are denser in T1D patients than in the healthy group, making it reasonable to hypothesize that brain connectivity in T1D is modified by compensation processes. The posterior parietal segment of the corpus callosum receives fibers from the primary sensory area and connects the somesthetic association cortex with thalamic nuclei. It is mostly referred to as the somesthetic association area that restores our sensation's memory, permitting assessment, for example, of the characteristics of an object held in the hand (48).

Associations between brain structure and neurobehavioral functions

VBA group differences were observed between T1D patients and healthy controls in the SLF and the posterior CR. WM connectivity is very important in many spheres—motor, emotional, behavioral, linguistic, and cognitive. These abilities develop through childhood and adolescence and into early adulthood. The SLF connects multiple regions and is involved in language, attention, memory, and executive data processing. Few studies have addressed brain tract structure in adolescents with T1D, and none have focused on its correlation with specific neurocognitive functions (49). Our results concur with others who report decreased WM integrity in several brain areas in T1D (11, 50), in addition to neurocognitive connection to poorer glycemic control (12, 46). Our findings, however, derived using meticulous new methods and specific diabetes-oriented testing, are new in that they demonstrate statistically significant associations between cognitive performance and specific tract properties, WM quality, and neurocognitive performance. Stroop food and nonfood response times are strongly correlated with the integrity of the corpus callosum's superior frontal segment. It has recently been reported that the prefrontal cortex plays a dominant role in controlling food selection and intake, and its function may change in patients with diabetes due to its dependence on insulin metabolism (51).

Our study has limitations. Its population is smaller than planned because of the difficulty in recruiting adolescents willing to comply with all study procedures, hospitalization, and the lack of gain for the individual involved in the study. This prevented a sufficient number of subjects from demonstrating conclusively significant differences between groups before glycemic parameter considerations. Our study population is, however, a consistent homogeneous demographic group without the confounding variables of age, gender, puberty, comorbidities, quality of life, socioeconomic scale, and T1D complications. A second limitation was the Ethics Committee's restriction of MRI protocols to 25 min because of the young age of the study population, preventing cognitive tasking with glucose manipulation under imaging (functional MRI).

To conclude, our study sheds new light on cognitive brain domains that may be specifically associated with glycemic control and are strongly linked to missed pre-meal boluses (52, 53). A potentially highly relevant issue, even in this era of hybrid closed-loop therapy, is that the only barrier to not only improving but also controlling the target may be pre-meal bolus delivery.

Our findings emphasize that networks responsible for learning, memory, and controlled reactivity to food may be compromised in adolescents with T1D. We present several exciting results that call for further investigation. Although the associations we discovered comply with the known functions of major brain tracts, further research is needed to identify the processes that establish these associations.

Data availability statement

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

Ethics statement

Studies involving human participants were reviewed and approved by Israel's Health Ministry's Helsinki Committee. Written informed consent to participate in this study was provided by the participants' legal guardian and an assent form was signed by the participant.

Author contributions

EL, RG and MR designed the study, analyzed and interpreted the data, and wrote the manuscript. AL and TS contributed to the study design, collected data, and reviewed and edited the manuscript. ML and EH collected data and reviewed and edited the manuscript. MG, JY, and ES collected data, analyzed and interpreted them, and reviewed and edited the manuscript. MR and EL designed the study, MR and AL were responsible for data collection . EL, RG and MR analyzed and interpreted the results. MR and El are the guarantors of this work. As such, they have full access to the dataset (EL is blinded to the names of the patients) and is responsible for the integrity of the data and the accuracy of the data analysis. All authors contributed to the article and approved the submitted version.

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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.

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References

1. Litmanovitch E, Geva R, Rachmiel M. Short and long term neuro-behavioral alterations in type 1 diabetes mellitus pediatric population. *World J Diabetes* (2015) 6:259–70. doi: 10.4239/wjd.v6.i2.259

2. Mauras N, Buckingham B, White NH, Tsalikian E, Weinzimer SA, Jo B, et al. Impact of type 1 diabetes in the developing brain in children: A longitudinal study. *Diabetes Care* (2021) 44:983–92. doi: 10.2337/dc20-2125

3. Miles R, Root F. Psychologic tests applied to diabetic patients. *Arch Intern Med* (1922) 30:767–77. doi: 10.1001/archinte.1922.00110120086003

4. Slattery D, Amiel SA, Choudhary P. Optimal prandial timing of bolus insulin in diabetes management: A review. *Diabetic Med* (2018) 35:306–16. doi: 10.1111/dme.13525

5. Tönnies T, Imperatore G, Hoyer A, Saydah SH, D'Agostino RB, Divers J, et al. Estimating prevalence of type I and type II diabetes using incidence rates: The SEARCH for diabetes in youth study. *Ann Epidemiol* (2019) 37:37–42. doi: 10.1016/j.annepidem.2019.07.006

6. ADA. 13. children and adolescents: Standards of medical care in diabetes–2019. *Diabetes Care* (2019) 42:S148–64. doi: 10.2337/dc19-S013

7. Prahalad P, Zaharieva DP, Addala A, New C, Scheinker D, Desai M, et al. Improving clinical outcomes in newly diagnosed pediatric type 1 diabetes: Teamwork, targets, technology, and tight control–the 4T study. *Front Endocrinol* (2020) 11. doi: 10.3389/fendo.2020.00360

8. Olinder AL, Kernell A, Smide B. Missed bolus doses: Devastating for metabolic control in CSII-treated adolescents with type 1 diabetes. *Pediatr Diabetes* (2009) 10:142–8. doi: 10.1111/j.1399-5448.2008.00462.x

9. Mauras N, Mazaika P, Buckingham B, Weinzimer S, White NH, Tsalikian E, et al. Longitudinal assessment of neuroanatomical and cognitive differences in young children with type 1 diabetes: Association with hyperglycemia. *Diabetes* (2015) 64:1770–9. doi: 10.2337/db14-1445

10. Arbelaez AM, Semenkovich K, Hershey T. Glycemic extremes in youth with T1DM: The structural and functional integrity of the developing brain. *Pediatr Diabetes* (2013) 14:541–53. doi: 10.1111/pedi.12088

11. Fox LA, Hershey T, Mauras N, Arbeláez AM, Tamborlane WV, Buckingham B, et al. Persistence of abnormalities in white matter in children with type 1 diabetes. *Diabetologia* (2018) 61:1538–47. doi: 10.1007/s00125-018-4610-6

12. DiMeglio LA, Kanapka LG, DeSalvo DJ, Anderson BJ, Harrington KR, Hilliard ME, et al. Time spent outside of target glucose range for young children with type 1 diabetes: a continuous glucose monitor study. *Diabetic Med* (2020) 37:1308–15. doi: 10.1111/dme.14276

13. Cato A, Hershey T. Cognition and type 1 diabetes in children and adolescents. *Diabetes Spectr* (2016) 29:197–202. doi: 10.2337/ds16-0036

14. Cameron FJ. The impact of diabetes on brain function in childhood and adolescence. *Pediatr Clinics North America* (2015) 62:911–27. doi: 10.1016/j.pcl.2015.04.003

15. Datye KA, Boyle CT, Simmons J, Moore DJ, Jaser SS, Sheanon N, et al. Timing of meal insulin and its relation to adherence to therapy in type 1 diabetes. *J Diabetes Sci Technol* (2017) 12:349–55. doi: 10.1177/1932296817728525

16. van Duinkerken E, Snoek FJ, de Wit M. The cognitive and psychological effects of living with type 1 diabetes: a narrative review. *Diabetic Med* (2020) 37:555–63. doi: 10.1111/dme.14216

17. Jongen C, Biessels GJ. Structural brain imaging in diabetes: A methodological perspective. *Eur J Pharmacol* (2008) 585:208–18. doi: 10.1016/j.ejphar.2007.11.085

18. Bassett DS, Brown JA, Deshpande V, Carlson JM, Grafton ST. Conserved and variable architecture of human white matter connectivity. *NeuroImage* (2011) 54:1262–79. doi: 10.1016/j.neuroimage.2010.09.006

19. Aye T, Barnea-Goraly N, Ambler C, Hoang S, Schleifer K, Park Y, et al. White matter structural differences in young children with type 1 diabetes: A diffusion tensor imaging study. *Diabetes Care* (2012) 35:2167–73. doi: 10.2337/dc12-0017

20. Brouwer RM, Mandl RC, Schnack HG, van Soelen IL, van Baal GC, Peper JS, et al. White matter development in early puberty: A longitudinal volumetric and diffusion tensor imaging twin study. *PloS One* (2012) 7:e32316. doi: 10.1371/journal.pone.0032316

21. Antenor-Dorsey JAV, Meyer E, Rutlin J, Perantie DC, White NH, Arbelaez AM, et al. White matter microstructural integrity in youth with type 1 diabetes. *Diabetes* (2013) 62:581–9. doi: 10.2337/db12-0696

22. Barnea-Goraly N, Raman M, Mazaika P, Marzelli M, Hershey T, Weinzimer SA, et al. Alterations in white matter structure in young children with type 1 diabetes. *Diabetes Care* (2013) 37. doi: 10.2337/dc13-1388

23. Biessels GJ, Reijmer YD. Brain changes underlying cognitive dysfunction in diabetes: What can we learn from MRI? *Diabetes* (2014) 63:2244–52. doi: 10.2337/db14-0348

24. Mills KL, Tamnes CK. Methods and considerations for longitudinal structural brain imaging analysis across development. *Dev Cogn Neurosci* (2014) 9:172–90. doi: 10.1016/j.dcn.2014.04.004

25. Jeurissen B, Descoteaux M, Mori S, Leemans A. Diffusion MRI fiber tractography of the brain. *NMR Biomed* (2019) 32:e3785. doi: 10.1002/nbm.3785

26. ADA. Standards of medical care in diabetes–2014. *Diabetes Care* (2014) 37 (Suppl. 1):S14–80. doi: 10.2337/dc14-S014

27. Miller KM, Foster NC, Beck RW, Bergenstal RM, DuBose SN, DiMeglio LA, et al. Current state of type 1 diabetes treatment in the U.S.: Updated data from the T1D exchange clinic registry. *Diabetes Care* (2015) 38:971–8. doi: 10.2337/dc15-0078

28. I.C.B.o.S. Characterization and classification of geographical units by the socio-economic level of the population, 2015. (2019).

29. Goldstein A, Haelyon U, Krolik E, Sack J. Comparison of body weight and height of Israeli schoolchildren with the tanner and centers for disease control and prevention growth charts. *Pediatrics* (2001) 108(6):E108. doi: 10.1542/peds.108.6.e108

30. Marshall WA, Tanner JM. Growth and physiological development during adolescence. *Annu Rev Med* (1978) 19:283–300. doi: 10.1146/annurev.me.19.020168.001435

31. Reynolds CR, Bigler ED. Factor structure, factor indexes, and other useful statistics for interpretation of the test of memory and learning (TOMAL). *Arch Clin Neuropsychol* (1996) 11:29–43. doi: 10.1093/arclin/11.1.29

32. Thaler NS. Cluster analysis of the TOMAL standardization sample. Developmental Psychology, University of Nevada, Las Vegas (2010).

33. Gerstadt CL, Hong YJ, Diamond A. The relationship between cognition and action: performance of children 3 1/2-7 years old on a stroop-like day-night test. *Cognition* (1994) 53:129–53. doi: 10.1016/0010-0277(94)90068-X

34. Ramon D, Geva R, Goldstein A. Trait and state negative affect interactions moderate inhibitory control performance in emotionally loaded conditions. *Pers Individ Dif* (2011) 51:95–101. doi: 10.1016/j.paid.2011.03.016

35. Miyake A, Friedman NP, Emerson MJ, Witzki AH, Howerter A, Wager TD. The unity and diversity of executive functions and their contributions to complex "Frontal lobe" tasks: A latent variable analysis. *Cogn Psychol* (2000) 41:49–100. doi: 10.1006/cogp.1999.0734

36. Kraut M, Calhoun V, Pitcock J, Cusick C, Hart J. Neural hybrid model of semantic object memory: Implications from event-related timing using fMRI. *J Int Neuropsychol Soc: JINS* (2003) 9:1031–40. doi: 10.1017/S135561770397007X

37. Assaf M, Calhoun VD, Kuzu CH, Kraut MA, Rivkin PR, Hart J, et al. Neural correlates of the object-recall process in semantic memory. *Psychiatry Res: Neuroimaging* (2006) 147:115–26. doi: 10.1016/j.pscychresns.2006.01.002

38. Yeatman JD, Dougherty RF, Myall NJ, Wandell BA, Feldman HM. Tract profiles of white matter properties: Automating fiber-tract quantification. *PloS One* (2012) 7:e49790. doi: 10.1371/journal.pone.0049790

39. Mori S, van Zijl PCM. Fiber tracking: priciples and strategies - a technical review. *NMR Biomed* (2002) 15:468–80. doi: 10.1002/nbm.781

40. Huang H, Zhang J, Jiang H, Wakana S, Poetscher L, Miller MI, et al. DTI tractography based parcellation of white matter: Application to the mid-sagittal morphology of corpus callosum. *NeuroImage* (2005) 26:195–205. doi: 10.1016/j.neuroimage.2005.01.019

41. Dougherty RF, Ben-Shachar M, Deutsch GK, Hernandez A, Fox GR, Wandell BA. Temporal-callosal pathway diffusivity predicts phonological skills in children. *Proc Natl Acad Sci U.S.A.* (2007) 104:8556–61. doi: 10.1073/pnas.0608961104

42. Mori S, Crain B, Chacko VP, van zijl P. Three dimensional tracking of axonal projections in the brain by magnetic resonance imaging. annal neurol 45: 265-269. *Ann Neurol* (1999) 45:265–9. doi: 10.1002/1531-8249(199902)45:2<265::AID-ANA21>3.0.CO;2-3

43. Sasson E, Doniger GM, Pasternak O, Tarrasch R, Assaf Y. White matter correlates of cognitive domains in normal aging with diffusion tensor imaging. *Front Neurosci* (2013) 7:32–2. doi: 10.3389/fnins.2013.00032

44. Nichols TE, Holmes AP. Nonparametric permutation tests for functional neuroimaging: A primer with examples. *Hum Brain Mapp* (2002) 15:1–25. doi: 10.1002/hbm.1058

45. Ryan CM. Why is cognitive dysfunction associated with the development of diabetes early in life? *diathesis hypothesis. Pediatr Diabetes* (2006) 7:289–97. doi: 10.1111/j.1399-5448.2006.00206.x

46. van Duinkerken E, Ryan CM. Diabetes mellitus in the young and the old: Effects on cognitive functioning across the life span. *Neurobiol Dis* (2020) 134:104608. doi: 10.1016/j.nbd.2019.104608

47. Aye T, Reiss AL, Kesler S, Hoang S, Drobny J, Park Y, et al. The feasibility of detecting neuropsychologic and neuroanatomic effects of type 1 diabetes in young children. *Diabetes Care* (2011) 34:1458–62. doi: 10.2337/dc10-2164

48. Kesner RP. The posterior parietal cortex and long-term memory representation of spatial information. *Neurobiol Learn Memory* (2009) 91:197–206. doi: 10.1016/j.nlm.2008.09.004

49. Pourabbasi A, Tehrani-Doost M, Qavam SE, Arzaghi SM, Larijani B. Association of diabetes mellitus and structural changes in the central nervous system in children and adolescents: A systematic review. *J Diabetes Metab Disord* (2017) 16:10. doi: 10.1186/s40200-017-0292-8

50. Toprak H, Yetis H, Alkan A, Filiz M, Kurtcan S, Aralasmak A, et al. Relationships of DTI findings with neurocognitive dysfunction in children with type 1 diabetes mellitus. *Br J Radiol* (2016) 89:20150680. doi: 10.1259/bjr.20150680

51. Kullmann S, Kleinridders A, Small DM, Fritsche A, Häring H-U, Preissl H, et al. Central nervous pathways of insulin action in the control of metabolism and food intake. *Lancet Diabetes Endocrinol* (2020) 8:524–34. doi: 10.1016/S2213-8587(20)30113-3

52. Gomes MB, Negrato CA. Adherence to insulin therapeutic regimens in patients with type 1 diabetes. a nationwide survey in Brazil. *Diabetes Res Clin Pract* (2016) 120:47–55. doi: 10.1016/j.diabres.2016.07.011

53. Bermeo-Cabrera J, Almeda-Valdes P, Riofrios-Palacios J, Aguilar-Salinas CA, Mehta R. Insulin adherence in type 2 diabetes in Mexico: Behaviors and barriers. *J Diabetes Res* (2018) 2018:3190849. doi: 10.1155/2018/3190849

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Investigation of treatment satisfaction and health-related quality of life after add-on to metformin-based therapy in patients with type 2 diabetes

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Background: The complexity of oral antidiabetic drug (OAD) regimens affects the quality of life (QOL) and treatment satisfaction. However, data on the QOL of patients with type 2 diabetes mellitus (T2DM) receiving metformin-based OAD treatment in Asia are limited. Therefore, this study aimed to evaluate the QOL and treatment satisfaction and explore the influencing factors and their correlations among patients with T2DM receiving metformin-based OADs.

Methods: This was a cross-sectional study conducted at the Outpatient Department of Metabolism and Endocrinology at a medical center in Taiwan. Data were collected using the Audit of Diabetes-Dependent Quality of Life (ADDQoL) and the Chinese version of the Satisfaction with Oral Anti-Diabetic Agent Scale (C-SOADAS) questionnaires from patients with T2DM using metformin. The outcomes were analyzed by group and stratified based on the use of two, three, and more than three OADs. The level of agreement between the questionnaires was analyzed using Spearman's rank correlation coefficient.

Results: A total of 153 patients with T2DM using metformin were included in this study. The average weighted impact score in the ADDQoL was −2.11, with no significant differences between the three groups. The C-SOADAS score showed a significant difference between the groups using two, three, and more than three OADs (21.42 [1.98] vs. 20.43 [2.09] vs. 19.00 [2.24], $p<0.0001$). The ADDQoL and C-SOADAS scores showed low correlations between patients' QOL and treatment satisfaction. However, the impact of diabetes on specific aspects of life was negatively correlated with the total C-SOADAS scores.

Conclusion: In Taiwan, a significantly greater effect on QOL was observed among patients with fewer OAD classes and higher treatment satisfaction. This study provides local evidence from self-reporting outcomes of patients with T2DM. Further studies focusing on different populations and treatment regimens for QOL are needed.

KEYWORDS

type 2 diabetes, ADDQoL, C-SOADAS, quality of life, treatment satisfaction

Introduction

Type 2 diabetes (T2D) has been one of the fastest growing diseases in the past decade, and its complications have a major impact on health-related quality of life (HRQoL) (1, 2). Previous studies showed that reducing the development of hypoglycemia can improve patients' treatment satisfaction and HRQoL and attain glycemic control (3–6). Furthermore, increasing treatment satisfaction and HRQoL in patients with T2D are associated with a lower occurrence of diabetes complications and mortality (7, 8).

Metformin is usually the first-line therapy for patients with T2D. Adding second-line regimens, including various combinations of oral antidiabetic drugs (OADs), when glycated hemoglobin (HbA1c) does not reach the target is recommended by current guidelines. However, medical benefits and risks should be considered when choosing second-line OADs, especially given the current limited information on treatment satisfaction and HRQoL. Previous studies showed that improving patients' treatment satisfaction can enhance treatment efficacy and adherence, and optimal glycemic control can reduce comorbidities and improve HRQoL. However, the high regimen complexity of OADs may lower treatment satisfaction and HRQoL due to the risk of side effects (9).

According to pharmacoepidemiology research, 87.5% of outpatients with diabetes use OADs in Taiwan (10). However, most research on OAD strategies in Taiwan has focused on diabetes control and treatment-related adverse events. Furthermore, data on patient-reported outcomes regarding treatment satisfaction and HRQoL after the addition of the second-line OAD strategy are limited. Thus, this study aimed to survey the treatment satisfaction and HRQoL for patients with T2D receiving add-on therapy to a metformin-based regimen.

Methods

Study population

Patients with T2D who visited endocrinology and metabolism outpatient clinics at Kaohsiung Chang Gung Memorial Hospital, a medical center in southern Taiwan, between April 2020 and June 2021 were enrolled. The inclusion criteria included patients who had been using metformin-based therapy for at least 12 weeks and who were receiving add-on therapy during the outpatient visit (index date). The exclusion criteria included patients who received injection therapy, including glucagon-like peptide-1 receptor agonist or insulin, had received chemotherapy within 6 months before the index date, had no willingness to fill out the consent form or questionnaire, and had a diagnosis of type 1 diabetes, gestational diabetes, or cognitive impairment. The included patients were asked to complete the Chinese version of the Audit of Diabetes-Dependent Quality of Life (ADDQoL) (11, 12) for Taiwan and the Chinese version of the Satisfaction with Oral Anti-Diabetic Agent Scale (C-SOADAS) (13) at the index date and 3 months after the index date, respectively.

This study was approved by the Institutional Review Board of the Chang Gung Medical Foundation (no. 202000024B0C501). All participants provided written informed consent.

Sample size calculation

Daniel's (14) sample size formula was used to calculate the minimum sample size, where 92 patients with T2D were considered a representative sample size for this study. Based on the previously published data on T2D prevalence and more than one OAD in Taiwan, a prevalence (P) of 60%, a desired precision of 10%, and a 95% confidence level were used (10).

Questionnaire measurement

In this study, the ADDQoL and C-SOADAS questionnaires in Taiwan were used to measure the diabetes-specific quality of life (QOL) and treatment satisfaction, respectively. The translated versions of these questionnaires were analyzed for reliability and validity, and all authors agreed to use them as research instruments for this study (11–13).

The Taiwanese version of the ADDQoL is a widely used tool for assessing diabetes-related QOL and consists of two overview items to measure generic QOL and 19 specific domains of life. The product of the impact rating and importance rating score for each domain is the weighted impact score, and the weighted impact scores are added and divided by the number of applicable domains to yield the overall average weighted impact (AWI) score, which ranges from −9 (maximum negative impact) to +3 (maximum positive impact). Negative AWI scores indicate that diabetes has a significant negative impact on QOL.

The Taiwan version of the C-SOADAS is a tool for evaluating treatment satisfaction based on a five-item scale, focusing on concepts related to satisfaction with OADs among patients with type 2 diabetes mellitus (T2DM), including (1) ability to control blood sugar, (2) effect on weight, (3) tolerability of the side effects, (4) convenience of drug taking, and (5) overall satisfaction. Each item was scored on a 5-point scale (ranging from 1 to 5). After adding up the scores of the five items, with 25 being the highest score, higher scores indicate higher satisfaction with OADs.

All participants were referred to the study by physicians. The study aim, methods, and consent form contents were explained clearly to eligible subjects by the researchers before inclusion. The questionnaire was completed after the consent form was completed and signed. The patients were allowed to complete the questionnaire by themselves during the index date. The questionnaires were administered once to each participant.

Outcomes

The primary outcome was the ADDQoL and C-SOADAS questionnaire scores, and the secondary outcome was ascertained to assess the convergent validity of the questionnaire correlations for ADDQoL and C-SOADAS.

Comorbidities and covariables

Basic data, including demographics, comorbidities, and laboratory values, were collected within 1 year from the index date and were

based on the most recent data available. Demographic data included age, sex, duration of diabetes, family history of diabetes, smoking status, alcohol status, self-monitoring of blood glucose levels, and body mass index. Comorbidities included hypertension, dyslipidemia, cerebra/cardiovascular disease, peripheral vascular disease, neurological disease, retinopathy, and nephropathy. Laboratory data included HbA1c (<7% and ≥ 7%) and urine albuminuria-to-creatinine ratio (UACR) (<30, 30–300, and > 300). The use of fixed-dose combination OADs was evaluated based on the index date prescription.

Statistical analysis

Basic data and questionnaire scores were presented as mean (standard deviation) for normally distributed variables and median (interquartile range) for non-normally distributed continuous variables. Categorical variables were presented as numbers and percentages and analyzed using the Chi-square test. Study participants were divided into three groups according to the number of OADs they used (two, three, and more than three OADs). The difference in C-SOADAS and ADDQoL items was determined using one-way ANOVA. The levels of agreement between questionnaires were analyzed using Spearman's rank correlation coefficient, which was interpreted as follows: <0.300, low correlation; 0.301–0.700, moderate correlation; and >0.701, high correlation. Data processing was performed using Microsoft Excel, and statistical analysis was performed using SPSS software.

Results

A total of 156 patients with T2D were enrolled. Of the 156 patients, three were excluded (one refused to complete the questionnaire, and two were diagnosed with malignancy). Thus, 153 participants were included in the analysis (Figure 1). The characteristics of the total and subgroups of the study population are shown in Table 1. The mean age was 60.4 years, and most of the patients were 55–69 years old. The mean duration of T2D was 9.2 years, and 64.7% of the patients had a family history of diabetes. Only 43.79% of the patients had the habit of self-monitoring their blood glucose levels. Most of the patients had no history of smoking (81.70%) or alcohol consumption (81.05%), and nearly half of them (47.75%) were obese. Hypertension (51.63%) and dyslipidemia (89.54%) were the most prevalent comorbidities. In this study, the proportion of fixed-dose combination OADs was 71.24%.

Based on the participant classification (Table 1), the mean duration of T2DM was 7.3 (4.9) years for the group with two OADs, 10.2 (6.4) years for the group with three OADs, and 11.1 (5.3) years for the group with more than three OADs, with a significant difference among the three groups ($p = 0.0015$). The proportion of patients with obesity was the highest in the group with more than three OADs (56.1%), with a significant difference between the three groups ($p = 0.0214$). The group with more than 3 OADs had the highest proportion of hypertension (68.3%). Family history of diabetes mellitus (DM), alcohol consumption, and home self-monitoring of blood glucose did not differ significantly among the three groups. In glycemic control, a significant difference in HbA1c levels was observed between the three groups (7.19% vs. 7.19% vs. 7.79%, $p = 0.0035$).

The ADDQoL scores are shown in Tables 2, 3. The mean and overall QOL scores were 0.86 (0.84) and 0.88 (0.86) for the group with two OADs, 0.79 (0.75) for the group with three OADs, and 0.92 (0.91) for the group with more than 3 OADs. No significant difference in the overall QOL was observed among the three groups ($p = 0.7274$). The mean scores for the impact of diabetes on specific life aspects were −1.68 (0.72) in all patients and −1.54 (0.73) for the group with two OADs, −1.87 (0.71) for the group with three OADs, and −1.68 (0.69) for the group with more than three OADs. The AWI scores were −2.11 (1.08) in the total participant population and −2.05 (1.08), −2.10 (0.90), and −2.23 (1.27) in the three groups, respectively, with no significant differences between the three groups. The group with more than three OADs had a higher negative impact on close personal relationships (−3.53) than the other two groups ($p = 0.0433$).

A significant difference in the C-SOADAS and total scores was observed among the three groups (21.42 [1.98] vs. 20.43 [2.09] vs.

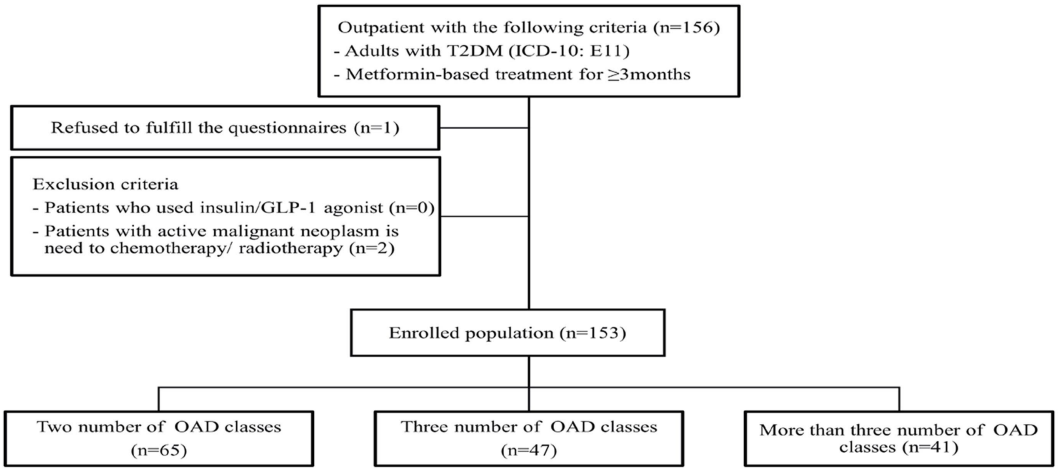

FIGURE 1
Study flow diagrams. T2DM, type 2 diabetes mellitus; OADs, oral anti-diabetic drugs; ICD-10, international classification of disease 10th revision.

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TABLE 1 Characteristics of all patients and stratified by the different oral antidiabetic drug groups.

Variable	Total (*n* =153)	2 OADs (*n* =65)	3 OADs (*n* =47)	>3 OADs (*n* =41)	*p* value
DM duration	9.22 (5.70)	7.32 (4.86)	10.17 (6.43)	11.07 (5.29)	0.002
Age(years)	60.39 (10.14)	61.32 (9.46)	58.72 (11.41)	60.84 (9.52)	0.388
Gender					0.035
Female	65 (42.48)	35 (53.85)	18 (38.30)	12 (29.27)	
Male	88 (57.52)	30 (46.15)	29 (67.70)	29 (70.73)	
DM family history					0.870
No	42 (27.45)	18 (27.69)	14 (29.79)	10 (24.39)	
Yes	99 (64.71)	43 (66.15)	28 (59.57)	28 (68.29)	
Unknown	12 (7.84)	4 (6.15)	5 (10.64)	3 (7.32)	
Smoke					0.036
Current	15 (9.80)	2 (3.08)	4 (8.51)	9 (21.95)	
Past	13 (8.50)	5 (7.69)	5 (10.64)	3 (7.32)	
Never	125 (81.70)	58 (89.23)	38 (80.85)	29 (70.73)	
Alcohol					0.132
Yes	29 (18.95)	9 (13.85)	8 (17.02)	12 (29.27)	
Never	124 (81.05)	56 (86.15)	39 (82.98)	29 (70.73)	
Self-monitoring blood glucose	67 (43.79)	32 (49.23)	17 (36.17)	18 (43.90)	0.389
Body mass index					0.021
Underweight	2 (1.31)	2 (3.07)	-	-	
Normal	34 (22.22)	18 (27.69)	14 (29.79)	2 (4.88)	
Overweight	47 (30.72)	21 (32.81)	10 (21.28)	16 (39.02)	
Obese	70 (45.75)	24 (36.92)	23 (48.94)	23 (56.10)	
Fixed-dose combination OADs					<0.0001
Yes	109 (71.24)	25 (38.46)	45 (95.74)	39 (95.12)	
No	44 (28.76)	40 (61.54)	2 (4.26)	2 (4.88)	
DM related disease					
Hypertension	79 (51.63)	27 (41.54)	24 (51.06)	28 (68.29)	0.027
Dyslipidemia	137 (89.54)	57 (87.69)	41 (87.23)	39 (95.12)	0.393
Cerebra/cardiovascular disease	10 (6.54)	4 (6.15)	2 (4.26)	4 (9.76)	0.649
Peripheral vascular disease	2 (1.31)	1 (1.54)	0 (0.00)	1 (2.44)	0.737
Neuropathy	14 (9.15)	3 (4.62)	6 (12.77)	5 (12.20)	0.218
Retinopathy	2 (1.31)	0 (0.00)	2 (4.26)	0 (0.00)	0.164
Nephropathy	60 (39.22)	20 (30.77)	19 (40.43)	21 (51.22)	0.108
Plasma glucose (mg/dL)	163.16 (50.62)	124.31 (48.06)	159.62 (46.68)	181.27 (55.31)	0.023
HbA1c (%)	7.35 (0.99)	7.11 (0.95)	7.26 (0.99)	7.79 (0.93)	0.004
<7%	54 (35.29)	26 (40.00)	21 (44.68)	7 (17.07)	0.012
≥7%	99 (64.71)	39 (60.00)	26 (55.32)	34 (82.93)	
UACR (mg/g)					0.093
<30	109 (71.71)	52 (81.25)	34 (72.34)	23 (56.10)	
30–300	31 (20.39)	9 (14.06)	9 (19.15)	13 (31.71)	
>300	12 (7.89)	3 (4.69)	4 (8.51)	5 (12.20)	

Values are displayed as *n* (%), mean ± standard deviation. DM, diabetes mellitus; HbA1c, glycated hemoglobin A1c; OADs, oral antidiabetic drugs.

TABLE 2 ADDQoL and C-SOADAS of all participants.

Item	All participants (n =153)
ADDQoL	
Overview questions	
Present QoL	0.86 ± 0.84
Diabetes-dependent QoL score	−1.68 ± 0.72
19 domain-specific items	
Leisure activities	−1.78 ± 1.71
Working life	−1.69 ± 1.66
Journeys	−1.75 ± 1.77
Holidays	−1.98 ± 1.81
Physical health	−2.38 ± 1.86
Family life	−3.46 ± 2.34
Friendship and social life	−1.35 ± 1.80
Close personal relationship	−2.78 ± 2.12
Sex life	−1.85 ± 1.89
Physical appearance	−1.90 ± 1.88
Self-confidence	−1.51 ± 1.92
Motivation	−1.48 ± 1.55
People's reaction	−0.76 ± 1.17
Feeling about future	−3.49 ± 2.33
Financial situation	−0.75 ± 1.36
Living conditions	−0.59 ± 1.16
Dependence on others	−1.48 ± 1.67
Freedom to eat	−4.75 ± 2.75
Freedom to drink	−4.57 ± 3.00
Average weighted impact score	−2.11 ± 1.08
C-SOADAS	
Q1: Ability to control blood sugar	4.20 ± 0.76
Q2: Effect of weight	3.90 ± 0.63
Q3: Tolerability of the side effect	4.29 ± 0.53
Q4: Convenience of drug taking	3.86 ± 0.76
Q5: Overall satisfaction	4.23 ± 0.58
Total C-SOADAS score	20.46 ± 2.29

Value is displayed as mean ± standard deviation. ADDQoL, Audit of Diabetes-Dependent Quality of Life; C-SOADAS, Chinese version of the Satisfaction with Oral Antidiabetic drugs; QoL, quality of life; AWI, Average weighted impact score.

19.00 [2.24], $p < 0.0001$) (Tables 2, 4). The mean scores for the ability to control blood sugar, tolerability of the side effects, convenience of drug taking, and overall satisfaction were significantly higher in the group using two OADs than in the other two groups ($p < 0.05$). The effect of OADs on body weight was higher in the group with two OADs than in the groups with three and more than three OADs, with no statistically significant difference (3.98 [0.67] vs. 3.91 [0.50] vs. 3.73 [0.67], $p = 0.1276$).

Regarding the correlation between the ADDQoL and AWI scores, as represented by Spearman's rank correlation coefficient (R_s), a weak correlation was observed between the general overall QOL and AWI scores. However, a statistically significant moderate correlation was observed between the impact of diabetes on specific aspects of life and AWI scores ($R_s = 0.350$, $p < 0.0001$). The four factors of ADDQoL were moderately to highly correlated with the AWI scores ($p < 0.0001$), as shown in Supplementary Appendix 1. The ADDQoL and C-SOADAS scores showed correlations between QOL and treatment satisfaction (Supplementary Appendix 2). Among the two overview items of ADDQoL, the impact of overall QOL was positively correlated with the total C-SOADAS score, while the impact of diabetes on specific aspects of life was negatively correlated with the total C-SOADAS score, but neither reached a statistically significant difference. A significant low-to-moderate correlation was observed between the items of effect on weight, but no significant correlations were observed between the other items of treatment satisfaction.

Discussion

The study results showed that patients with T2DM receiving metformin-based therapy using more than three OADs had the lowest HRQoL compared with those with less than or equal to three OADs. Furthermore, the treatment satisfaction of patients receiving two OADs was significantly higher than that of the other treatment combinations.

In the study, HbA1c was significantly higher in the group with more than three OADs (7.8% ± 0.9%) than in the other two groups, and 80% of those had poorer glycemic control (HbA1c ≥ 7%). These results are consistent with those of previous studies (15). A greater number of prescribed OADs together were correlated with more comorbidities, obesity, smoking, and high blood pressure, which increased insulin resistance. Previous studies showed that the use of multiple (>3) OADs for treating patients has been highly correlated with poor glycemic control and increasing duration of diabetes (16, 17). Furthermore, physical impairment has been observed in frail hypertensive older adults with hyperglycemia (18). Although this study included patients using OADs that had a negative impact on HRQoL, patients treated with two OADs had a higher HRQoL in relation to diabetes than those treated with three or more OADs.

Furthermore, this study revealed that whether patients used two, three, or more than three OADs, the domain of "dietary freedom" had the most significant effect on diabetes. A strong association was observed between diabetes and diet, and dietary control was involved in the management of T2DM as a major life factor and influenced the long-term outcomes of the disease, including depression (19, 20). Diabetes has a significant negative impact on diet, and the same results are seen in patients treated with OADs or insulin. Several studies on factors associated with diabetes treatment satisfaction showed that the use of insulin is negatively associated with treatment satisfaction (21). However, whether T2DM treated with OADs is positively or negatively associated with treatment satisfaction remains controversial and is only compared with dietary control (6, 22). These inconsistent results may be partly due to differences in the OADs tested.

However, the longer the duration of DM, the greater the number of OADs required for combination therapy owing to insufficient insulin secretion and poor glycemic control (15). Patients using OADs have been shown to have higher satisfaction levels than those using insulin. Furthermore, those treated with fewer drugs have significantly better satisfaction scores than those treated with multiple combination OADs (23, 24). The European multinational PANORAMA study (25)

TABLE 3 ADDQoL score in different oral antidiabetic drug groups.

Item	2 OADs (*n* =65)	3 OADs (*n* =47)	>3 OADs (*n* =41)	*p* value
Overview questions				
Present QoL	0.88 ± 0.86	0.79 ± 0.75	0.92 ± 0.91	0.727
Diabetes-dependent QoL score	−1.54 ± 0.73	−1.87 ± 0.71	−1.68 ± 0.69	0.053
19 domain-specific items				
Leisure activities	−1.54 ± 1.43	−1.81 ± 1.73	−2.15 ± 2.05	0.203
Working life	−1.52 ± 1.42	−1.68 ± 1.39	−1.96 ± 2.25	0.623
Journeys	−1.60 ± 1.52	−1.89 ± 1.88	−1.80 ± 2.03	0.669
Holidays	−1.66 ± 1.58	−2.07 ± 1.64	−2.44 ± 2.30	0.1351
Physical health	−2.38 ± 1.81	−2.45 ± 1.73	−2.29 ± 2.11	0.928
Family life	−3.14 ± 2.17	−3.96 ± 2.56	−3.38 ± 2.30	0.192
Friendship and social life	−1.31 ± 1.76	−1.38 ± 1.76	−1.37 ± 1.96	0.974
Close personal relationship	−2.39 ± 1.98	−2.66 ± 1.55	−3.53 ± 2.64	0.043*
Sex life	−1.50 ± 1.72	−1.69 ± 1.42	−2.61 ± 2.38	0.042*
Physical appearance	−1.91 ± 1.95	−1.80 ± 1.67	−2.00 ± 2.04	0.890
Self-confidence	−1.75 ± 2.13	−1.21 ± 1.53	−1.46 ± 1.98	0.337
Motivation	−1.74 ± 1.83	−1.17 ± 1.12	−1.44 ± 1.45	0.157
People's reaction	−0.89 ± 1.34	−0.62 ± 1.03	−0.73 ± 1.03	0.462
Feeling about future	−3.48 ± 2.40	−3.60 ± 2.25	−3.02 ± 2.34	0.485
Financial situation	−0.85 ± 1.36	−0.74 ± 1.21	−0.61 ± 1.55	0.688
Living conditions	−0.66 ± 1.03	−0.50 ± 0.94	−0.56 ± 1.53	0.763
Dependence on others	−1.48 ± 1.87	−1.26 ± 1.36	−1.77 ± 1.64	0.374
Freedom to eat	−4.46 ± 2.68	−4.89 ± 2.67	−5.05 ± 2.97	0.518
Freedom to drink	−4.32 ± 2.98	−4.55 ± 2.90	−4.98 ± 3.20	0.556
AWI	−2.05 ± 1.08	−2.10 ± 0.90	−2.23 ± 1.27	0.716

Value is displayed as mean ± standard deviation. ADDQoL, Audit of Diabetes-Dependent Quality of Life; OAD, oral antidiabetic drug; QoL, quality of life; AWI, Average weighted impact score; *$p < 0.05$.

TABLE 4 C-SOADAS in different oral antidiabetic drug groups.

Item	2 OADs (*n* =65)	3 OADs (*n* =47)	>3 OADs (*n* =41)	*p* value
Q1: Ability to control blood sugar	4.44 ± 0.56	4.09 ± 0.75	3.93 ± 0.93	0.001
Q2: Effect of weight	3.98 ± 0.67	3.91 ± 0.50	3.73 ± 0.67	0.127
Q3: Tolerability of the side effect	4.43 ± 0.50	4.25 ± 0.49	4.10 ± 0.58	0.006
Q4: Convenience of drug taking	4.11 ± 0.59	3.96 ± 0.66	3.34 ± 0.85	<0.0001
Q5: Overall satisfaction	4.45 ± 0.53	4.21 ± 0.46	3.90 ± 0.62	<0.0001
Total C-SOADAS score	21.42 ± 1.98	20.43 ± 2.09	19.00 ± 2.24	<0.0001

Value is displayed as mean ± standard deviation. C-SOADAS, Chinese version of the Satisfaction with Oral Antidiabetic drugs; OADs, oral antidiabetic drugs; Q, question.

showed that the use of one or two and three or more OADs had no significant effect on treatment satisfaction compared with diet or exercise control, whereas the combination of OADs and insulin had significantly lower treatment satisfaction. However, a very limited number of studies have evaluated the treatment satisfaction of patients using various OAD classes. The study findings showed that the number of OAD classes was significantly associated with treatment satisfaction, and the treatment satisfaction scores decreased as the number of OADs increased. These results are consistent with those of previous studies showing that the use of more drugs may increase the complexity of the dosing regimen and may lead to other adverse drug reactions, such as weight gain and increased incidence of severe hypoglycemia, to achieve intensive glycemic control. Thus, poor adherence leads to worse treatment satisfaction (26). An analysis of the UK study on drug attribute preference in T2DM showed that the most important factors determining patients' preference for OAD were the likelihood of hypoglycemic events, weight change, the likelihood of gastrointestinal side effects or nausea, especially for patients taking two or more drugs, and drug efficacy (27).

The AWI score reflects the overall impact of diabetes on a person's life. The study results indicated that the mean weighted impact score had a low correlation with the overall (current) QOL score ($R_s = 0.148$, $p = 0.0682$) and, as expected, a moderate correlation with the diabetes impact score on specific aspects of life ($R_s = 0.350$, $p < 0.0001$). These results are similar to the results of the ADDQoL-CnTW validation study (12). The correlation between the AWI score and the diabetes-specific impact score ($R_s = 0.52$, $p < 0.01$) was better than the overall QOL score ($R_s = 0.07$, $p > 0.05$), which is consistent with the findings of previous similar studies, which showed that the diabetes-specific QOL psychometric instrument is more sensitive to individual changes. The European PANORAMA study (25), a cross-sectional investigation of 5,817 individuals, showed a higher correlation between AWI scores and diabetes impact scores on specific life aspects than overall QOL scores ($R_s = 0.21$, $p < 0.001$ and $R_s = 0.60$, $p < 0.001$). The 2016 Hong Kong study (11) suggests that the AWI score of the ADDQoL is less relevant to general generic instruments such as the SF-36 or EQ-5D for assessing a broad range of health states. A moderate-to-high correlation was observed between the mean weighted impact scores of the ADDQoL and the four factors of its instrument

($R_s = 0.490–0.916$, $p < 0.0001$), which is consistent with the results of the ADDQoL-CnTW validation study ($R_s = 0.390–0.82$) (12). Furthermore, a significant moderate-to-high correlation was observed between the mean weighted impact scores of the ADDQoL and 19 specific life domains in a Polish study ($R_s = 0.42–0.80$, $p < 0.001$) (28). Therefore, it can be concluded that the ADDQoL can be used as a standard tool to measure diabetes-related QOL across ethnic groups, especially in relation to the impact of specific life domains of diabetes. Strengthening the appreciation of risk factors associated with HRQoL or treatment satisfaction has become an important program in diabetes healthcare. Assessing the association between treatment satisfaction and HRQoL may help healthcare providers identify patients' perceptions of their disease, predict various aspects of the life of individuals with diabetes, and identify diabetes management that needs to be reinforced to improve treatment outcomes. However, few studies have evaluated the relationship between HRQoL and treatment satisfaction. To the best of our knowledge, this is the first study to evaluate the correlation between ADDQoL and C-SOADAS in patients with T2DM. This study showed a low positive correlation between the two, which is consistent with earlier findings. Although the questionnaires used in this study were inconsistent with previous studies in that they did not yield a significant correlation between HRQoL and treatment satisfaction, both the most commonly used general QOL measure (29) and the Diabetes-Related QOL Questionnaire (30) provided the same confirmation that perceptions of treatment satisfaction and perceptions and descriptions of burdens or limitations in QOL disagree, which suggests that treatment satisfaction and HRQoL are two distinct phenomena. Therefore, QOL and treatment satisfaction should be assessed concurrently in the comprehensive care of patients with diabetes.

Strengths and limitations

To the best of our knowledge, this is the first study to investigate the QOL and treatment satisfaction of patients with T2DM receiving OADs in Taiwan. The first advantage of this study is that the majority of outpatients with diabetes in Taiwan use only OADs for blood glucose control, and this is increasing significantly. However, few outcome measures have been reported for diabetes-related patients using OADs alone. Second, this study used a validated, standardized instrument to assess diabetes-related QOL and treatment satisfaction. However, this study has several limitations.

First, the total sample size was relatively small. Patients with T2DM were recruited from a single medical center in southern Taiwan. Although the characteristics of the study population were similar to those of the Taiwan Annals of Diabetes data, it was not possible to address the issue of treatment patterns affecting the results and limit the generalizability of the study. Second, this cross-sectional study was inconclusive in establishing a causal relationship between sociodemographic and clinical characteristics, HRQoL, and treatment satisfaction, and assessing differences in changes in QOL at different time points was not feasible. However, previous studies have suggested that SGLT2 inhibitors can be anti-frailty drugs (31). Although 43% of our study population used SGLT2 inhibitors, we are uncertain about the effect of hypoglycemic drugs on QOL. Third, while respondents were encouraged to answer honestly to ensure that no relevant findings were affected, it may not be possible to completely avoid response bias in social expectations, especially for treatment satisfaction surveys, which is a common limitation of survey research. Fourth, previous studies showed a positive relationship between treatment satisfaction and HRQoL and medication adherence, especially in chronic diseases such as diabetes and hypertension (32). In this study, medication adherence was not explored, so bias in producing good glycemic control could not be avoided.

Conclusion

This study evaluated the HRQoL and treatment satisfaction of patients with T2DM undergoing metformin-based treatment in combination with other OADs using the ADDQoL and C-SOADAS questionnaire in Taiwan. The results showed a significantly greater positive effect of fewer OAD classes on treatment satisfaction. Further studies with a larger sample size are needed to provide clinical healthcare providers with a more comprehensive understanding of the QOL and treatment satisfaction of patients with T2DM in Taiwan and worldwide.

Data availability statement

The data analyzed in this study are subject to the following licenses/restrictions: First author had full access to all the data in the study and took responsibility for the integrity of the data and the accuracy of the data analysis. Data will be available with appropriate request to first author (wun0910@gmail.com) or corresponding author by e-mail (jk2975525@hotmail.com). Requests to access these datasets should be directed to Y-WC, wun0910@gmail.com, and C-YC, jk2975525@hotmail.com.

Ethics statement

This study was approved by the Institutional Review Board of the Chang Gung Medical Foundation (no. 202000024B0C501). All participants provided written informed consent. The patients/participants provided their written informed consent to participate in this study.

Author contributions

Y-WC, F-CS, and C-YC were involved in the conception of the study, analysis, provided data, clinical information, and study design. All authors were involved in the interpretation of the findings, drafting the manuscript, reviewed, and approved the final manuscript and agreed to be held accountable for all aspects of the work.

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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.

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Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fpubh.2023.1152284/full#supplementary-material

References

1. Saeedi P, Petersohn I, Salpea P, Malanda B, Karuranga S, Unwin N, et al. Global and regional diabetes prevalence estimates for 2019 and projections for 2030 and 2045: results from the international diabetes federation diabetes atlas, 9(th) edition. *Diabetes Res Clin Pract*. (2019) 157:107843. doi: 10.1016/j.diabres.2019.107843

2. Atkinson MJ, Kumar R, Cappelleri JC, Hass SL. Hierarchical construct validity of the treatment satisfaction questionnaire for medication (TSQM version II) among outpatient pharmacy consumers. *Value Health*. (2005) 8:S9–S24. doi: 10.1111/j.1524-4733.2005.00066.x

3. Nicolucci A, Cucinotta D, Squatrito S, Lapolla A, Musacchio N, Leotta S, et al. Clinical and socio-economic correlates of quality of life and treatment satisfaction in patients with type 2 diabetes. *Nutr Metab Cardiovasc Dis*. (2009) 19:45–53. doi: 10.1016/j.numecd.2007.12.005

4. Bradley C, de Pablos-Velasco P, Parhofer KG, Eschwège E, Gönder-Frederick L, Simon D. PANORAMA: a European study to evaluate quality of life and treatment satisfaction in patients with type-2 diabetes mellitus—study design. *Prim Care Diabetes*. (2011) 5:231–9. doi: 10.1016/j.pcd.2011.04.004

5. Williams SA, Pollack MF, Dibonaventura M. Effects of hypoglycemia on health-related quality of life, treatment satisfaction and healthcare resource utilization in patients with type 2 diabetes mellitus. *Diabetes Res Clin Pract*. (2011) 91:363–70. doi: 10.1016/j.diabres.2010.12.027

6. Biderman A, Noff E, Harris SB, Friedman N, Levy A. Treatment satisfaction of diabetic patients: what are the contributing factors? *Fam Pract*. (2009) 26:102–8. doi: 10.1093/fampra/cmp007

7. Sundaram M, Kavookjian J, Patrick JH, Miller LA, Suresh Madhavan S, Scott V. Quality of life, health status and clinical outcomes in type 2 diabetes patients. *Qual Life Res*. (2007) 16:165–77. doi: 10.1007/s11136-006-9105-0

8. Khunti K, Seidu S, Kunutsor S, Davies M. Association between adherence to pharmacotherapy and outcomes in type 2 diabetes: a meta-analysis. *Diabetes Care*. (2017) 40:1588–96. doi: 10.2337/dc16-1925

9. Brod M, Cobden D, Lammert M, Bushnell D, Raskin P. Examining correlates of treatment satisfaction for injectable insulin in type 2 diabetes: lessons learned from a clinical trial comparing biphasic and basal analogues. *Health Qual Life Outcomes*. (2007) 5:8. doi: 10.1186/1477-7525-5-8

10. Chu CH, Hsu CC, Lin SY, Chuang LM, Liu JS, Tu ST. Trends in antidiabetic medical treatment from 2005 to 2014 in Taiwan. *J Formos Med Assoc*. (2019) 118:S74–82. doi: 10.1016/j.jfma.2019.06.001

11. Fung CS, Wan EY, Yu CL, Wong CKH, et al. Validity and reliability of the 19-item audit of diabetes-dependent quality of life (ADDQoL-19) questionnaire in Chinese patients with type 2 diabetes mellitus in primary care. *Qual Life Res*. (2016) 25:2373–8. doi: 10.1007/s11136-016-1263-0

12. Wang HF, Bradley C, Chang TJ, Chuang LM, Yeh MC. Assessing the impact of diabetes on quality of life: validation of the Chinese version of the 19-item audit of diabetes-dependent quality of life for Taiwan. *Int J Qual Health Care*. (2017) 29:335–42. doi: 10.1093/intqhc/mzx028

13. Lin YJ, Wang CY, Chang EH, Cheng SW, Ko Y. Translation, revision, and validation of the Chinese version of the satisfaction with Oral anti-diabetic agent scale (C-SOADAS) in patients with type 2 diabetes mellitus. *Patient Prefer Adherence*. (2018) 12:667–72. doi: 10.2147/PPA.S162268

14. Daniel WW, Cross CL. *Biostatistics: A Foundation for Analysis in the Health Science 11/e AE*. New York: John Wiley & Sons (2019).

15. Turner RC, Cull CA, Frighi V, Holman RR. Glycemic control with diet, sulfonylurea, metformin, or insulin in patients with type 2 diabetes mellitus: progressive requirement for multiple therapies (UKPDS 49). UK prospective diabetes study (UKPDS) group. *JAMA*. (1999) 281:2005–12. doi: 10.1001/jama.281.21.2005

16. Willey CJ, Andrade SE, Cohen J, Fuller JC, Gurwitz JH. Polypharmacy with oral antidiabetic agents: an indicator of poor glycemic control. *Am J Manag Care*. (2006) 12:435–40.

17. Singla R, Bindra J, Singla A, Gupta Y, Kalra S. Drug prescription patterns and cost analysis of diabetes therapy in India: audit of an endocrine practice. *Indian J Endocrinol Metab*. (2019) 23:40–5. doi: 10.4103/ijem.IJEM_646_18

18. Pansini A, Lombardi A, Morgante M, Frullone S, Marro A, Rizzo M, et al. Hyperglycemia and physical impairment in frail hypertensive older adults. *Front Endocrinol*. (2022) 13:831556. doi: 10.3389/fendo.2022.831556

19. Dipnall JF, Pasco JA, Meyer D, Berk M, Williams LJ, Dodd S, et al. The association between dietary patterns, diabetes and depression. *J Affect Disord*. (2015) 174:215–24. doi: 10.1016/j.jad.2014.11.030

20. Papazafiropoulou AK, Bakomitrou F, Trikallinou A, Ganotopoulou A, Verras C, Christofilidis G, et al. Diabetes-dependent quality of life (ADDQOL) and affecting factors in patients with diabetes mellitus type 2 in Greece. *BMC Res Notes*. (2015) 8:786. doi: 10.1186/s13104-015-1782-8

21. Bradley C, Lewis KS. Measures of psychological well-being and treatment satisfaction developed from the responses of people with tablet-treated diabetes. *Diabet Med*. (1990) 7:445–51. doi: 10.1111/j.1464-5491.1990.tb01421.x

22. Bener A, Al-Hamaq AO, Yousafzai MT, Abdul-Ghani M, et al. Relationship between patient satisfactions with diabetes care and treatment. *Niger J Clin Pract*. (2014) 17:218–25. doi: 10.4103/1119-3077.127562

23. Chaturvedi R, Desai C, Patel P, Shah A, Dikshit RK. An evaluation of the impact of antidiabetic medication on treatment satisfaction and quality of life in patients of diabetes mellitus. *Perspect Clin Res*. (2018) 9:15–22. doi: 10.4103/picr.PICR_140_16

24. Naegeli AN, Hayes RP. Expectations about and experiences with insulin therapy contribute to diabetes treatment satisfaction in insulin-naïve patients with type 2 diabetes. *Int J Clin Pract*. (2010) 64:908–16. doi: 10.1111/j.1742-1241.2010.02363.x

25. Bradley C, Eschwege E, de Pablos-Velasco P, Parhofer KG, Simon D, Vandenberghe H, et al. Predictors of quality of life and other patient-reported outcomes in the PANORAMA multinational study of people with type 2 diabetes. *Diabetes Care*. (2018) 41:267–76. doi: 10.2337/dc16-2655

26. Baumgartner A, Drame K, Geutjens S, Airaksinen M. Does the Polypill improve patient adherence compared to its individual formulations? A Systematic Review. *Pharmaceutics*. (2020) 12:190. doi: 10.3390/pharmaceutics12020190

27. Gelhorn HL, Stringer SM, Brooks A, Thompson C, Monz BU, Boye KS, et al. Preferences for medication attributes among patients with type 2 diabetes mellitus in the UK. *Diabetes Obes Metab*. (2013) 15:802–9. doi: 10.1111/dom.12091

28. Bak E, Marcisz C, Nowak-Kapusta Z, Dobrzyn-Matusiak D, Marcisz E, Krzeminska S. Psychometric properties of the audit of diabetes-dependent quality of life (ADDQoL) in a population-based sample of polish adults with type 1 and 2 diabetes. *Health Qual Life Outcomes*. (2018) 16:53. doi: 10.1186/s12955-018-0878-y

29. Nicolucci A, Giorgino R, Cucinotta D, Zoppini G, Muggeo M, Squatrito S, et al. Validation of the Italian version of the WHO-well-being questionnaire (WHO-WBQ) and the WHO-diabetes treatment satisfaction questionnaire (WHO-DTSQ). *Diabetes Nutr Metab*. (2004) 17:235–43.

30. Gurkova E, Cap J, Ziakova K. Quality of life and treatment satisfaction in the context of diabetes self-management education. *Int J Nurs Pract*. (2009) 15:91–8. doi: 10.1111/j.1440-172X.2009.01733.x

31. Mone P, Varzideh F, Jankauskas SS, Pansini A, Lombardi A, Frullone S, et al. SGLT2 inhibition via Empagliflozin improves endothelial function and reduces mitochondrial oxidative stress: insights from frail hypertensive and diabetic patients. *Hypertension*. (2022) 79:1633–43. doi: 10.1161/HYPERTENSIONAHA.122.19586

32. Chiolero A, Burnier M, Santschi V. Improving treatment satisfaction to increase adherence. *J Hum Hypertens*. (2016) 30:295–6. doi: 10.1038/jhh.2015.89

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Gender-specific effects of oxidative balance score on the prevalence of diabetes in the US population from NHANES

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Background: The relationship between oxidative balance score (OBS) and diabetes remains poorly understood and may be gender-specific. We conducted a cross-sectional study to investigate the complex association between OBS and diabetes among US adults.

Methods: Overall, 5,233 participants were included in this cross-sectional study. The exposure variable was OBS, composed of scores for 20 dietary and lifestyle factors. Multivariable logistic regression, subgroup analysis, and restricted cubic spline (RCS) regression were applied to examine the relationship between OBS and diabetes.

Results: Compared to the lowest OBS quartile group (Q1), the multivariable-adjusted odds ratio (OR) (95% confidence interval (CI) for the highest OBS quartile group (Q4) was 0.602 (0.372–0.974) (p for trend = 0.007), and for the highest lifestyle, the OBS quartile group was 0.386 (0.223–0.667) (p for trend < 0.001). Moreover, gender effects were found between OBS and diabetes (p for interaction = 0.044). RCS showed an inverted-U relationship between OBS and diabetes in women (p for non-linear = 6e–04) and a linear relationship between OBS and diabetes in men.

Conclusions: In summary, high OBS was negatively associated with diabetes risk in a gender-dependent manner.

KEYWORDS

oxidative balance score, gender, diabetes, cross-sectional study, adults

1 Introduction

The spread of Western lifestyles has gradually increased the use of high-calorie and high-fat diets, sedentary lifestyles, and the number of adults with diabetes (1). According to the International Diabetes Federation 2019 report, the overall prevalence of diabetes among adults aged 20 to 79 years was 9.3%. This is expected to rise to 693 million worldwide by 2045 (2–4). Diabetes and its complications are life-threatening problems.

A large body of evidence demonstrated that oxidative stress plays a crucial role in the development and progression of diabetes (5). The oxidative balance score (OBS) is a comprehensive indicator containing 20 different dietary and lifestyle components, which highlights the overall balance of pro- and antioxidants at dietary and lifestyle levels. In general, a higher OBS indicates that antioxidants prevail over pro-oxidants. Numerous studies have reported a negative correlation between OBS and the incidence of different diseases, such as breast cancer (6), new-onset hypertension (7), osteoporosis (8), and leukocyte telomere length (9). However, the potential relationship between OBS and diabetes risk remains to be known.

Herein, we investigated the relationship between OBS and the prevalence of diabetes. We examined the possible effects of OBS on diabetes using data from the National Health and Nutrition Examination Survey (NHANES) from 2007 to March 2020.

2 Method

2.1 Source of data and study population

The NHANES was a national cross-sectional study assessing the health and nutrition status of adults and children in the US population. The study used a "stratified multistage probability sampling," in which the information was collected from relevant interviews, examinations, dietary questionnaires, and laboratory measurements. In total, 5,233 participants were chosen from 2007 to March 2020. Exclusion criteria were as follows: age of participants was <20 or ≥80 years, participants without dietary or lifestyle data, participants without known diabetes status, and variables with missing values (Figure 1). All participants provided signed written informed consent, and the study conformed to ethical standards.

2.2 Calculation of the oxidative balance score

The development and calculation of the OBS have been reported previously (9). The total OBS components were assigned a score by intake, property, and gender. Overall scores were the sum of dietary and lifestyle scores. Higher OBS was positively correlated with participants' antioxidant activity. Dietary components and lifestyle components were used to calculate OBS. Dietary components of OBS consisted of dietary fiber, carotene, riboflavin, niacin, calcium, magnesium, zinc, total folate, vitamins (B_6, B_{12}, C, and E), copper, selenium, total fat, and iron. The lifestyle components of OBS consisted of physical activity, alcohol drinking, body mass index, and cotinine. Fat, iron, alcohol drinking, cotinine, and body mass index were classified as pro-oxidants, and the remaining components were classified as antioxidants.

2.3 Evaluation of diabetes

The diagnostic criteria for diabetes were as follows: previous diagnosis of diabetes by a physician, glycated hemoglobin (HbA1c) >6.5%, fasting glucose ≥7.0 mmol/L, random blood glucose ≥11.1 mmol/L, 2-h oral glucose tolerance test (OGTT) ≥11.1 mmol/L, and use of diabetes medication or insulin.

2.4 Covariates

Based on the existing literature and clinical consideration, we selected covariates that could play roles as potential confounders in the associations between OBS and diabetes. The standardized household interviews were used to obtain the demographic characteristics, including age, gender, race, educational level, and poverty income ratio (PIR). Age was divided into three groups (20–39, 40–59, and 60–79 years), with 20–39 years as the reference. Race was divided into non-Hispanic white, non-Hispanic black, Mexican American, and others, with non-Hispanic black as the reference. Education level was graded into primary school or less, middle and high school, and college or higher, with college or higher as the reference. Poverty was defined as PIR ≤ 1.0 and divided into two categories of PIR (≤1.0 and >1.0), with PIR ≤ 1.0 as the reference. White blood cell (WBC) count, platelet (Plt) count, neutrophil (Neu) count, lymphocyte (Lym) count, and hemoglobin (Hb) level were obtained from the laboratory data. Chronic kidney disease (CKD), cardiovascular disease (CVD), hypertension, dyslipidemia, and smoking are important risk factors for diabetes. Therefore, these diseases were included in the analysis. According to the KDIGO 2021 Clinical Practice Guideline for the Management of Glomerular Diseases, albumin-to-creatinine ratio (ACR) and

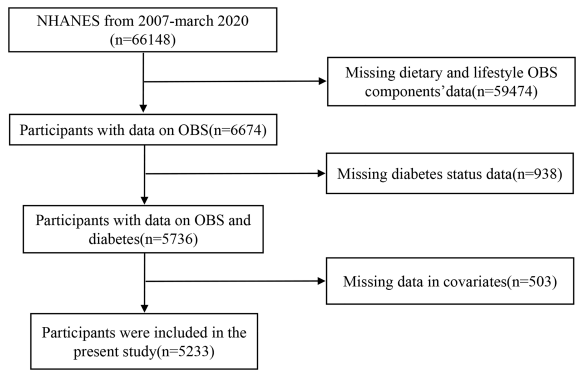

FIGURE 1
Flow diagram.

estimated glomerular filtration rate (eGFR) were used to define CKD. ACR ≥ 30 mg/g (3 mg/mmol) and eGFR < 60 ml/min/1.73 m^2 were defined as diagnostic criteria of CKD. CVD was defined as congestive heart failure, coronary heart disease, heart attack, angina, and stroke. The diagnostic criteria for hypertension were as follows: average systolic blood pressure ≥140 mmHg or average diastolic blood pressure ≥90 mmHg after at least three times of measurement, use of anti-hypertensive drugs, and subject- or physician-reported diagnosis of hypertension. The diagnostic criteria for dyslipidemia were as follows: total cholesterol level ≥5.18 mmol/L, triglyceride level ≥150 mg/dl, high-density lipoprotein-cholesterol <1.04 mmol/L in men and <1.30 mmol/L in women, low-density lipoprotein-cholesterol ≥3.37 mmol/L, or the use of cholesterol-lowering drugs. Smoking status was defined into three categories: never (smoked less than 100 cigarettes in life), former (smoked more than 100 cigarettes in life and does not smoke now), and smoked more than 100 cigarettes in life and smokes some days or every day. No or never was taken as the reference for all of the above conditions.

2.5 Statistical analysis

Considering the complexity of the sampling method, the study subject was a weighted statistical analysis. Continuous variables are presented as mean (standard deviation (SD)), and categorical variables are summarized as frequency (percentage). For baseline characteristics, categorical variables were compared using the chi-square test, and continuous variables were compared using the t-test or one-way analysis of variance. Multivariable logistic regression models (crude models to model 3) were used to investigate the relationship between OBS and diabetes after adjusting for different potential confounders. The crude model was not adjusted for any covariates. Model 1 was adjusted for age, gender, race, and education. Model 2 was further adjusted for WBC count, Neu count, Hb count, and Plt count. Model 3 was adjusted for the variables in model 2 and additional confounders, including CKD, CVD, smoking status, hypertension, and dyslipidemia. We further assessed the heterogeneity between OBS and diabetes through subgroup analysis for the following variables: age groups, gender, race, education, CKD, CVD, hypertension, dyslipidemia, and smoking status. We applied restricted cubic spline (RCS) analysis with four knots to evaluate the non-linear associations between OBS and diabetes risk. R statistical software (version 4.2.2) was applied for all statistical analyses and mapping. Alpha was set at <0.05 for statistical significance, and all analyses were two-sided. A two-sided p-value <0.05 was defined as the significance threshold.

3 Results

3.1 Baseline characteristics

In total, 5,233 participants from NHANES (2007 to March 2020) were enrolled in the present study, of whom 622 (11.89%) had diabetes. Among all participants with diabetes, men (461, 11.64%) showed a higher prevalence than women (161, 5.61%). Participants with diabetes were older and had lower education levels, but PIR did not differ between the two groups (Table 1). The comparison of baseline characteristics showed that patients with diabetes had a higher prevalence of CKD, CVD, hypertension, and dyslipidemia. Participants without diabetes showed significantly higher OBS and lifestyle OBS when compared with participants with diabetes. There was no significant difference in Lym count ($p = 0.7$) and dietary OBS ($p = 0.06$).

All 5,233 individuals were categorized into four groups according to OBS quartiles: Q1 (OBS, 5 to 28; median, 26), Q2 (OBS, 29 to 30; median, 30), Q3 (OBS, 31 to 32; median, 31), and Q4 (OBS, 32 to 36; median, 33). As the reference group, participants in the first quartile group (Q1) with lower OBS were more likely to be white and have higher educational levels and PIR. In addition, participants in the Q1 had a lower incidence of CKD, diabetes, hypertension, dyslipidemia, and smoking. Gender was not significantly different between OBS quartiles, suggesting an evenly balanced distribution of men and women in all quartiles (Table 2). We also performed the same analysis on dietary OBS and lifestyle OBS. Dietary OBS did not significantly affect the prevalence of diabetes, CKD, CVD, hypertension, and dyslipidemia. Increased lifestyle OBS was associated with the prevalence of diabetes, CKD, CVD, hypertension, and dyslipidemia. Similar to OBS, lifestyle OBS was associated with the prevalence of diabetes (Supplementary Tables 1, 2).

3.2 Relationship between OBS and diabetes

The results of the multivariable logistic regressions showed that OBS was significantly associated with diabetes (Table 3). In model 3, the risk of diabetes decreased by 3.8% with a 1-unit increase in OBS (OR = 0.962, 95% confidence interval (CI) 0.935–0.990), revealing that OBS was negatively correlated with the risk of diabetes. Compared with participants in Q1, those in Q2, Q3, and Q4 were at lower risk of diabetes in all models. Compared with participants in Q1, subjects in Q4 had 39.8% (OR = 0.602; 95% CI, 0.378–0.974) decreased risk of diabetes in model 3. We further explored the effects of dietary OBS and lifestyle OBS on diabetes using logistic regression models. Although dietary OBS and lifestyle OBS were supposed to be protective factors for diabetes, results for dietary OBS were not statistically significant (Supplementary Tables 3, 4).

3.3 Subgroup analysis

Subgroup analysis was performed based on gender, age group, race, education, CVD, CKD, dyslipidemia, hypertension, and smoking status (Table 4). Statistical significance was found in gender subgroups of diabetes (p for interaction = 0.044). Subgroup analysis showed that the female subgroup was more sensitive to OBS compared with the male subgroup. Women had an 88.0% lower risk of developing diabetes in the fourth quartile of age compared with the first quartile. In contrast, the risk of diabetes only decreased by 38.6% in men (all p for trend <0.05).

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TABLE 1 Baseline characteristics of all participants by diabetes.

Variables	Overall (n = 5,233)	Non-DM (n = 4,611)	DM (n = 622)	*p*-Value
Age (years)	45.23 (0.41)	44.23 (0.42)	55.11 (0.78)	<0.0001
Gender, n (%)				<0.0001
Female	2,062 (39.4)	1,901 (94.39)	161 (5.61)	
Male	3,171 (60.6)	2,710 (88.36)	461 (11.64)	
Age group, n (%)				<0.0001
20–39	2,103 (40.19)	2,023 (97.12)	80 (2.88)	
40–59	1,951 (37.28)	1,675 (89.39)	276 (10.61)	
60–79	1,179 (22.53)	913 (81.29)	266 (18.71)	
Education, n (%)				<0.001
College and higher	3,495 (66.79)	3,136 (91.79)	359 (8.21)	
Middle and high school	1,535 (29.33)	1,314 (88.92)	221 (11.08)	
Primary school and less	203 (3.88)	161 (79.08)	42 (20.92)	
Race, n (%)				0.01
Black	1,029 (19.66)	868 (87.89)	161 (12.11)	
Mexican	623 (11.91)	520 (86.97)	103 (13.03)	
Other	989 (18.9)	873 (90.43)	116 (9.57)	
White	2,592 (49.53)	2,350 (91.57)	242 (8.43)	
PIR				0.65
≤1	812 (15.52)	719 (90.23)	93 (9.77)	
>1	4,421 (84.48)	3,892 (90.94)	529 (9.06)	
WBC ($\times10^9$/L)	7.01 (0.05)	6.98 (0.05)	7.36 (0.14)	0.01
Neu ($\times10^9$/L)	4.12 (0.04)	4.08 (0.04)	4.45 (0.10)	<0.001
Lym ($\times10^9$/L)	2.10 (0.01)	2.11 (0.01)	2.08 (0.07)	0.7
Hb (g/L)	14.52 (0.03)	14.50 (0.03)	14.69 (0.08)	0.03
Plt ($\times10^6$/L)	237.68 (1.57)	238.57 (1.61)	228.78 (4.45)	0.03
CKD, n (%)				<0.0001
No	4,693 (89.68)	4,246 (92.36)	447 (7.64)	
Yes	540 (10.32)	365 (75.37)	175 (24.63)	
CVD, n (%)				<0.0001
No	4,910 (93.83)	4,390 (91.86)	520 (8.14)	
Yes	323 (6.17)	221 (73.09)	102 (26.91)	
Hypertension, n (%)				<0.0001
No	3,485 (66.6)	3,273 (95.35)	212 (4.65)	
Yes	1,748 (33.4)	1,338 (80.82)	410 (19.18)	
Dyslipidemia, n (%)				<0.0001
No	1841,(35.18)	1,747 (96.01)	94 (3.99)	
Yes	3,392 (64.82)	2,864 (88.28)	528 (11.72)	
Smoking status, n (%)				<0.001

(Continued)

TABLE 1 Continued

Variables	Overall (n = 5,233)	Non-DM (n = 4,611)	DM (n = 622)	*p*-Value
Never	2,570 (49.11)	2,327 (92.49)	243 (7.51)	
Former	1,364 (26.07)	1,139 (87.38)	225 (12.62)	
Now	1,299 (24.82)	1,145 (91.65)	154 (8.35)	
OBS	29.69 (0.08)	29.76 (0.08)	28.99 (0.20)	<0.001
Dietary OBS	25.55 (0.06)	25.58 (0.07)	25.28 (0.16)	0.06
Lifestyle OBS	4.13 (0.04)	4.17 (0.04)	3.71 (0.09)	<0.0001

All values represented are weighted means (standard deviation) or counts (weighted percentage).
SD, standard deviation; PIR, poverty income ratio; WBC, white blood cells; Neu, neutrophil; Lym, lymphocyte; Hb, hemoglobin; Plt, platelet; CKD, chronic kidney disease; CVD, cardiovascular disease; OBS, oxidative balance score; DM, diabetes.

TABLE 2 Baseline characteristics of all participants by the OBS quartile.

Variables	Overall (n = 5,233)	Q1 (n = 1,608)	Q2 (n = 1,310)	Q3 (n = 1,422)	Q4 (n = 893)	*p*-value
Age (years)	45.23 (0.41)	43.32 (0.60)	44.62 (0.64)	46.13 (0.60)	47.31 (0.78)	<0.001
Gender, n (%)						0.1
Female	2,062 (39.4)	604 (25.23)	494 (23.55)	577 (30.41)	387 (20.81)	
Male	3,171 (60.6)	1,004 (27.72)	816 (26.18)	845 (28.22)	506 (17.88)	
Age group, n (%)						<0.001
20–39	2,103 (40.19)	681 (29.60)	537 (25.45)	545 (27.61)	340 (17.34)	
40–59	1,951 (37.28)	597 (26.93)	515 (25.96)	538 (29.31)	301 (17.80)	
60–79	1,179 (22.53)	330 (20.14)	258 (22.41)	339 (31.87)	252 (25.57)	
Education, n (%)						<0.0001
College and higher	3,495 (66.79)	854 (21.20)	827 (23.76)	1,054 (31.39)	760 (23.64)	
Middle and high school	1,535 (29.33)	677 (42.07)	425 (28.62)	319 (22.96)	114 (6.35)	
Primary school and less	203 (3.88)	77 (40.38)	58 (31.00)	49 (20.89)	19 (7.73)	
Race, n (%)						<0.0001
Black	1,029 (19.66)	499 (49.20)	269 (26.55)	192 (17.63)	69 (6.61)	
Mexican	623 (11.91)	177 (28.60)	189 (29.51)	182 (30.09)	75 (11.81)	
Other	989 (18.9)	247 (25.12)	214 (23.42)	285 (28.86)	243 (22.60)	
White	2,592 (49.53)	685 (24.32)	638 (24.81)	763 (30.34)	506 (20.53)	
PIR						<0.0001
≤1	812 (15.52)	377 (44.18)	197 (23.27)	167 (21.67)	71 (10.89)	
>1	4,421 (84.48)	1,231 (24.87)	1,113 (25.27)	1,255 (29.90)	822 (19.95)	
WBC ($\times10^9$/L)	7.01 (0.05)	7.51 (0.10)	7.21 (0.07)	6.81 (0.07)	6.37 (0.09)	<0.0001
Neu ($\times10^9$/L)	4.12 (0.04)	4.46 (0.07)	4.24 (0.05)	3.97 (0.05)	3.71 (0.07)	<0.0001
Lym ($\times10^9$/L)	2.10 (0.01)	2.21 (0.03)	2.16 (0.03)	2.07 (0.02)	1.93 (0.03)	<0.0001
Hb (g/L)	14.52 (0.03)	14.63 (0.06)	14.58 (0.06)	14.47 (0.05)	14.35 (0.06)	0.002
Plt ($\times10^6$/L)	237.68 (1.57)	243.98 (2.24)	237.34 (2.58)	237.81 (2.56)	229.12 (3.06)	0.002
DM, n (%)						0.004

(Continued)

TABLE 2 Continued

Variables	Overall (n = 5,233)	Q1 (n = 1,608)	Q2 (n = 1,310)	Q3 (n = 1,422)	Q4 (n = 893)	*p*-value
No	4,611 (88.11)	1,360 (25.79)	1,156 (24.98)	1,269 (29.61)	826 (19.62)	
Yes	622 (11.89)	248 (35.61)	154 (26.12)	153 (24.37)	67 (13.89)	
CKD, n (%)						0.003
No	4,693 (89.68)	1,399 (25.90)	1,190 (24.99)	1,297 (29.85)	807 (19.26)	
Yes	540 (10.32)	209 (34.87)	120 (26.07)	125 (21.63)	86 (17.43)	
CVD, n (%)						0.65
No	4,910 (93.83)	1,485 (26.66)	1,227 (24.94)	1,348 (29.08)	850 (19.32)	
Yes	323 (6.17)	123 (27.07)	83 (27.62)	74 (30.06)	43 (15.25)	
Hypertension, n (%)						<0.0001
No	3,485 (66.6)	960 (24.48)	861 (24.30)	962 (29.26)	702 (21.95)	
Yes	1,748 (33.4)	648 (31.62)	449 (26.84)	460 (28.85)	191 (12.70)	
Dyslipidemia, n (%)						<0.001
No	1,841 (35.18)	527 (23.98)	440 (23.82)	499 (28.70)	375 (23.50)	
Yes	3,392 (64.82)	1,081 (28.05)	870 (25.72)	923 (29.35)	518 (16.88)	
Smoking status, n (%)						<0.0001
Never	2,570 (49.11)	573 (18.40)	590 (21.97)	802 (34.28)	605 (25.35)	
Former	1,364 (26.07)	345 (22.86)	345 (26.36)	407 (29.35)	267 (21.43)	
Now	1,299 (24.82)	690 (50.65)	375 (30.62)	213 (17.01)	21 (1.72)	
OBS	29.69 (0.08)	25.10 (0.13)	29.55 (0.02)	31.50 (0.02)	33.50 (0.03)	<0.0001
Dietary OBS	25.55 (0.06)	22.13 (0.15)	25.89 (0.05)	26.97 (0.04)	27.73 (0.03)	<0.0001
Lifestyle OBS	4.13 (0.04)	2.96 (0.05)	3.66 (0.05)	4.53 (0.04)	5.77 (0.03)	<0.0001

All values represented are weighted means (standard deviation) or counts (weighted percentage). The OBS was divided into four levels by quartile (5 < Q1 ≤ 28, 29 < Q2 ≤ 30, 31 < Q3 ≤ 32, and 32 < Q4 ≤36).
WBC, white blood cells; Neu, neutrophil; Lym, lymphocyte; Hb, hemoglobin; Plt, platelet; CKD, chronic kidney disease; CVD, cardiovascular disease; OBS, oxidative balance score; DM, diabetes.

3.4 RCS analysis

The associations between OBS and diabetes in men and women were further evaluated using the RCS curves and the multivariable logistic regression (model 3). First, we found an inverted-U relationship (p for non-linear = 0.0118) between OBS and the risk of diabetes (Figure 2A). The turning point appeared around the OBS of 25.20, and the median number was 30.00. The risk of diabetes slightly increased at the beginning and then declined with the OBS after reaching the turning point. Results of the RCS analysis by gender revealed that OBS

TABLE 3 Association of the OBS with diabetes, NHANES 2007–March 2020.

Diabetes	OR (95% CI); *p*-value							
	Crude model		Model 1		Model 2		Model 3	
Continuous	0.95 (0.93, 0.97)	<0.0001	0.94 (0.92, 0.97)	<0.0001	0.95 (0.92, 0.98)	<0.001	0.96 (0.94, 0.99)	<0.009
Q1	1.00 (ref)		1.00 (ref)		1.00 (ref)		1.00 (ref)	
Q2	0.76 (0.56, 1.02)	0.07	0.71 (0.52, 0.98)	0.04	0.73 (0.53, 1.00)	0.05	0.76 (0.55, 1.06)	0.10
Q3	0.60 (0.42, 0.85)	0.005	0.55 (0.38, 0.78)	0.001	0.58 (0.40, 0.83)	0.003	0.62 (0.43, 0.90)	0.01
Q4	0.51 (0.33, 0.79)	0.003	0.44 (0.28, 0.69)	<0.001	0.48 (0.30, 0.76)	0.002	0.60 (0.37, 0.97)	0.04
p for trend		<0.001		<0.0001		<0.001		0.007

The OBS was converted from a continuous variable to a categorical variable (quartiles). Data are presented as OR (95% CI). Crude model was adjusted with no covariates.
OBS, oxidative balance score; NHANES, National Health and Nutrition Examination Survey.

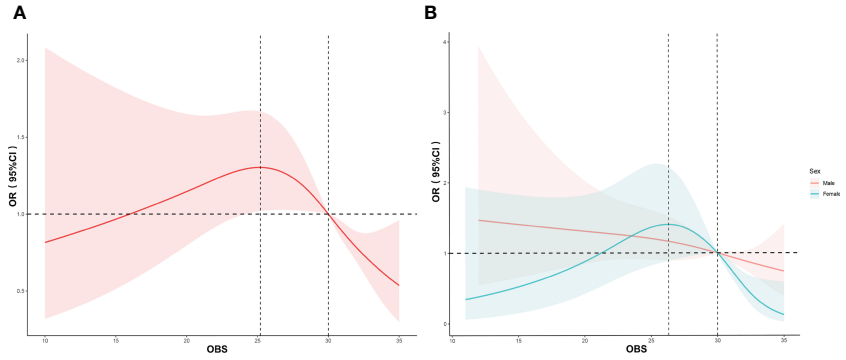

FIGURE 2
RCS analysis of the association between OBS and diabetes. The association was adjusted for age, gender, race, education, WBC count, Neu count, Hb level, Plt count, CKD, CVD, smoking status, hypertension, and dyslipidemia. The median OBS was chosen as the reference. **(A)** RCS curve of the association between OBS and diabetes among all participants. **(B)** RCS curve of the association between OBS and diabetes among female and male participants. RCS, restricted cubic spline; WBC, white blood cells; Plt, platelet; Neu, neutrophil; Lym, lymphocyte; Hb, hemoglobin; OR, odds ratio; CI, confidence interval.

was negatively correlated with the incidence of diabetes in male individuals, and it showed a linear relationship (p for non-linear = 0.7816). Consistent with the relationship between the overall OBS and diabetes, we found a non-linear inverted-U relationship (p for non-linear < 0.001) between OBS and diabetes in female individuals (Figure 2B). The turning point appeared around the OBS of 26.32, and the median number was 30.00. After the OBS of 30 was reached, the risk of diabetes decreased with the increase of OBS, and the decrease in diabetes risk was more pronounced in women than in men. It was also consistent with the results of the subgroup analysis.

4 Discussion

For the first time, our large-scale cross-sectional study evaluated the association of OBS with diabetes based on NHANES (from 2007 to March 2020). In this study, we confirmed that the OBS and lifestyle OBS in participants without diabetes were significantly higher than those in participants with diabetes. Higher OBS and lifestyle OBS were associated with decreased risk of diabetes. After confounding factors were adjusted, it was shown that the effect of OBS on diabetes significantly relied on gender. In women and all participants, the association between OBS and diabetes showed an inverted-U relationship. In men, there was a linear relationship between OBS and the risk of diabetes.

Numerous clinical and animal studies linked oxidative stress to diabetes incidence and progression. Oxidative stress occurs when the amount of reactive oxygen species (ROS) exceeds the neutralizing capacity of antioxidants (10). Oxidative stress can interfere with the oxidation–reduction reactions in glycolysis and the electron transport chain, causing hyperglycemia. Oxidative stress activates the secondary pathways of glucose metabolism, such as glucose autoxidation and the polyol pathway, which leads to excessive ROS production and lipid peroxidation, triggering oxidative stress and exacerbating hyperglycemia (5, 10–12). Previous studies have not evaluated the relationship between OBS and diabetes, and most of them focused on the relationship between a single component of OBS and diabetes risk. Oxidative stress is a major contributor to the progression of diabetes, but not all strategies for reducing ROS are protective against diabetes (13–15). Several reasons may account for this difference. First, it is not easy to determine the effects of individual oxidative stress-related components on blood glucose control. Second, pro-oxidants or antioxidants may have antagonistic and synergistic interactions. Special attention should also be paid to the properties of the antioxidants themselves. Antioxidants can exert a pro-oxidant effect in high doses, have poor solubility and low permeability in biomembranes, and lack stability and specificity of action (16). Hence, we employed OBS as a comprehensive evaluation metric to measure the oxidative balance in an individual and investigate its influence on diabetes. Our study demonstrated that OBS was significantly higher in the non-diabetic group than in the diabetic group, and higher OBS predicted a lower risk of diabetes. RCS analysis showed that the relationship between OBS and diabetes mellitus was an inverted-U relationship. Our findings are consistent with previous epidemiological evidence indicating a significant association between oxidative stress and diabetes (5, 10, 17).

Similar to previous studies, subgroup analysis and RCS analysis revealed that gender significantly affected the correlations between OBS and diabetes. Studies have shown that the serum glucose level and oxidative stress of female diabetic rats were lower than those of male diabetic rats. At the same time, female diabetic rats had lower hydrogen peroxide levels and xanthine oxidase activity (18). Estrogen has antioxidant properties and can enhance the activity of antioxidants (19, 20). On the contrary, androgen can promote ROS production and induce oxidative stress (21). Studies have shown that estrogen, as a protective factor, can reduce the risk of insulin resistance and diabetes in women, while androgen has the opposite role (22, 23). It is worth noting that the risk of diabetes decreased more rapidly in women than in men when OBS was greater than 30. It can be concluded that it is more efficient in combating oxidative stress to keep OBS within a certain range, resulting in better glycemic control in patients with diabetes. Our results may provide new policies to reduce the burden of diabetes complications and improve the management of diabetes. Several factors such as diet, lifestyle, and genetic factors can lower

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TABLE 4 Subgroup analyses of the association between OBS and diabetes, NHANES 2007–March 2020.

Variables	Q1	Q2	Q3	Q4	*p* for trend	*p* for interaction
Age group						0.211
60–79	Ref	1.063 (0.637, 1.776)	1.031 (0.579, 1.833)	0.821 (0.480, 1.403)	0.456	
20–39	Ref	0.574 (0.290, 1.138)	0.334 (0.153, 0.726)	0.212 (0.067, 0.672)	0.001	
40–59	Ref	0.639 (0.416, 0.982)	0.423 (0.261, 0.684)	0.345 (0.142, 0.838)	0.001	
Gender						0.044
Male	Ref	0.838 (0.570, 1.232)	0.592 (0.410, 0.855)	0.614 (0.363, 1.041)	0.016	
Female	Ref	0.456 (0.214, 0.970)	0.468 (0.246, 0.891)	0.120 (0.036, 0.401)	<0.0001	
Race						0.832
White	Ref	0.723 (0.460, 1.136)	0.552 (0.326, 0.933)	0.520 (0.290, 0.933)	0.009	
Mexican	Ref	1.008 (0.478, 2.123)	0.665 (0.298, 1.485)	0.617 (0.152, 2.499)	0.26	
Black	Ref	0.835 (0.483, 1.446)	0.719 (0.381, 1.357)	0.186 (0.063, 0.550)	0.017	
Other	Ref	0.545 (0.236, 1.256)	0.603 (0.242, 1.502)	0.305 (0.129, 0.721)	0.029	
Education						0.187
College and higher	Ref	0.835 (0.551, 1.264)	0.696 (0.432, 1.118)	0.519 (0.304, 0.887)	0.009	
Middle and high school	Ref	0.667 (0.387, 1.149)	0.419 (0.207, 0.848)	0.626 (0.220, 1.782)	0.05	
Primary school and less	Ref	0.192 (0.044, 0.841)	0.091 (0.024, 0.345)	0.074 (0.011, 0.501)	<0.001	
CKD						0.069
No	Ref	0.598 (0.420, 0.852)	0.516 (0.344, 0.774)	0.470 (0.289, 0.764)	<0.001	
Yes	Ref	1.309 (0.599, 2.859)	0.776 (0.371, 1.624)	0.312 (0.121, 0.804)	0.011	
CVD						0.268
No	Ref	0.727 (0.526, 1.005)	0.508 (0.339, 0.760)	0.393 (0.242, 0.638)	<0.0001	
Yes	Ref	0.543 (0.201, 1.462)	0.693 (0.320, 1.500)	0.958 (0.253, 3.627)	0.872	
Smoking status						0.472
Never	Ref	0.534 (0.283, 1.007)	0.415 (0.236, 0.729)	0.353 (0.182, 0.685)	0.001	
Former	Ref	0.947 (0.532, 1.686)	0.736 (0.439, 1.236)	0.493 (0.254, 0.955)	0.015	
Now	Ref	0.621 (0.334, 1.154)	0.285 (0.147, 0.551)	0.203 (0.070, 0.590)	<0.001	
Hypertension						0.121
No	Ref	0.436 (0.256, 0.744)	0.538 (0.298, 0.971)	0.470 (0.229, 0.962)	0.038	
Yes	Ref	1.022 (0.694, 1.506)	0.611 (0.417, 0.894)	0.601 (0.334, 1.082)	0.01	
Dyslipidemia						0.239
Yes	Ref	0.675 (0.480, 0.949)	0.572 (0.382, 0.858)	0.531 (0.328, 0.860)	0.002	
No	Ref	0.944 (0.412, 2.163)	0.427 (0.160, 1.136)	0.188 (0.073, 0.486)	<0.001	

Data are presented as OR (95% CI). Adjusted for age, gender, race, education, WBC, Neu, Hb, Plt, CKD, CVD, smoking status, hypertension, and dyslipidemia.
WBC, white blood cells; Neu, neutrophil; Hb, hemoglobin; Plt, platelet; CKD, chronic kidney disease; CVD, cardiovascular disease; OBS, oxidative balance score.

the risk of chronic diseases by regulating oxidation and reducing ROS generation (24). Recently, researchers suggested that the risk of chronic disease can be reduced through lifestyle interventions (25). The risk of diabetes can be significantly reduced by lifestyle modifications, especially in male subjects (26). The mechanisms underlying the gender-specific associations merit further investigation (27). As reported in diabetic patients, female patients have lower overall muscle mass and physical function, poorer health conditions, and higher prevalence of depression, which indicate that gender-specific differences in diabetes deserve more attention (28). Thus, the lower overall muscle mass in women with diabetes may be related to OBS, especially lifestyle OBS. A study found that decreased estrogen levels in postmenopausal women increased insulin resistance and elevated the risk of diabetes (29).

The study has several advantages. First, our study found the association between OBS and diabetes for the first time and uncovered the gender-specific effects of OBS on the prevalence of diabetes. Second, the NHANES used a stratified, multistage sampling method, which increases the generalizability of our findings to non-institutionalized populations. Third, this study adjusted the results for several confounders. In addition, there are several limitations to this study. Even though we controlled for potential confounders, the role of unknown or unmeasured confounders cannot be ruled out. However, the cross-sectional nature of our study makes it difficult to infer causality. To increase the utility of our findings, the predictive value of OBS in diabetes needs to be further verified through prospective studies. Finally, dietary OBS was not significantly different between diabetic and non-diabetic groups in our study; therefore, the effects of dietary OBS in predicting diabetes risk remain unclear.

5 Conclusion

In conclusion, this cross-sectional study indicated that OBS, especially lifestyle OBS, was negatively associated with the prevalence of diabetes. OBS had an inverted-U relationship with the prevalence of diabetes in nationally representative adults of the USA. In addition, we found that the negative correlation between OBS and diabetes was clearer among female participants than in male participants.

Data availability statement

Publicly available datasets were analyzed in this study. This data can be found here: https://www.cdc.gov/nchs/nhanes/index.htm.

Ethics statement

The studies involving human participants were reviewed and approved by National Center for Health Statistics Ethics Review Board Approval. The patients/participants provided their written informed consent to participate in this study.

Author contributions

CW and CR conceptualized this study; performed the literature search, study design, data curation, data analysis, and data interpretation; and drafted the original manuscript. YS participated in the study design and critically revised the manuscript. YS, HG, XP and LZ conceived the study and participated in study design, coordination, data collection, and analysis. All authors contributed to the article and approved the submitted version.

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Conflict of interest

The authors declare that this study was conducted in the absence of any commercial or financial relationships that could serve 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/fendo.2023.1148417/full#supplementary-material

References

1. Cunha DA, Cito M, Carlsson PO, Vanderwinden JM, Molkentin JD, Bugliani M, et al. Thrombospondin 1 protects pancreatic β-cells from lipotoxicity *via* the PERK-NRF_2 pathway. *Cell Death Differ* (2016) 23(12):1995–2006. doi: 10.1038/cdd.2016.89

2. Cho NH, Shaw JE, Karuranga S, Huang Y, da Rocha Fernandes JD, Ohlrogge AW, et al. IDF diabetes atlas: global estimates of diabetes prevalence for 2017 and projections for 2045. *Diabetes Res Clin Pract* (2018) 138:271–81. doi: 10.1016/j.diabres.2018.02.023

3. Climie RE, van Sloten TT, Périer MC, Tafflet M, Fayosse A, Dugravot A, et al. Change in cardiovascular health and incident type 2 diabetes and impaired fasting glucose: the Whitehall II study. *Diabetes Care* (2019) 42(10):1981–7. doi: 10.2337/dc19-0379

4. Kato S, Ando M, Honda H, Yoshida Y, Imaizumi T, Yamamoto N, et al. Effectiveness of lifestyle intervention using the Internet of things system for individuals with early type 2 diabetes mellitus. *Intern* (2020) 59(1):45–53. doi: 10.2169/internalmedicine.3150-19

5. Tangvarasittichai S. Oxidative stress, insulin resistance, dyslipidemia and type 2 diabetes mellitus. *World J Diabetes* (2015) 6(3):456–80. doi: 10.4239/wjd.v6.i3.456

6. Sohouli MH, Baniasadi M, Hernández-Ruiz Á, Melekoglu E, Zendehdel M, José Soto-Méndez M, et al. Adherence to oxidative balance scores is associated with a reduced risk of breast cancer; a case-control study. *Nutr Cancer* (2023) 75(1):164–73. doi: 10.1080/01635581.2022.2102658

7. Lee JH, Son DH, Kwon YJ. Association between oxidative balance score and new-onset hypertension in adults: a community-based prospective cohort study. *Front Nutr* (2022) 9:1066159. doi: 10.3389/fnut.2022.1066159

8. Shahriarpour Z, Nasrabadi B, Hejri-Zarifi S, Shariati-Bafghi SE, Yousefian-Sanny M, Karamati M, et al. Oxidative balance score and risk of osteoporosis among postmenopausal Iranian women. *Arch Osteoporos* (2021) 16(1):43. doi: 10.1007/s11657-021-00886-w

9. Zhang W, Peng SF, Chen L, Chen HM, Cheng XE, Tang YH. Association between the oxidative balance score and telomere length from the national health and nutrition examination survey 1999-2002. *Oxid Med Cell Longev* (2022) 2022:1345071. doi: 10.1155/2022/1345071

10. Opara EC. Role of oxidative stress in the ediology of type 2 diabetes and the effect of antioxidant supplementation on glycemic control. *J Invest Med* (2004) 52 (1):19–23. doi: 10.1136/jim-52-01-22

11. Cutiongco MFA, Chua BMX, Neo DJH, Rizwan M, Yim EKF. Functional differences between healthy and diabetic endothelial cells on topographical cues. *Biomaterials* (2018) 153:70–84. doi: 10.1016/j.biomaterials.2017.10.037

12. Dariya B, Nagaraju GP. Advanced glycation end products in diabetes, cancer and phytochemical therapy. *Drug Discov Today* (2020) 25(9):1614–23. doi: 10.1016/j.drudis.2020.07.003

13. Grundy SM, Vega GL, McGovern ME, Tulloch BR, Kendall DM, Fitz-Patrick D, et al. Efficacy, safety, and tolerability of once-daily niacin for the treatment of dyslipidemia associated with type 2 diabetes: results of the assessment of diabetes control and evaluation of the efficacy of niaspan trial. *Arch Intern Med* (2002) 162:1568–76. doi: 10.1001/archinte.162.14.1568

14. Stranges S, Marshall JR, Natarajan R, Donahue RP, Trevisan M, Combs GF, et al. Effects of long-term selenium supplementation on the incidence of type 2 diabetes: a randomized trial. *Ann Intern Med* (2007) 147:217–23. doi: 10.7326/0003-4819-147-4-200708210-00175

15. Song Y, Cook NR, Albert CM, Van Denburgh M, Manson JE. Effects of vitamins c and e and beta-carotene on the risk of type 2 diabetes in women at high risk of cardiovascular disease: a randomized controlled trial. *Am J Clin Nutr* (2009) 90(2):429–37. doi: 10.3945/ajcn.2009.27491

16. Nasri H, Shirzad H, Baradaran A, Rafieian-Kopaei M. Antioxidant plants and diabetes mellitus. *J Res Med Sci* (2015) 20(5):491–502. doi: 10.4103/1735-1995.163977

17. Rehman K, Akash MSH. Mechanism of generation of oxidative stress and pathophysiology of type 2 diabetes mellitus: how are they interlinked? *J Cell Biochem* (2017) 118(11):3577–85. doi: 10.1002/jcb.26097

18. Díaz A, López-Grueso R, Gambini J, Monleón D, Mas-Bargues C, Abdelaziz KM, et al. Sex differences in age-associated type 2 diabetes in rats-role of estrogens and oxidative stress. *Oxid Med Cell Longev* (2019) 2019:6734836. doi: 10.1155/2019/6734836

19. Badeau M, Adlercreutz H, Kaihovaara P, Tikkanen MJ. Estrogen a-ring structure and antioxidative effect on lipoproteins. *J Steroid Biochem Mol Biol* (2005) 96:271–8. doi: 10.1016/j.jsbmb.2005.04.034

20. Chainy GBN, Sahoo DK. Hormones and oxidative stress: an overview. *Free Radic Res* (2020) 54(1):1–26. doi: 10.1080/10715762.2019.1702656

21. Fernando SM, Rao P, Niel L, Chatterjee D, Stagljar M, Monks DA. Myocyte androgen receptors increase metabolic rate and improve body composition by reducing fat mass. *Endocrinology* (2010) 151(7):3125–32. doi: 10.1210/en.2010-0018

22. Greenhill C. Obesity: sex differences in insulin resistance. *Nat Rev Endocrinol* (2018) 14:65. doi: 10.1038/nrendo.2017.168

23. Kautzky-Willer A, Harreiter J, Pacini G. Sex and gender differences in risk, pathophysiology and complications of type 2 diabetes mellitus. *Endocr Rev* (2016) 37 (3):278–316. doi: 10.1210/er.2015-1137

24. Hermsdorff HHM, Barbosa KB, Volp ACP, Puchau B, Bressan J, Zulet M Á, et al. Gender-specific relationships between plasma oxidized low-density lipoprotein cholesterol, totalantioxidant capacity, and central adiposity indicators. *Prev Cardiol* (2012) 21(7):884–91. doi: 10.1177/2047487312472420

25. Wang Y, Han X, Yang J. Revisiting the blended learning literature: using a complex adaptive systems framework. *Educ Technol Soc* (2015) 18(2):380–93.

26. Harreiter J, Kautzky-Willer A. Sex and gender differences in prevention of type 2 Diabetes.Front. *Endocrinol (Lausanne)* (2018) 9:220. doi: 10.3389/fendo.2018.00220

27. Feitosa MF, Reiner AP, Wojczynski MK, Graff M, North KE, Carr JJ, et al. Borecki. sex-influenced association of nonalcoholic fatty liver disease with coronary heart disease. *Atherosclerosis.* (2013) 227(2):420–4. doi: 10.1016/j

28. Merchant RA, Soong JTY, Morley JE. Gender differences in body composition in pre-frail older adults with diabetes mellitus. *Front Endocrinol (Lausanne)* (2022) 13:795594. doi: 10.3389/fendo.2022.795594

29. Yang K, Liu W. Triglyceride and glucose index and sex differences in relation to major adverse cardiovascular events in hypertensive patients without diabetes. *Front Endocrinol* (2021) 12: doi: 10.3389/fendo

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Cyclocarya paliurus leaves extracts alleviate metabolic phenotypes in Chinese T2DM patients by modulating gut microbiota and metabolites: a clinical randomized controlled trial

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Objective: We aimed to investigate the effect of Cyclocarya paliurus leaves extracts (CP) on glucose and blood lipid metabolism and its relationship with intestinal flora in type 2 diabetes mellitus (T2DM) patients.

Methods: In this open-label, 84-day randomized controlled trial, a total of 38 T2DM patients were randomly assigned to the CP group or the Glipizide group (G group) in a 2:1 ratio. T2DM-associated metabolic phenotypes, gut microbiota and metabolites including short-chain fatty acids (SCFAs) and bile acids (BAs) were detected.

Results: At the end of intervention, CP, like Glipizide, significantly improved HbA1c level and other glucose metabolism parameters (fasting plasma glucose (FBG), 2-hour post-meal blood glucose (2hPBG), the area under curve of oral glucose tolerance test glucose (OGTT glucose AUC)). Moreover, CP also resulted in the significant improvement in the levels of blood lipid and blood pressure. Notably, the improvement in blood lipid(triglycerides (TG) and high-density lipoprotein cholesterol (HDL-c)) and blood pressure (diastolic blood pressure (DBP)) was significantly greater in the CP group compared with the G group. Furthermore, the liver and kidney function parameters did not significantly change in both CP group and the G group over the 84-day period. Additionally, the enrichment of potentially beneficial bacteria (Faecalibacterium and Akkermansia), SCFAs and unconjugated BAs and the depletion of potential pathogenic bacteria (Prevotella_9) and conjugated BAs were observed in the CP group, while the abundances of the gut microbial were kept stable in the G group after intervention.

Conclusion: CP displays a more beneficial effect in the alleviation of T2DM-associated metabolic phenotypes than glipizide by regulating gut microbiota and metabolites in T2DM patients, with no significant effects on liver and kidney function.

KEYWORDS

Cyclocarya paliurus leaves extracts (CP), type 2 diabetes mellitus (T2DM), randomized controlled trial (RCT), metabolic phenotypes, gut microbiota

Introduction

Type 2 diabetes mellitus (T2DM), whose prevalence has been estimated to raise from 536.6 million people in 2021 to 783.2 million by 2045 (1), causes lots of linked complications, such as kidney failure, heart disease, amputation, and blindness (2), and a heavy burden on public health. At present, the two main methods to control T2DM are lifestyle changes and drug therapy (3). Although there are multiple anti-diabetic medications, it is still difficult to control the progression of T2DM. Additionally, anti-diabetic medications have many unwanted side effects such as lactic acidosis and hypoglycemia, and ultimately fail to control blood glucose levels (4). Therefore, it is necessary to find out safer and more effective antidiabetic medications for T2DM.

Cyclocarya paliurus (C. paliurus) (Batal) IIjinskajia, an endemic plant belonging to Cyclocarya genus of the Juglandaceae family, is also known as the money tree, and it is distributed in 420–2500 m mountainous regions of Jiangxi, Hubei, Hunan, Guizhou and other provinces in China (5). Studies have shown that the leaves of C. paliurus contain a variety of bioactive compounds, such as polysaccharides, flavonoids, triterpenoids and phenolic compounds (6, 7). It has been reported that C. paliurus can reduce the levels of blood glucose, blood lipid and blood pressure in rats (8, 9). Based on its characteristics, in 1999, the United States Food and Drug Administration (FDA) approved the leaves of C. paliurus as a dietary supplement product and it is the first Chinese herbal tea approved by the institution (10). Therefore, C. paliurus has attracted more and more attention, and several studies found that polysaccharides and polyphenols (flavonoids, tannins, phenolic acids, and anthocyanins) isolated from C. paliurus leaves exert a potent hypoglycemic effect to ameliorate high-fat diet-low dose streptozotocin (STZ)-induced experimental T2DM in mice (11, 12). However, the mechanism of the effect of bioactive constituents of C. paliurus leaves on glucose and blood lipid metabolism improvement is still unclear.

Gut microbiota plays an important role in T2DM. Moderate dysbiosis with a decrease in butyric-producing bacteria and an increase in opportunistic pathogens was found in T2DM patients in China (13). Recently, accumulating evidence demonstrated that C. paliurus could impact T2DM through gut microbiota and its metabolites. Li et al. found that polysaccharide from C. paliurus leaves attenuated diabetes through multi-path of gut microbiota and host metabolism in T2DM rats (14). Yao et al. revealed that C. paliurus alleviate T2DM symptoms by modulating gut microbiota and short-chain fatty acids (SCFAs) in T2DM rats (15).

As above mentioned, C. paliurus seems to have the potential to be a new treatment medication for T2DM. However, to our knowledge, no existing literature or research is available to address the question concerning whether C. paliurus can improve glucose and lipid metabolism in T2DM patients. Thus, we conducted a pilot clinical randomized controlled study to compare the effect of C. paliurus leaves extracts (CP) and an antidiabetic drug Glipizide on metabolic phenotypes, including the intestinal microbiota and their metabolites (SCFAs and bile acids) in T2DM patients.

Method

Study design and subjects

A randomized and open-label, controlled clinical trial was designed and conducted according to Consolidated Standards of Reporting Trials guidelines (16). Participants were all recruited from the community healthcare center at West Renmin Road, Yanquan Road and Xiameiqiao Road in Chenzhou, Hunan, China from January 2019 to December 2020. All participants with T2DM signed written informed consentforms before enrollments. The trial protocol was registered at Chinese Clinical Trial Registry (http://www.chictr.org.cn/) and got a registration number of ChiCTR1900020482. Chinese newly onset, treatment-naive T2D patients from Han nationality with the age of 35- to 70-year old were enrolled in this trial. The inclusion criteria were as follows: (1) fasting plasma glucose (FBG) ≥7 mmol/L or/and 2-h oral glucose tolerance test ≥11.1 mmol/L; (2) HbA1c ≥6.5% (6.5%≦HbA1c≦12%) (17, 18). The exclusion criteria were listed in Table S1 (Supplementary Material Content 1). The ethics committee of Affiliated Hospital of Xiangnan University approved the study protocol (Ethical approval department No. KY-20181226001).Patients who met all inclusion criteria were randomly assigned to either the C. paliurus leaves extracts group (CP group) or the Glipizide group (G group) in a 2: 1 ratio using a computer-generated random number allocation by a researcher not involved in the study.

Sample sizes calculation

According to Movahed et al., the HbA1c difference between intervention group and control group was expected to be 1.2% at the end of the intervention (19). Combined with a standard deviation of 1.09%, α=0.05 and power=0.80, we calculated that a minimum of 11 T2DM patients were required in each group. Assuming a shedding rate of 10%, each group needed12 T2DM patients at least according to the actual situation. For this clinical trial, a minimum of 36 T2DMpatients were required.

The medication intervention

Before starting the clinical trial, all the enrolled participants underwent a 2-week run-in period and received a diet and exercise education about the daily management of T2DM. Once the 2-week run-in period completed, participants entered the intervention study immediately. In the CP group, the participants received CP powder (2g/time, 3 times/day orally before meals) (CP were provided by Chenzhou Mingrun biological products Co. LTD, Hunan, China) (Figure S1). In the G group (the positive-control group), the participants received Glipizide Controlled-Release Tablets (5mg/time, 2 times/day orally before meals) (Pfizer Pharmaceuticals LLC, hereinafter referred to as Glipizide) that has no significant effect on gut microbiota of T2DM patients (20). During the intervention, all participants were required to monitor FBG, and 2-hour post-meal blood glucose (2hPBG) using Roche glucometer (Accu-Chek Performa) every day.

Preparation of Cyclocarya paliurus leaves extracts and phytochemical analysis

Cyclocarya paliurus leaves extracts (CP)were provided by Chenzhou Ming run biological products Co. LTD Hunan, China. The license number of this product is QS431006011417.The leaves of C. paliurus has long been used as a bitter Traditional Chinese Medicine which has also been historically used as an herbal tea in the folk. Nowadays, the products derived from the leaves of C. paliurus have become a very popular health product in China (11). CP were prepared according to the method described previously with some modifications (21). Briefly, the leaves of Cyclocarya paliurus were dried and pulverized before extraction. Cyclocarya paliurus leaves powders were soaked with distilled water at 90 °C for 1 h (1:30, g/mL), and the extraction solution was centrifuged at 4500 × g for 15 min. The above operation was repeated once. The supernatants (extraction solutions, two times) were combined and concentrated using a rotary evaporator under vacuum. The resulting residue was dissolved with deionized water, filtered through a 800 mesh standard sieve. The elution was performed using 80% ethanol and the effluent of ethanol solution was collected and concentrated, resulting in the CP.

The composition of the C. paliurus leaves extracts was determined by high performance liquid chromatography (HPLC, Agilent 1200, USA) equipped with Agilent 5 TC-C18 (4.6 mm ×250 mm, 5μm). The system parameters were set as follows: injection volume, 5μL; the column temperature, 30°C; and the mobile phase flow rate, 0.8mL/min. The mobile phase consisted of two solvents: (A) 0.2% aqueous acetic acid and (B) acetonitrile. The elution conditions were as follows: 0–5min, gradient 5% B; 5–10 min, linear gradient 5–10% B; 10–15 min, linear gradient 10–25% B; 15–25 min, linear gradient 25–40% B; 25–30min, linear gradient 40–90% B. Peaks were detected at 254 nm. The quantification of different component monomers was based on the peak areas and calculated as equivalents of the standard compounds; all contents were expressed as mg/g component dry weight. The temperature of the column oven was set at 40 °C, the flow rate was set at 0.6 mL/ min and the injection volume was 5 μL.

Clinical assessment

Participants were asked not to make any change to their daily physical activity and to maintain body weight. On Day 0, Day 42, and Day 84, the 7-day recall method was used to measure physical activity energy expenditure through an interview-administered survey instrument modified from the Cross-Cultural Activity Participation Study (22). Metabolic Equivalent Tasks (METs) were conducted to measure the physical activity energy expenditure of all the participants. On Day 0 and Day 84, the medical history and anthropometric evaluation (body weight (BW), waist circumference (WC), waist to hip ratio (WHR), body mass index (BMI), systolic blood pressure (SBP) and diastolic blood pressure (DBP)) were recorded and fasting blood samples and fecal samples were collected. Oral glucose tolerance test(OGTT), insulin release test and C-Peptide release test were also performed. Further details about the physical activity assessment can be found online (Supplementary Methods, Supplementary Material Content 2). Adverse effects were also recorded by the research team.

Clinical outcomes

The primary clinical outcome was a change in the level of HbA1c over 84 days in the two treatment groups. The secondary clinical outcomes included the changes in the levels of FBG, 2hPBG, the area under curve of OGTT blood glucose, fasting insulin (FINS), 2-hour post-meal insulin (2hPINS), the area under curve of insulin release test, fasting C- peptide (FCP), 2-hour post-meal C-Peptide (2hPCP), the area under curve of C-peptide release test, total cholesterol (TC), triglycerides (TG), low-density lipoprotein cholesterol (LDL-c), high-density lipoprotein cholesterol (HDL-c), BW, WC, WHR, BMI, SBP, and DBP.

Gut microbiome, short-chain fatty acids and bile acids analysis

The stool samples were immediately transferred to the -80° Cfreezer for storage after collection on day 0 and day 84. Then the stool samples were used for bacterial 16sRNA gene sequencing, and SCFAs as well as BAs analysis. The detailed information was shown on Supplementary Material Content 2.

Safety assessment

The liver function (the total bilirubin (TBIL), direct bilirubin (DBIL), indirect bilirubin (IBIL), albumin (ALB), alanine aminotransferase (ALT), aspartate aminotransferase (AST)) and renal function (serum creatinine (Scr) and blood urea nitrogen (BUN))of all participants was assessed on day 0 and day 84.

Fecal microbiota transplantation in pseudo-sterile mice

The animal experimental protocol was approved by the animal ethics committee of Jinan University (20190821-02). A total 50 of C57BL/6J mice (weight, 25.00 ± 2g; age, 10 weeks) were purchased from Biotechnology Company Limited (Beijing, China). The mice were raised in the specific pathogen-free animal experiment center at Jinan University, with constant temperature (24 ± 2°C), constant humidity of 55 ± 10% and a 12-h light/dark cycle. All mice received adaptive feeding for 7 days, and then were randomly divided into a control group (NC, n=10) and an antibiotic treatment group (AT, n=40). Within the next 4 weeks, the mice in AT group were fed with antibiotic-containing sterile water and then divided into 4 groups. More detailed information was shown in Supplementary Material Content 2.

Statistical analysis

Outcomes were calculated on a strict intention-to-treat (ITT) basis. The raw data of Day 84 – the raw data of Day 0 represented the changes in all variables. The Kolmogorov-Smirnov Test was used to detect whether the continuous data conform to normal distribution. If normally distributed, paired T-test was used for intra-group comparison and independent T-test was used for inter-group comparison, and the data were expressed as mean and standard deviations. If not normally distributed, non-parametric two-tailed Wilcoxon matched-pairs signed-ranks was used for intra-group comparison and non-parametric K-independent Wilcoxon signed-ranks were used for inter-group comparison, and the data were expressed as the median and inter-quartile ranges. Fold changes of diabetes-induced markers after treatments of CP & G were calculated as (the raw data of day 84/the raw data of day 0). Statistical Package for Social Science (SPSS) 28.0 was used to analyze all data and we defined $P<0.05$ as statistically significant. All data visualization were made by GraphPad Prism (version 9.0). We used the OmicStudio tools at https://www.omicstudio.cn.to make a clustering correlation heatmap.

Results

Phytochemical characterization of CP

Phytochemical compositions of the CP were clarified by using high performance liquid chromatography (HPLC). The 5 components in the CP were identified in the chromatogram with peaks at 17.19 min to3-cafeoylquinic acid, 20.19 min to isoquercitrin, 22.19 min to kaempferol-3-glucoside, 23.43 min to kaempferol 3-rhamnoside, 27.56 min to quercetin by the comparison to external standards (Figure S2). Also, the contents of five compounds in the CP were measured by HPLC and their contents were 53.4, 42.5, 92.6, 4.8 and 2.1 mg/g in the CP, successively (Table S2, Supplementary Material Content 1).

Clinical baseline characteristics and physical activity of participants

According to the inclusion and exclusion criteria, a total of 38 participants with T2DM were randomly divided into the G group (n=13) and CP group (n=25) in the study. During the intervention, 6 participants dropped out (1 participant in the G group and 5 participants in the CP group). Finally, a total of 32 participants completed the intervention (Figure S3). At baseline, anthropometric parameters (BW, WC, WHR, BMI, SBP and DBP), blood glucose homeostasis (HbA1c, FBG, 2hPBG, FINS, 2hPINS, HOMA-IR, Fasting C-Peptide, 2hPCP, OGTT glucose AUC, Insulin release test AUC, C-Peptide release test AUC), blood lipid homeostasis (TG, TC, LDL-c and HDL-c) and other blood markers (TBIL, DBIL, IBIL, ALT, AST, ALB, Scr, BUN) were not statistically different between the two groups (Table S3, Supplementary Material Content 1). In addition, there were no significant differences in physical activity levels in the intra-group and inter-group comparisons on Day 0 and on Day 84 (Table S4, Supplementary Material Content 1).

CP alleviates HbA1c level in T2DM patients

Compared with Day 0, the level of HbA1c, the primary clinical outcome, was significantly decreased both in the G group and in the CP group on Day 84. At the end of intervention, there was no significant difference in the reduction of HbA1c between the two groups (Figure 1A; Table 1). We also calculated the proportion of participants who achieved adequate glycemic control (HbA1c<7%). On Day 0, there were no significant differences in the proportion of participants who achieved HbA1c<7% between the two groups, and both the G group and the CP group had a significant increase in the proportion of participants who achieved HbA1c<7% on Day 84 compared with Day 0. We also found that there was no significant difference in the proportion of participants who achieved HbA1c<7% on Day 84 between the two groups (Figure 1B; Table S5, Supplementary Material Content 1). These results demonstrate that the CP induce a significant reduction in HbA1c, which is similar to the effect of Glipizide on the reduction of HbA1c in T2DM patients.

CP ameliorates several other metabolic phenotypes in T2DM patients

From Day 0 to Day 84, the significant improvement in blood glucose homeostasis (FBG, 2hPBG, OGTT glucose AUC) (Table 1;

Figures 1C, D, 2A), blood lipid homeostasis (TG, TC, LDL-c and HDL-c) (Table 1; Figures 3A–D) and anthropometric parameters (SBP and DBP) (Table 1; Figures S4E, F)was observed, whereas the other blood glucose homeostasis (FINS, 2hPINS, HOMA-IR, fasting C-peptide, 2hPCP, insulin release test AUC and C-peptide release test AUC) (Table 1; Figures 1E–I, 2B, C) and anthropometric parameters (BW, WC, WHR and BMI) (Table 1; Figures S4A–D) were not significantly affected in the CP group. From Day 0 to Day 84, the significant amelioration in blood glucose homeostasis (FBG, 2hPBG, OGTT glucose AUC, FINS, 2hPINS, fasting C-peptide, 2hPCP, insulin release test AUC and C-peptide release test AUC) was found, while the other blood glucose homeostasis (HOMA-IR), blood lipid homeostasis (TG, TC, LDL-c and HDL-c) and anthropometric parameters (BW, WC, WHR and BMI) were not significantly affected in the G group. The fold changes in blood lipid homeostasis (TG and HDL-c) and anthropometric parameters (DBP) was significantly greater in the CP group compared with the G group. Moreover, there were no significant differences in the fold changes in the other blood glucose homeostasis (FBG, 2hPBG, FINS, 2hPINS, fasting C-peptide, 2hPCP, HOMA-IR, OGTT glucose AUC, insulin release test AUC and C-peptide release test AUC), blood lipid homeostasis (TC and LDL-c) and anthropometric parameters (BW, WC, WHR, BMI and SBP) over the 84-days between the two groups. Also, the liver and kidney function parameters (TBIL, DBIL, IBIL, ALT, AST, ALB, Scr and BUN)did not significantly change in both CP group and the G group over the 84-day period (Table 1; Figures 1-3, S4, 5).These results suggest that CP can alleviate T2DM-associated clinical markers, blood lipid and blood pressure in addition to alleviating HbA1c in T2DM patients, having no significant impacts on the liver and kidney function.

CP redresses gut microbiota dysbiosis in T2DM patients

To explore the potential role of the gut microbiota in CP-induced metabolic phenotypes improvement in T2DM patients, 16S rRNA gene sequencing was performed in the fecal samples from the patients on Day 0 and Day 84. At the phylum level, the overall microbial compositions were shown in Table S6 (Supplementary Material Content 1). The dominant species (mean relative abundance >1%) were Firmicutes, Bacteroidetes, Proteobacteria, Actinobacteria, Verrucomicrobia and Fusobacteria (Figure 4A). From Day 0 to Day 84, only the abundance of Verrucomicrobia was significantly increased, whereas there were no significant differences in the abundances of Firmicutes, Bacteroidetes, Proteobacteria, Actinobacteria, Fusobacteria, Synergistetes, Cyanobacteria and Patescibacteria in CP group. In the G group, the abundances of the gut microbial at the phylum level were kept stable during the intervention (Table S6, Supplementary Material Content 1; Figures 4B–F).

At the genus level, the top 10 dominant genera was presented in Figure 5A. Compared with Day 0, the abundances of potentially beneficial bacteria (Faecalibacterium and Akkermansia) were significantly enriched, while the abundance of potentially pathogenic bacteria (Prevotella_9) was significantly reduced on Day 84 in the CP group (Table S7, Supplementary Material Content 1; Figures 5B–D). In addition, the abundances of other genera of bacteria showed no significant change in the CP group during the intervention. In the G group, the abundances of the gut microbiota at the genus level were not significantly affected from Day 0 to Day 84 (Table S7, Supplementary Material Content 1; Figures 5E–G).

Then we conducted a spearman correlation analysis to explore the potential relationships between bacterial abundance and metabolic phenotypes. We found that Faecalibacterium, which was enriched in the participants on Day 84 in the CP group, had a negative correlation with several metabolic phenotypes (HbA1c, FBG, 2hPBG, SBP and TC). Akkermansia, which was also enriched in the participants on Day 84 in the CP group, was negatively related to a few metabolic markers (HbA1c, FBG and TC). Prevotella_9 had a positive relationship with several metabolic markers (FBG, OGTT Glucose AUC, 2hPBG and TC) (Figure 5H). These findings indicate that CP-induced metabolic phenotype alleviation maybe associated with improved intestinal flora.

CP induces metabolites alteration in T2DM patients

To reveal metabolites alteration, related to the gut microbiota, which is potentially involved in CP-induced T2DM improvement, we performed metabolic profiling of feces from the participants. From Day 0 to Day 84, a significant increase was observed in total SCFAs, propionic acid (PA) and butyric acid (BA), while the levels of acetic acid (AA), isobutyric acid (IBA), valeric acid (VA), isovaleric acid (IVA) and hexanoic acid (HA) had no significant change in the CP group. In the G group, the level of total SCFAs, AA, PA, BA, IBA, VA, IVA and HA were not affected over the 84-day period (Table S8, Supplementary Material Content 1; Figures 6A–E).

To explore the potential effect of CP on BA metabolism, a total of 20 species of BAs were detected. Compared to Day 0, the levels of unconjugated BAs (chenodeoxycholic acid (CDCA), 12-ketolithocholic acid (12-KLCA) and β-muricholic acid (β-MCA)) were significantly increased and the levels of conjugated BAs (taurodeoxycholic acid (TDCA),glycochenodeoxycholic acid (GCDCA) were significantly reduced, while no significant differences were found in other BAs in the CP group on Day 84. From Day 0 to Day 84, the levels of BAs showed no significant changes in the G group (Table S9, Supplementary Material Content 1; Figures 7A–F).

Correlation analyses were performed to determine the potential relationships between metabolites and gut microbiota. Coincidently, we observed that Faecalibacterium enriched in the CP group on Day 84 was positively related to SCFAs (total SCFAs, BA and PA) and unconjugated BAs (CDCA, 12-KLCA, β-MCA and LCA), but negatively correlated with conjugated BAs (GCA). Akkermansia, which was also enriched in the CP group on Day 84, showed a positive relationship with SCFAs (total SCFAs, BA,

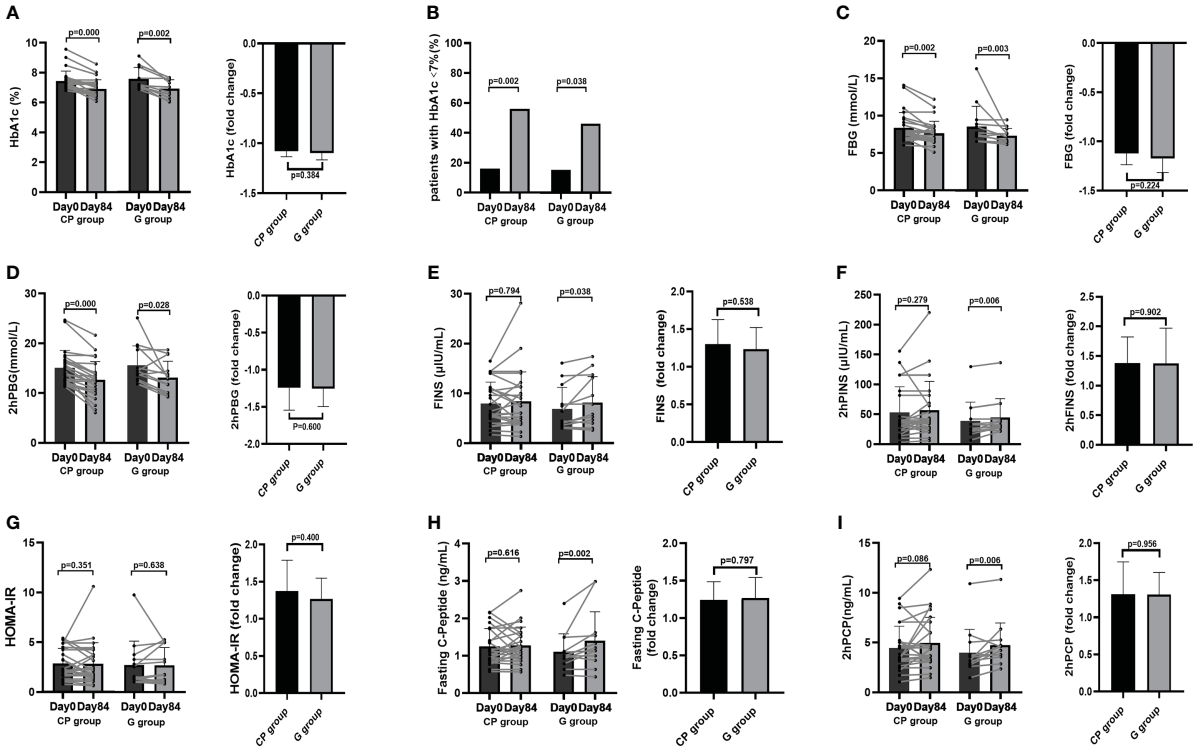

FIGURE 1
The changes in glucose metabolism phenotypes in T2DM patients. **(A)** The change in HbA1c level. **(B)** The percentage of participants with HbA1c<7%. **(C-I)** The changes in the levels of FBG **(C)**, 2hPBG **(D)**, FINS **(E)**, 2hPINS **(F)**, HOMA-IR **(G)**, Fasting C-Peptide **(H)**, 2hPCP **(I)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the fold changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; T2DM, type 2 diabetes mellitus; FBG, fasting blood glucose; FINS, fasting insulin; 2hPBG, 2-hour post-meal blood glucose; 2hPINS, 2-hour post-mealinsulin; HOMA-IR, homeostasis model assessment for insulin resistance = (fasting blood glucosexfasting insulin/22.5), 2hPCP, 2-hour post-meal C-Peptide.

TABLE 1 Comparison of metabolic parameters in T2DM patients.

Outcomes	CP group			G group			*p*
	Day 0 (n=25)	Day 84 (n=25)	*P-value*	Day 0 (n=13)	Day 84 (n=13)	*P-value*	
Blood glucose homeostasis							
HbA1c (%)	7.29 (7.07, 7.63)	6.70 (6.50, 7.28)	0.000	7.14 (7.08, 8.44)	7.01 (6.42, 7.55)	0.002	0.384
FBG (mmol/L)	8.37±2.05	7.64±1.62	0.002	7.80±1.26	6.73±0.98	0.003	0.224
2hPBG (mmol/L)	14.56 (12.47, 16.58)	12.72 (10.27, 14.05)	0.000	14.45 (12.60, 19.00)	12.8 (10.05, 16.08)	0.028	0.600
OGTT glucose AUC (mmol/L/min)	2226.15 (1972.72, 2688.37)	1925.55 (1733.40, 2352.15)	0.000	2258.10 (2026.90, 2827.05)	2063.70 (1838.62, 2317.87)	0.008	0.877
FINS (μIU/mL)	6.35(4.39,10.93)	7.42(4.02,11.22)	0.794	4.46 (3.40, 9.80)	5.82 (3.51, 13.42)	0.038	0.538
2hPINS (μIU/mL)	37.34 (27.02, 85.43)	40.48 (27.33, 75.23)	0.279	26.77 (21.56, 51.74)	32.34 (28.07, 55.25)	0.006	0.902
Insulin release test AUC (μIU/mL/min)	4942.2 (3613.35, 10822.20)	5579.55 (3932.55, 11280.75)	0.145	4249.2 (2807.7, 5489.25)	4915.5 (3871.05, 5895.75)	0.004	0.195
HOMA-IR	2.26 (1.65, 4.45)	2.11 (1.38, 3.97)	0.351	1.46 (1.24, 3.18)	1.96 (1.01, 3.77)	0.638	0.400
Fasting C-Peptide (ng/mL)	1.11 (0.88, 1.73)	1.22 (0.94, 1.56)	0.616	0.99 (0.85, 1.30)	1.21 (0.93, 1.57)	0.002	0.797
2hPCP (ng/mL)	3.82 (3.15, 5.11)	4.47 (3.17, 6.51)	0.086	3.53 (2.83, 4.40)	4.58 (3.67, 4.99)	0.006	0.956

(Continued)

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TABLE 1 Continued

Outcomes	CP group			G group			*p*
	Day 0 (n=25)	Day 84 (n=25)	*P-value*	Day 0 (n=13)	Day 84 (n=13)	*P-value*	
C-Peptide release test AUC (ng/mL/min)	541.80 (419.60, 711.80)	576.6 (462.40, 891.00)	0.064	420.750 (378.45, 634.20)	579.150 (516.00, 640.80)	0.015	0.453
Blood lipid homeostasis							
TG(mmol/L)	2.38±1.51	1.98±1.01	0.014	1.67±0.92	1.80±0.77	0.250	0.014
TC(mmol/L)	5.65±1.35	5.13±1.26	0.001	4.54±089	4.28±0.90	0.064	0.672
LDL-c(mmol/L)	3.40±1.24	2.88±1.15	0.007	2.55±0.71	2.44±0.85	0.231	0.735
HDL-c(mmol/L)	1.17±0.42	1.27±0.33	0.006	1.22±0.32	1.16±0.33	0.523	0.043
Anthropometric markers							
BW (kg)	60.88±7.95	60.66±8.06	0.408	63.1±8.36	62.69±8.19	0.203	0.955
WC (cm)	87.16±5.74	87.03±6.14	0.817	89.88±6.80	88.92±7.14	0.126	0.589
WHR	0.92±0.38	0.93±0.47	0.298	0.94±0.50	0.93±0.51	0.231	0.421
BMI	23.95±2.07	23.82±2.01	0.165	24.38±2.33	24.23±2.34	0.229	0.853
SBP (mmHg)	136.08±19.23	129.80±16.25	0.011	125.84±19.13	121.30±16.73	0.116	0.820
DBP (mmHg)	81.00 (73.00, 88.50)	74.00 (68.50, 82.50)	0.001	67.00 (60.00, 87.50)	73.00 (63.00, 77.00)	0.789	0.028
Liver and renal function							
TBIL (umol/L)	16.34±5.38	17.74±496	0.149	16.82±5.00	15.62±5.02	0.334	0.511
DBIL (umol/L)	4.80 (3.35, 6.05)	4.60(3.45, 5.60)	0.681	5.20 (4.95, 6.45)	5.00 (4.10, 6.90)	0.386	0.557
IBIL (umol/L)	11.00(9.15, 13.00)	9.80 (8.25,11.25)	0.097	9.90 (9.20, 12.35)	10.30 (7.60, 11.20)	0.666	0.566
ALT (U/L)	19.00 (15.00, 27.00)	18.00 (13.50,23.50)	0.092	23.00 (15.00, 25.50)	21.00 (14.50, 28.00)	0.397	0.164
AST (U/L)	19.00 (16.50, 22.50)	20.00 (17.00, 22.50)	0.749	19.00 (17.50, 25.50)	18.00 (17.00, 22.50)	0.180	0.331
ALB (g/L)	43.73±1.91	43.76±2.28	0.347	43.53±2.88	44.54±3.74	0.295	0.123
Scr(umol/L)	70.47±15.51	70.06±14.62	0.721	69.9±13.44	66.96±10.27	0.286	0.136
BUN(mmol/L)	5.15±1.10	5.39±1.14	0.199	4.66±1.03	4.87±1.19	0.465	0.702

The data are shown as the mean ± SD for normal variables or median (IQR) for non-normal variables. Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons. P-value, comparison between Day 0 and Day 84 at the same group. Independent T-test and non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the changes between the two groups. P, comparison of the fold changes between the CP group and the G group.
CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; HbA1c, hemoglobin A1c; FBG, fasting blood glucose; 2hPBG, 2-hour post-meal blood glucose; FINS, fasting insulin; 2hPINS, 2-hour post-meal insulin; HOMA-IR, homeostasis model assessment for insulin resistance = (fasting blood glucose×fasting insulin/22.5); 2hPCP, 2-hour post-meal C-Peptide; OGTT, oral glucose tolerance test; AUC, area under curve; TG, triglycerides; TC, total cholesterol; LDL-c, low-density lipoprotin cholesterol; HDL-c, high-density lipoprotein cholesterol; BW, body weight; WC, waist circumference; WHR, waist to hip ratio; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; TBIL, total bilirubin; DBIL, direct bilirubin; IBIL, indirect bilirubin; ALB, albumin;ALT, alanine aminotransferase; AST, aspartate aminotransferase; Scr, serum creatinine; BUN, blood urea nitrogen.

HA, IBA and PA), and unconjugated BAs (CDCA, 12-KLCA and β-MCA), but had a negative correlation with conjugated BAs (GUCDA, GCA, TDCA and TCDCA). Prevotella_9, which was depleted in the CP group on Day 84, was negatively correlated with SCFAs (total SCFAs, IBA and PA) and unconjugated BAs (CDCA and β-MCA), but positively associated with conjugated BAs (GCA, GLCA and TLCA). Megamonas was negatively correlated with conjugated BAs (GCA and GLCA), but positively associated with SCFAs (HA) and unconjugated BAs (β-MCA and CA) (Figures 6F, 7G).Taken together, the abovementioned results reveal that the changes in gut microbiome abundances induced by CP may be responsible for the alteration of metabolites, thus affecting metabolic phenotypes in patients with T2DM.

FMT ameliorates the glucose metabolism in pseudo-sterile mice

Experimental design for animal study for microbiota transplantation (Figure 8A). After the FMT, we observed that body weight was not also statistically significant both between intragroup and between intergroup (Figure 8B). Mice transplanted with the postintervention microbiota from the CP group showed better glucose metabolic parameters than those with the preintervention microbiota from the CP group, while mice transplanted with the postintervention microbiota from the G group showed no significant change in glucose metabolism parameters compared with the preintervention microbiota from the G group (Figures 8C–E).

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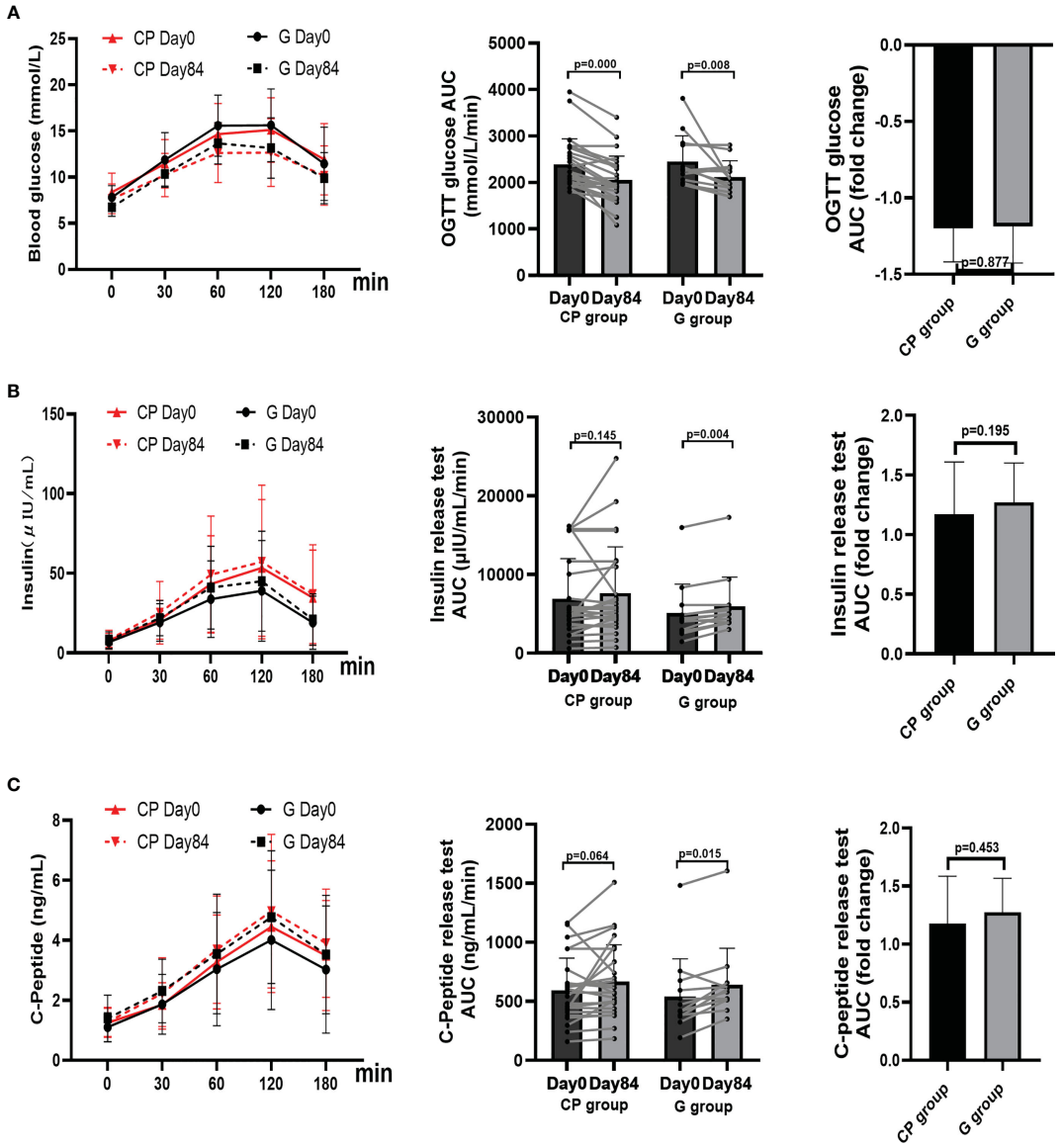

FIGURE 2
The changes in OGTT glucose AUC, insulin release test, and C-Peptide release test in T2DM patients. **(A–C)** The changes in the levels of OGTT glucose AUC **(A)**, insulin release test AUC **(B)**, and C-Peptide release test AUC **(C)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the fold changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; AUC, area under curve.

Discussion

In this pilot clinical randomized trial, we demonstrated for the first time that CP, like glipizide, significantly improved HbA1c level and other T2DM-associated glucose metabolism parameters, independent of physical activity and body weight and with no significant effects on liver and kidney function in the patients with T2DM. Moreover, CP also resulted in the improvement in the levels of blood lipid and blood pressure. These results indicate that CP displays a more beneficial effect in the alleviation of T2DM-associated metabolic phenotypes than Glipizide.

The improvements in T2DM-associated metabolic phenotypes induced by CP in the study may be explained in part by multiple chemical components in the CP, such as3-caffeoylquinic acid, isoquercitrin and kaempferol-3-glucoside. It has been shown that 3-caffeoylquinic acid is associated with a wide range of biological activities, such as antioxidation, antibacterial, antiparasitic, neuroprotective, anti-inflammatory, anticancer, antiviral, and antidiabetic effect (23). In the diabetic mice, isoquercitrin could significantly improve the sensitivity of adipose tissue to insulin and attenuate insulin resistance (24). Quercetin-3-glucuronide and kaempferol-3-glucuronideisolated from C. paliurus leaves showed potential anti-diabetic activity in STZ-stimulated mice (25). The above results were coincident with our findings that CP resulted in the alleviation of HbA1c and other T2DM-associated clinical markers and blood pressure, which was mainly associated with

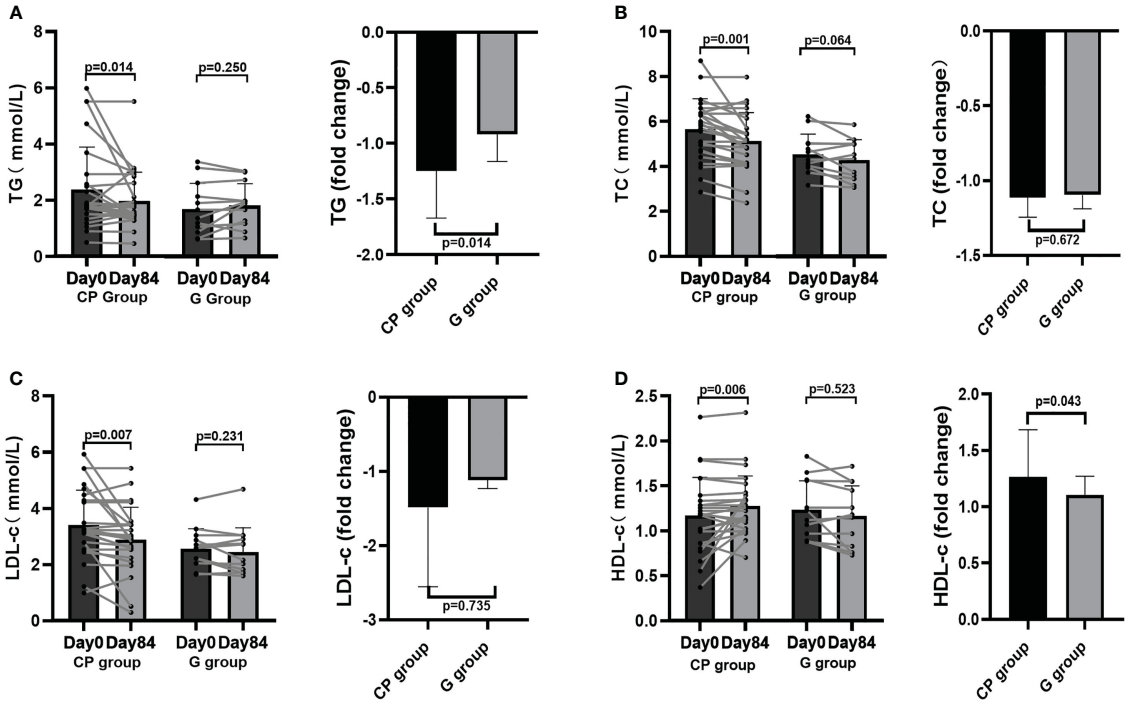

FIGURE 3
Blood lipid homeostasis in T2DM patients. **(A–D)** The changes in the levels of TG **(A)**, TC **(B)**, LDL-c **(C)**, HDL-c **(D)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the fold changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; TG, triglycerides; TC, total cholesterol; LDL-c, low-density lipoprotin cholesterol; HDL-c, high-density lipoprotein cholesterol.

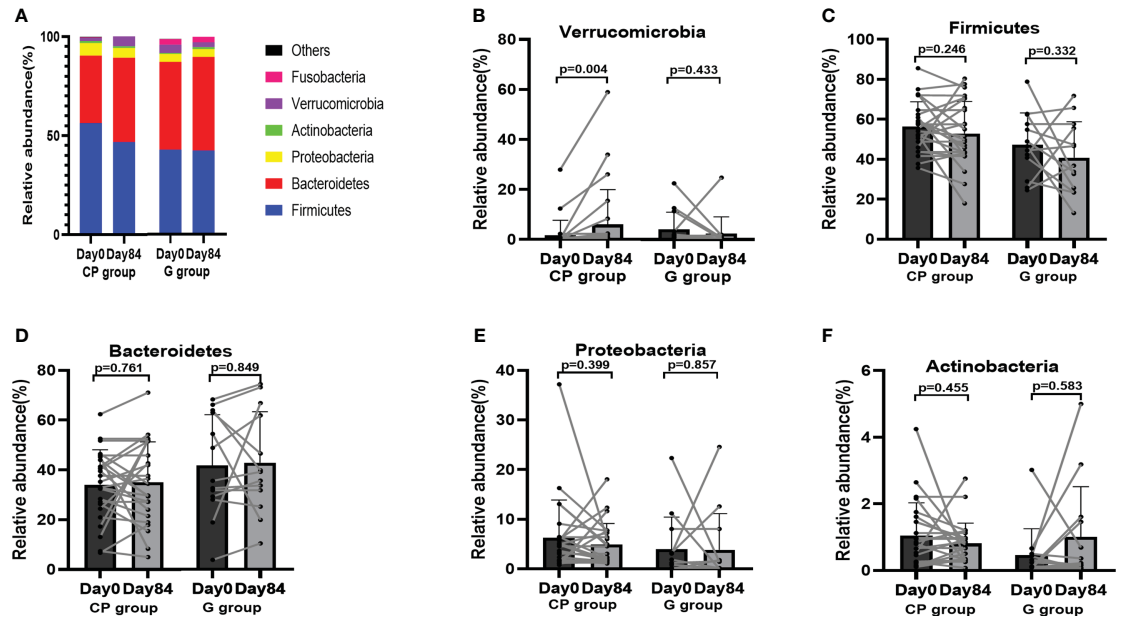

FIGURE 4
The abundances of the gut microbiota at the phylum level in T2DM patients. **(A)** The abundances of the gut microbiota at the phylum level. **(B-F)** The abundance of Verrucomicrobia **(B)**, Firmicutes **(C)**, Bacteroidetes **(D)**, Proteobacteria **(E)** and Actinobacteria **(F)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range.

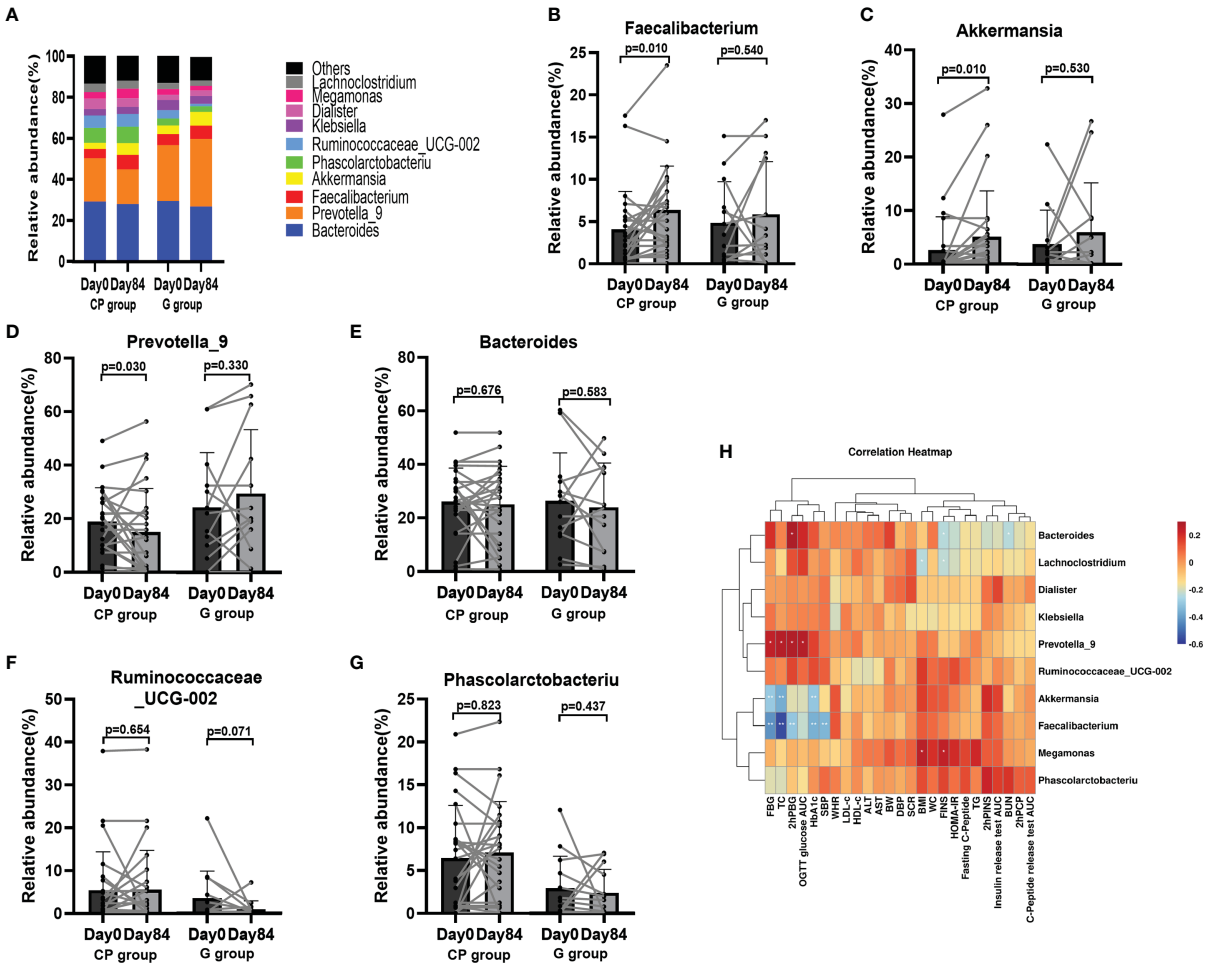

FIGURE 5
The abundances of the gut microbiota at the genus level in T2DM patients. **(A)** The abundances of the gut microbiota at genus level. **(B–G)** The abundance of Faecalibacterium **(B)**, Akkermansia **(C)**, Prevotella_9 **(D)**, Bacteroides **(E)** and Ruminococcaceae_UCG-002 **(F)** and Phascolarctobacteriu **(G)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons. **(H)** Theassociations between bacterial abundance at the genus level and metabolic parameters. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; BW, body weight; WC, waist circumference; WHR, waist to hip ratio; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; HbA1c, hemoglobin A1c; FBG, fasting blood glucose; PBG, 2-hour post-meal blood glucose; FINS, fasting insulin; hPINS, 2-hour post-mealinsulin; 2hPCP, 2-hour post-mealC-Peptide; OGTT, oral glucose tolerance test; AUC, area under curve; HOMA-IR, homeostasis model assessment for insulin resistance = (fasting blood glucose×fasting insulin/22.5); TG, triglycerides; TC, total cholesterol; LDL-c, low-density lipoprotein cholesterol; HDL-c, high-density lipoprotein cholesterol, ALB, albumin; ALT, Alanine aminotransferase, Scr, serum creatinine; BUN, blood urea nitrogen.

phytochemical compositions 3-caffeoylquinic acid, isoquercitrin and kaempferol-3-glucoside in the CP.

There is mounting evidence that gut microbiota involves the etiology and progression of T2DM, which has attracted the attention of researchers (26–28). Hence, we investigated the potential involvement in gut microbiota in mediating CP-induced T2DM improvement. In a multicenter, randomized, open label clinical trial, Chinese herbal formula alleviatedHbA1c, FBG, 2-h PBG, HOMA-IR, and TC levels through enriching Faecalibacteriumin T2DM patients with hyperlipidemia (29). Another study showed that participants with diabetes taking metformin had higher relative abundance of Akkermansia muciniphila and several SCFA-producing microbiota compared with participants without diabetes (30). In addition, a randomized, double-blind clinical trial showed that a freshwater fish-based diet ameliorated hepatic steatosis and other metabolic phenotypes by increasing Faecalibacterium abundance in patients with NAFLD (31). In agreement with human studies, the increase in the Akkermansia by metformin treatment contributed to the antidiabetic effects of metformin in diet-induced obese mice (32). Several clinical studies have found that Prevotell_9was enriched in patients with advanced liver fibrosis, liver cirrhosis, insulin resistance, type 2 diabetes, inflammation and obesity (33–35). In our study, we observed that the abundances of Faecalibacterium and Akkermansia were enriched, and the abundance of Prevotella_9 was decreased at the end of the intervention in the CP group, along with the improvement in HbA1c and other metabolic phenotypes. Supporting the aforementioned findings, our study provided new

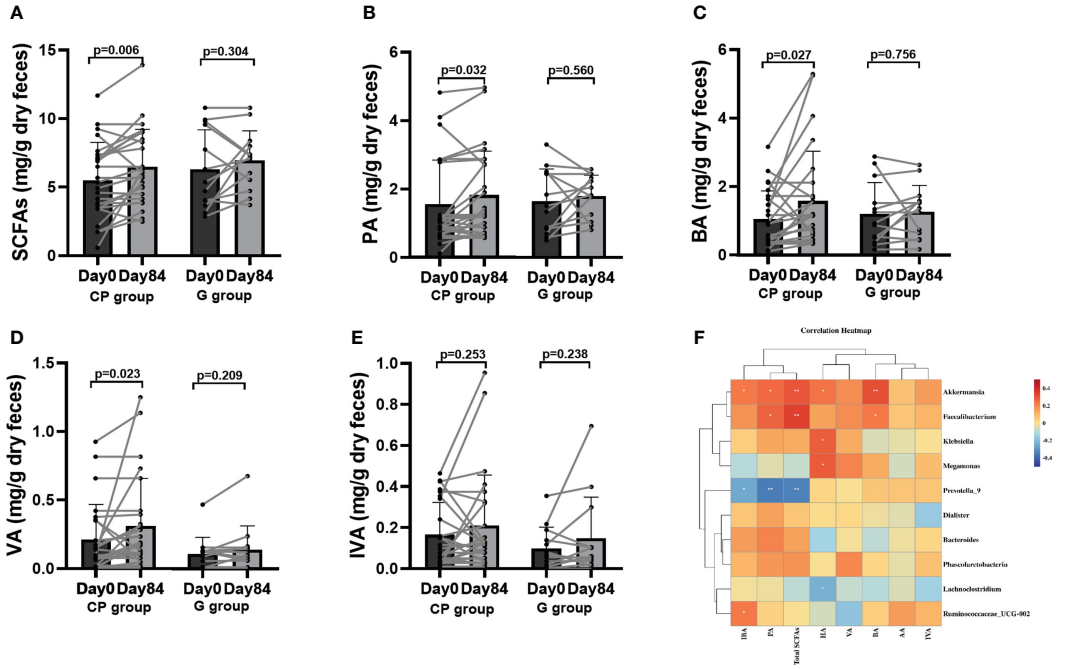

FIGURE 6
The levels of SCFAs in T2DM patients. **(A–E)** Total SCFAs **(A)**, PA **(B)**, BA **(C)**, VA **(D)** and IVA **(E)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons. **(F)** the associations between bacterial abundance and SCFAs. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; SCFAs, short chain fatty acids; AA, acetic acid; PA, propionic acid; BA, butyric acid; IBA, isobutyric acid; VA, valeric acid; IVA, isovalerlc acid; HA, hexanoic acid.

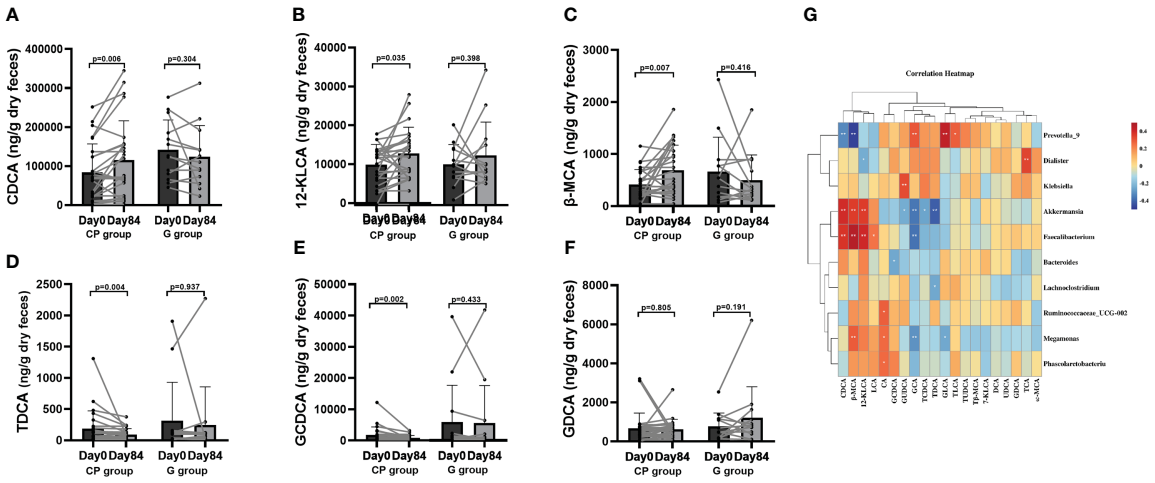

FIGURE 7
The levels of BAs in T2DM patients. **(A–F)** The changes in the levels of CDCA **(A)**,12-KLCA **(B)**, β-MCA **(C)**, TDCA **(D)**, GCDCA **(E)**, GDCA **(F)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons. **(G)** The associations between bacterial abundance and BAs. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; BA, bile acids; CDCA, chenodeoxycholic acid; DCA, deoxycholic acid; LCA, lithocholic acid; UDCA, ursodeoxycholic acid; 7-KLCA, 7-ketolithocholic acid; CA, cholic acid; 12-KLCA, 12-ketolithocholic acid; β-MCA, β-muricholic acid; ω-MCA, ω-muricholic acid; GCDCA, glycochenodeoxycholic acid; GCA, glycocholic acid; TCA, taurocholic acid; TCDCA, taurochenodeoxycholic acid; GUDCA, glycoursodeoxycholic acid; TDHCA, taurodehydrocholic acid; GDCA, glycodeoxycholic acid; TUDCA, tauroursodeoxycholic acid; TLCA, taurolithocholic acid; Tβ-MCA, tauro-β-muricholic acid; GLCA, glycolithocholic acid; TDCA, taurodeoxycholic acid.

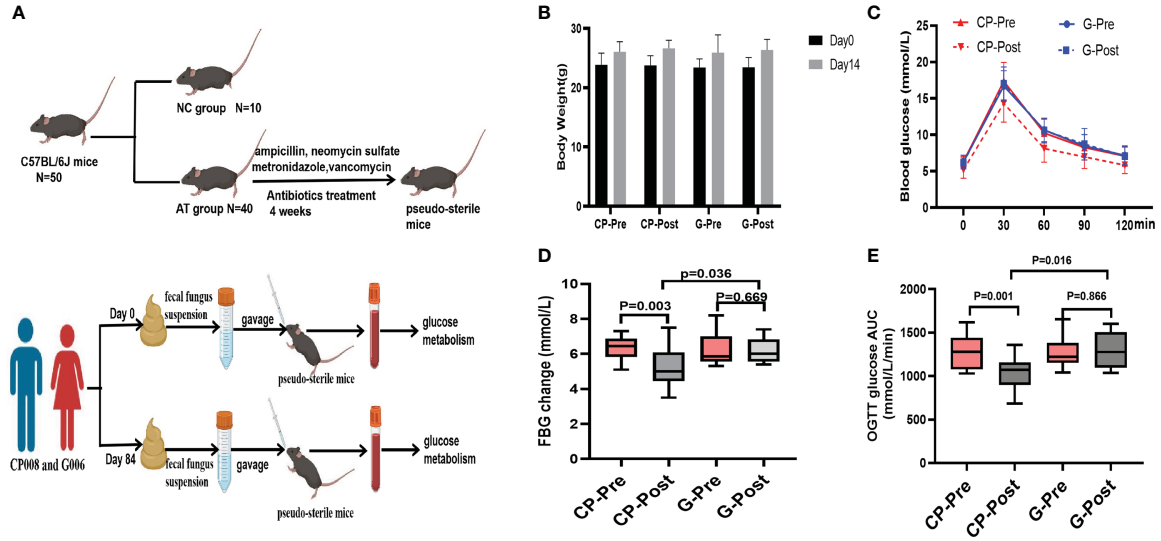

FIGURE 8
FMT improves glucose metabolism in pseudo-sterile mice. **(A)** Experimental design for animal study for microbiota transplantation. **(B)** Body weight. **(C)** OGTT blood glucose (2 weeks after transplantation) of mice receiving transplanted pre- and post-intervention human gut microbiota. **(D)** FBG level. **(E)** OGTT AUC. Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; AT group, antibiotic treatment group; NC group, normal control group.

insights into the potential role of gut microbiota in mediating the more beneficial effect of CP on T2DM than Glipizide.

On the other hand, our results showed that Glipizide did not significantly affect the composition of intestinal flora during the intervention, which was consistent with the report by Wamg et al (20). Glipizide works by binding to the sulphonylurea receptor (SUR)-1 subunit of pancreatic β-cell ATP-sensitive potassium channels, leading to their closure; the resultant membrane depolarization opens voltage-dependent calcium channels, causing intracellular calcium concentrations to increase, with subsequent release of insulin (36).

In addition to gut microbiota, metabolites including SCFAs, amino acid catabolites, and BAs produced by gut microbiota play an important role in T2DM development. These metabolites alleviate or exacerbate T2DM via their homologous receptors-mediated signaling (27, 37). We therefore attempted to clarify the potential role of metabolites (SCFAs and BAs) in mediate the beneficial impact of CP on T2DM patients. The favorable effects of SCFAs on metabolic phenotypes in T2DMhave been demonstrated. Zhao et al.indicated that SCFAs such as BA were significantly increased by gut microbiota and alleviated metabolic phenotype in T2DM patients after dietary fiber intervention (28). CP polysaccharides alleviated T2DM symptoms by increasing the SCFA-producing bacteria in type 2 diabetic rat models (15). Consistently, our study showed that total SCFAs, PA and BA were significantly enriched in the CP group at the end of the intervention. The mechanism responsible for the beneficial effects of SCFAs on T2DM involves the combination of SCFAs with G protein-coupled receptor 41 (GLPR41) and GLPR43 expressed by pancreatic β-cells (38, 39).

The results of BAs profiling indicated that the regulation of BA signal might be involved in the effect of CP on T2DM patients. On Day 84 of intervention, unconjugated BAs (CDCA, 12-KLCA and β-MCA) levels were increased, while conjugated BAs (GCDCA and TDCA) levels were reduced in the CP group compared with Day 0. Mantovani et al. reported that T2DM was significantly associated with the higher levels of conjugated BAs (TCDCA, TDCA, GCDCA, GDCA and GCA) as well as the lower level of unconjugated BAs (CA) (40). The report by Zhao showed that radix scutellariae water extract ameliorated hyperglycaemia, hyperlipidaemia and liver and kidney damage by significantly decreasing the contents of conjugated BAs (GDCA, GLCA, TLCA and TUDCA) in T2DM rats (41). The above findings supported our results. BAs are conformed to be involved in regulation of glucose and lipid response. BAs can regulate the gene expression of glucose, lipid synthesis and metabolism via binding with farnesoid X receptor and Gprotein-coupled bile acid receptor 5in the enterohepatic circulation (37).

Collectively, our results indicate that CP increases the levels of SCFAs and unconjugated BAs and decreases the levels of conjugated BAs, thus leading to the improvements in HbA1c and other metabolic indicators.

At last, FMT in pseudo-sterile mice was conducted to confirm the causal relationship between the gut microbiota and the effect of CP on host glucose metabolism. The results showed that glucose

metabolism parameters (FBG and OGTT glucose AUC) had a significant improvement in the mice transplanted with the post-intervention microbiota from the CP group compared with the mice transplanted with pre-intervention microbiota. Zhang et al. Reported that transplanted fecal bacteria from individuals with normal glucose tolerance altered gut microbiota composition and improved FBG, 2hPBG, TC, TG, and LDL-c in db/db mice (42). Our findings supported the above results, suggesting that better glucose metabolic parameters in mice was associated with the improvement in gut microbiota dysbiosis in the patient CP008 after CP intervention.

Strengths and limitations

There were several strengths in our study. First, all existing studies on CP were based on animals or *in vitro* experiments and this is the first clinical RCT to reveal the effect of CP on T2DM patients. Second, we explored the potential mechanism of CP-induced metabolic phenotypes improvement. Third, a pseudo-sterile mouse experiment was performed to verify the causality between the gut microbiota and CP-induced changes in glucose metabolism. However, there were some limits to the study. First, this was a single, open-label, controlled clinical trial, and the participants and researchers were aware of the intervention. Third, the sample size of diabetes patients included in the clinical study was relatively small. In addition, germ-free mice were not used in the FMT experiment.

Conclusion

In conclusion, this 84-day clinical randomized controlled trial in T2DM patients demonstrates that CP displays a more beneficial effect in the alleviation of T2DM-associated metabolic phenotypes than Glipizide by regulating gut microbiota and metabolites, with no significant effects on liver and kidney function. However, these results should be considered preliminary, and large-scale clinical controlled studies are required to confirm these findings.

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 ethical approval department No. KY-20181226001. The patients/participants provided their written informed consent to participate in this study. The animal study was reviewed and approved by Jinan University (20190821-02).

Author contributions

The authors' responsibilities were as follows—XP, SC, LuZ, YL, CW and ST contributed to the conception and design of this study. XP, SC, LuZ, YL, CW, LiZ, WC, JZ and JY were involved in the acquisition and analysis of the data. XP, LiZ and LuZ interpreted the results. XP, SC and ST drafted the manuscript. All authors read and approved the final manuscript.

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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/fendo.2023.1176256/full#supplementary-material

SUPPLEMENTARY FIGURE 1
Business License of Chenzhou Mingrun biological products Co. LTD, Hunan, China and cyclocarya paliurus leaves extracts. **(A)** Business license. **(B)** Food production license. **(C)** List of permitted varieties of food production. **(D)** Packaging of Cyclocarya paliurus leaves extracts. **(E)** Cyclocarya paliurus leaves extracts.

SUPPLEMENTARY FIGURE 2
HPLC fingerprinting of cyclocarya paliurus leaves extracts. HPLC chromatogram of standard mixture **(A)** and cyclocarya paliurus leaves

extracts **(B)**. External standard: (1) 3-caffeoylquinic acid, (2) 5-caffeoylquinic acid, (3) isoquercitrin, (4) kaempferol-3-glucoside, (5) kaempferol 3-rhamnoside, and (6) quercetin.

SUPPLEMENTARY FIGURE 3

The flow chart of participant recruitment and withdrawal.

SUPPLEMENTARY FIGURE 4

Anthropometric markers in T2DM patients. **(A-F)** The changes in the levels of weight **(A)**, Waist **(B)**, Waist-hip ratio **(C)**, BMI **(D)**, SBP **(E)** and DBP **(F)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the fold changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure.

SUPPLEMENTARY FIGURE 5

Liver and renal function in T2DM patients. (A-H) The changes in the levels of TBIL **(A)**, DBIL **(B)**, IBIL **(C)**, ALB **(D)**, ALT **(E)**, AST **(F)**, Scr **(G)**, BUN **(H)**. Data are presented as means ± SD or medians (IQRs). Paired T-test or non-parametric two-tailed Wilcoxon paired sign rank test was used for intra-group comparisons and independent T-test or non-parametric K independent Wilcoxon signed-ranks was used for comparisons of the fold changes between the two groups. CP group, cyclocarya paliurus leaves extracts group; G group, Glipizide group; SD, standard deviation; IQR, interquartile range; TBIL, total bilirubin; DBIL, direct bilirubin; IBIL, indirect bilirubin;ALB, albumin;ALT, Alanine aminotransferase; AST, Aspartate aminotransferase; Scr, serum creatinine; BUN, blood urea nitrogen.

References

1. Sun H, Saeedi PY, Karuranga SV, Pinkepank M, Ogurtsova K, Duncan BB, et al. IDF diabetes atlas: global, regional and country-level diabetes prevalence estimates for 2021 and projections for 2045. *Diabetes Res Clin Pract* (2022) 183:109119. doi: 10.1016/j.diabres.2021.109119

2. Eckel RH, Grundy SM, Zimmet PZ. The metabolic syndrome. *Lancet* (2005) 365 (9468):1415–28. doi: 10.1016/S0140-6736(05)66378-7

3. Dehghan-Kooshkghazi M, Mathers JC. Starch digestion, large-bowel fermentation and intestinal mucosal cell proliferation in rats treated with the alpha-glucosidase inhibitor acarbose. *Br J Nutr* (2004) 91(3):357–65. doi: 10.1079/BJN20031063

4. Baig MA, Panchal SS. Streptozotocin-induced diabetes mellitus in neonatal rats: an insight into its applications to induce diabetic complications. *Curr Diabetes Rev* (2019) 16(1):26–39. doi: 10.2174/1573399815666190411115829

5. Li QQ, Hu JL, Xie JH, Nie SP, Xie MY. Isolation, structure, and bioactivities of polysaccharides from cyclocarya paliurus (Batal.) iljinskaja. *Ann N Y Acad Sci* (2017) 1398(1):20–9. doi: 10.1111/nyas.13357

6. Liu Y, Qian CY, Ding SH, Shang XL, Yang WX, Fang SZ, et al. Effect of light regime and provenance on leaf characteristics, growth and flavonoid accumulation in cyclocarya paliurus (Batal) iljinskaja coppices. *Bot Stud* (2016) 57(1):28. doi: 10.1186/s40529-016-0145-7

7. Xie J-H, Xie M-Y, Nie S-P, Shen M-Y, Wang Y-X, Li C. Isolation, chemical composition and antioxidant activities of a water-soluble polysaccharide from cyclocarya paliurus (Batal.) iljinskaja. *Food Chem* (2010) 119(4):1626–32. doi: 10.1016/j.foodchem.2009.09.055

8. Yang ZW, Wang J, Li JG, Xiong L, Chen H, Liu X, et al. Antihyperlipidemic and hepatoprotective activities of polysaccharide fraction from cyclocarya paliurus in high-fat emulsion-induced hyperlipidaemic mice. *Carbohydr Polym* (2018) 183:11–20. doi: 10.1016/j.carbpol.2017.11.033

9. Wang X, Li W, Kong D. Cyclocarya paliurus extract alleviates diabetic nephropathy by inhibiting oxidative stress and aldose reductase. *Ren Fail* (2016) 38 (5):678–85. doi: 10.3109/0886022X.2016.1155394

10. Zhao LC, Wang X, Li JX, Tan XM, Fan LL, Zhang ZW, et al. Effect of cyclocarya paliurus on hypoglycemic effect in type 2 diabetic mice. *Med Sci Monit* (2019) 25:2976–83. doi: 10.12659/MSM.913368

11. Xiao HT, Wen B, Ning ZW, Zhai LX, Liao CH, Lin CY, et al. Cyclocarya paliurus tea leaves enhances pancreatic β cell preservation through inhibition of apoptosis. *Sci Rep* (2017) 7(1):9155. doi: 10.1038/s41598-017-09641-z

12. Yang ZW, Ouyang KH, Zhao J, Chen H, Xiong L, Wang WL, et al. Structural characterization and hypolipidemic effect of cyclocarya paliurus polysaccharide in rat. *Int J Biol Macromol* (2016) 91:1073–80. doi: 10.1016/j.ijbiomac.2016.06.063

13. Chen ZL, Jian YQ, Wu Q, Wu J, Sheng WB, Jiang S, et al. Cyclocarya paliurus (Batalin) iljinskaja: botany, ethnopharmacology, phytochemistry and pharmacology. *J Ethnopharmacol* (2022) 285:114912. doi: 10.1016/j.jep.2021.114912

14. Li QQ, Hu JL, Nie QX, Chang X, Fang QY, Xie JH, et al. Hypoglycemic mechanism of polysaccharide from cyclocarya paliurus leaves in type 2 diabetic rats by gut microbiota and host metabolism alteration. *Sci China Life Sci* (2021) 64(1):117–32. doi: 10.1007/s11427-019-1647-6

15. Yao Y, Yan LJ, Chen H, Wu N, Wang WB, Wang DS, et al. Cyclocarya paliurus polysaccharides alleviate type 2 diabetic symptoms by modulating gut microbiota and short-chain fatty acids. *Phytomedicine* (2020) 77:153268. doi: 10.1016/j.phymed.2020.153268

16. Schulz KF, Altman DG, Moher D. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *Bmj* (2010) 340:c332. doi: 10.1136/bmj.c332

17. Gillet MJ. International expert committee report on the role of the A1C assay in the diagnosis of diabetes. *Diabetes Care* (2009) 32(7):1327–34.

18. Report of the expert committee on the diagnosis and classification of diabetes mellitus. *Diabetes Care* (1997) 20(7):1183–97. doi: 10.2337/diacare.20.7.1183

19. Movahed A, Nabipour I, Louis XL, Thandapilly SJ, Yu LP, Kalantarhormozi M, et al. Antihyperglycemic effects of short term resveratrol supplementation in type 2 diabetic patients. *Evid Based Complement Alternat Med* (2013) 2013:851267. doi: 10.1155/2013/851267

20. Gu Y, Wang X, Li J, Zhang Y, Zhong H, Liu R, et al. Analyses of gut microbiota and plasma bile acids enable stratification of patients for antidiabetic treatment. *Nat Commun* (2017) 8(1):1785. doi: 10.1038/s41467-017-01682-2

21. Zhai L, Ning ZW, Huang T, Wen B, Liao CH, Lin CY, et al. Cyclocarya paliurus leaves tea improves dyslipidemia in diabetic mice: a lipidomics-based network pharmacology study. *Front Pharmacol* (2018) 9:973. doi: 10.3389/fphar.2018.00973

22. Ainsworth BE, Addy CL, Whitt MC, Stolarczyk LM. Moderate physical activity patterns of minority women: the cross-cultural activity participation study. *J Womens Health Gend Based Med* (1999) 8(6):805–13. doi: 10.1089/152460999319129

23. Liang N, Kitts DD. Role of chlorogenic acids in controlling oxidative and inflammatory stress conditions. *Nutrients* (2015) 8(1):16. doi: 10.3390/nu8010016

24. Li SH, Ji T, Su SL, Zhu Y, Chen XL, Shang EX, et al. Mulberry leaves ameliorate diabetes via regulating metabolic profiling and AGEs/RAGE and p38 MAPK/NF-κB pathway. *J Ethnopharmacol* (2022) 283:114713. doi: 10.1016/j.jep.2021.114713

25. Liu Y, Cao Y, Fang S, Wang T, Yin Z, Shang X, et al. Antidiabetic effect of cyclocarya paliurus leaves depends on the contents of antihyperglycemic flavonoids and antihyperlipidemic triterpenoids. *Molecules* (2018) 23(5). doi: 10.3390/molecules23051042

26. Tanase DM, Gosav EM, Neculae E, Costea CF, Ciocoiu M, Hurjui LL, et al. Role of gut microbiota on onset and progression of microvascular complications of type 2 diabetes (T2DM). *Nutrients* (2020) 12(12). doi: 10.3390/nu12123719

27. Canfora EE, Meex RCR, Venema K, Blaak EE. Gut microbial metabolites in obesity, NAFLD and T2DM. *Nat Rev Endocrinol* (2019) 15(5):261–73. doi: 10.1038/s41574-019-0156-z

28. Zhao LP, Zhang F, Ding XP, Wu GJ, Lam YY, Wang XJ, et al. Gut bacteria selectively promoted by dietary fibers alleviate type 2 diabetes. *Science* (2018) 359 (6380):1151–6. doi: 10.1126/science.aao5774

29. Tong X, Xu J, Lian FM, Yu XT, Zhao YF, Xu LP, et al. Structural alteration of gut microbiota during the amelioration of human type 2 diabetes with hyperlipidemia by metformin and a traditional Chinese herbal formula: a multicenter, randomized, open label clinical trial. *mBio* (2018) 9(3):e02392–17. doi: 10.1128/mBio.02392-17

30. Cuesta-Zuluaga JDL, Mueller NT, Corrales-Agudelo VEP, Mejía V, Carmona JA, Abad JM, et al. Metformin is associated with higher relative abundance of mucin-degrading akkermansia muciniphila and several short-chain fatty acid-producing microbiota in the gut. *Diabetes Care* (2017) 40(1):54–62. doi: 10.2337/dc16-1324

31. He KY, Guo LLZ, Tang HJ, Peng XJ, Li J, Feng SF, et al. A freshwater fish-based diet alleviates liver steatosis by modulating gut microbiota and metabolites: a clinical randomized controlled trial in Chinese participants with nonalcoholic fatty liver disease. *Am J Gastroenterol* (2022) 117(10):1621–31. doi: 10.14309/ajg.0000000000001885

32. Shin NR, Lee JC, Lee HY, Kim MS, Whon TW, Lee MS, et al. An increase in the akkermansia spp. population induced by metformin treatment improves glucose homeostasis in diet-induced obese mice. *Gut* (2014) 63(5):727–35. 10.1136/gutjnl-2012-303839

33. Dong TS, Katzka W, Lagishetty V, Luu KT, Hauer M, Pisegna J, et al. A microbial signature identifies advanced fibrosis in patients with chronic liver disease mainly due to NAFLD. *Sci Rep* (2020) 10(1):2771. doi: 10.1038/s41598-020-59535-w

34. Fugmann M, Breier M, Rottenkolber M, Banning F, Ferrari U, Sacco V, et al. The stool microbiota of insulin resistant women with recent gestational diabetes, a high risk group for type 2 diabetes. *Sci Rep* (2015) 5:13212. doi: 10.1038/srep13212

35. Qin JJ, Li YR, Cai ZM, Li SH, Zhu JF, Zhang F, et al. A metagenome-wide association study of gut microbiota in type 2 diabetes. *Nature* (2012) 490(7418):55–60. doi: 10.1038/nature11450

36. Deacon CF, Lebovitz HE. Comparative review of dipeptidyl peptidase-4 inhibitors and sulphonylureas. *Diabetes Obes Metab* (2016) 18(4):333–47. doi: 10.1111/dom.12610

37. Yang Q, Vijayakumar A, Kahn BB. Metabolites as regulators of insulin sensitivity and metabolism. *Nat Rev Mol Cell Biol* (2018) 19(10):654–72. doi: 10.1038/s41580-018-0044-8

38. Veprik A, Laufer D, Weiss S, Rubins N, Walker MD. GPR41 modulates insulin secretion and gene expression in pancreatic β-cells and modifies metabolic homeostasis in fed and fasting states. *FASEB J* (2016) 30(11):3860–9. doi: 10.1096/fj.201500030R

39. McNelis JC, Lee YS, Mayoral R, Kant RVD, Johnson AMF, Wollam J, et al. GPR43 potentiates β-cell function in obesity. *Diabetes* (2015) 64(9):3203–17. doi: 10.2337/db14-1938

40. Mantovani A, Dalbeni A, Peserico D, Cattazzo F, Bevilacqua M, Salvagno JL, et al. Plasma bile acid profile in patients with and without type 2 diabetes. *Metabolites* (2021) 11(7):453. doi: 10.3390/metabo11070453

41. Zhao LJ, Ma P, Peng Y, Wang MY, Peng CS, Zhang YL, et al. Amelioration of hyperglycaemia and hyperlipidaemia by adjusting the interplay between gut microbiota and bile acid metabolism: radix scutellariae as a case. *Phytomedicine* (2021) 83:153477. doi: 10.1016/j.phymed.2021.153477

42. Zhang PP, Li LL, Han X, Li QW, Zhang XH, Liu JJ, et al. Fecal microbiota transplantation improves metabolism and gut microbiome composition in db/db mice. *Acta Pharmacol Sin* (2020) 41(5):678–85. doi: 10.1038/s41401-019-0330-9t

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Glossary

T2DM	type 2 diabetes mellitus
FDA	Food and Drug Administration
SCFAs	short-chain fatty acids
CP group	cyclocarya paliurus leaves extracts group
G group	Glipizide group
FBG	fasting plasma glucose
METs	Metabolic Equivalent Tasks
BMI	body mass index
BW	body weight
WC	waist circumference
WHR	waist to hip ratio
SBP	systolic blood pressure
DBP	diastolic blood pressure
OGTT	oral glucose tolerance test
FINS	fasting insulin
2hPINS	2-hour post-meal insulin
FCP	fasting C- peptide, 2hPCP, 2-hour post-meal C-Peptide, the area under curve of C-peptide release test, TC, total cholesterol
TG	triglycerides
LDL-c	low-density lipoprotein cholesterol
HDL-c	high-density lipoprotein cholesterol
TBIL	the total bilirubin
DBIL	direct bilirubin
IBIL	indirect bilirubin
ALB	albumin
ALT	alanine aminotransferase
AST	aspartate aminotransferase
Scr	serum creatinine
BUN	blood urea nitrogen
FMT	fecal microbiota transplantation
NC	control group
AT	antibiotic treatment
ITT	intention-to-treat
SPSS	Statistical Package for Social Science
HPLC	high-performance liquid chromatography
Rt	retention times
UV	ultraviolet

(Continued)

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PA	propionic acid
BA	butyric acid
AA	acetic acid
IBA	isobutyric acid
VA	valeric acid
IVA	isovaleric acid
HA	hexanoic acid
BAs	bile acids
CDCA	chenodeoxycholic acid
12-KLCA	12-ketolithocholic acid
β-MCA	β-muricholic acid
TDCA	taurodeoxycholic acid
GCDCA	glycochenodeoxycholic acid
SUR	sulphonylurea receptor
GLPR41	G protein-coupled receptor 41.

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Clonal hematopoiesis as a novel risk factor for type 2 diabetes mellitus in patients with hypercholesterolemia

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Introduction: Clonal hematopoiesis of indeterminate potential (CHIP) is associated with atherosclerosis and cardiovascular disease. It has been suggested that CHIP may be related to diabetes, so we investigated the association between CHIP and new-onset type 2 diabetes.

Methods: This study included 4,047 subjects aged >=40 years without diabetes. To detect CHIP, targeted gene sequencing of genomic DNA from peripheral blood cells was performed. The incidence of new-onset type 2 diabetes during the follow-up period was evaluated.

Results: Of the total subjects, 635 (15.7%) had CHIP. During the median follow-up of 5.1 years, the incidence of new-onset diabetes was significantly higher in CHIP carriers than in subjects without CHIP (11.8% vs. 9.1%, $p = 0.039$). In a univariate analysis, CHIP significantly increased the risk of new-onset diabetes (HR 1.32, 95% CI 1.02–1.70, $p = 0.034$), but in a multivariate analysis, it was not significant. The CHIP-related risk of new onset diabetes differed according to LDL cholesterol level. In the hyper-LDL cholesterolemia group, CHIP significantly increased the risk of diabetes (HR 1.64, 95% CI 1.09–2.47, $p = 0.018$), but it did not increase the risk in the non-hyper-LDL cholesterolemia group. The subjects with CHIP and hyper-LDL-cholesterolemia had approximately twice the risk of diabetes than subjects without CHIP and with low LDL cholesterol (HR 2.05, 95% CI 1.40–3.00, $p < 0.001$).

Conclusion: The presence of CHIP was a significant risk factor for new-onset type 2 diabetes, especially in subjects with high LDL cholesterol. These results show the synergism between CHIP and high LDL cholesterol as a high-risk factor for diabetes.

KEYWORDS

clonal hematopoiesis, diabetes mellitus, hematopoiesis, hypercholesterolemia, LDL cholesterol

1. Introduction

The development of type 2 diabetes is very complex, involving several genetic and environmental factors. Age, race/ethnicity, family history of diabetes, obesity, and physical inactivity are known as traditional risk factors (1). However, these traditional risk factors alone cannot fully explain the development of type 2 diabetes. The previous prediction models generated with these traditional risk factors had limited ability to predict future type 2 diabetes (2–4). Therefore, studies have been conducted to find new factors conferring

residual risk for type 2 diabetes. For example, genome-wide association studies have recently been performed to find new genetic risk factors, and based on these studies, it is estimated that genetic variation can explain 18%−45% of the risk for type 2 diabetes (5, 6). Even so, there are still risk factors for type 2 diabetes that have not been discovered.

Aging is accompanied by the accumulation of somatic mutations in hematopoietic stem cells. These mutations can cause hematologic malignancies, such as acute leukemia, but this does not happen in most individuals. The clonal expansion of hematopoietic cells with aging-related recurrent somatic mutations in the absence of other hematologic abnormalities is called clonal hematopoiesis of indeterminate potential (CHIP) (7). CHIP has been reported to be associated with an increased risk of cardiovascular disease, including coronary heart disease and stroke, and higher mortality (8, 9). CHIP is an emerging risk factor for atherosclerosis (10, 11).

Type 2 diabetes is a potent risk factor for cardiovascular disease and atherosclerosis. Like CHIP, type 2 diabetes is associated with aging. A meta-analysis reported that type 2 diabetes is associated with an increased risk of hematologic malignancy (12). These results suggest that CHIP may have some relationship with type 2 diabetes. Bonnefond et al. reported that large clonal mosaicism in peripheral blood cells was associated with type 2 diabetes (13). Jaiswal et al. reported that CHIP was modestly but significantly associated with an increased risk of type 2 diabetes (odds ratio 1.3) (14). Owing to the limitations of cross-sectional studies, whether CHIP promotes the development of type 2 diabetes is not yet understood. Therefore, in this study, we investigated the association between CHIP and new-onset type 2 diabetes in a longitudinal retrospective cohort study.

2. Materials and methods

2.1. Study population

The Gene-ENvironmental Interaction and phEnotype (GENIE)-CHIP cohort was designed to investigate the effects of CHIP on health outcomes. This retrospective cohort included subjects aged ≥65 years or aged 40–64 years with more than one of the risk factors for cardiovascular disease (diabetes, hypertension, dyslipidemia, chronic kidney disease, and current smoking) who came to the Seoul National University Hospital Healthcare System Gangnam Center for a health check-up for screening purposes between January 2014 and January 2017. The GENIE-CHIP cohort was a subcohort of the GENIE cohort, and details of the GENIE cohort were described previously (15). Of the total 4,991 subjects in the GENIE-CHIP cohort, subjects who did not have glucose or low-density lipoprotein (LDL) cholesterol (n = 64) were excluded, and 4,927 subjects were finally analyzed (Supplementary Figure S1). Type 2 diabetes was defined as a fasting glucose ≥126 mg/dl or HbA1c ≥6.5% and/or treatment with glucose-lowering medication. Hypertension was defined as a systolic blood pressure (BP) ≥140 mmHg or diastolic BP ≥90 mmHg or the use of anti-hypertensive medications. Dyslipidemia was defined as total cholesterol ≥240 mg/dL, LDL cholesterol ≥160 mg/dL, triglycerides ≥200 mg/dL, high-density lipoprotein (HDL) cholesterol <40 mg/dL, or the use of medication for dyslipidemia.

Informed consent was obtained from participants in the GENIE cohort, and blood samples were collected with the approval of the Seoul National University Hospital Institutional Review Board (H-1103-127-357). The protocol of this retrospective cohort study was additionally approved by the institutional review board (H-1908-121-1056), and informed consent was waived owing to its retrospective nature.

2.2. Targeted gene sequencing

Genomic DNA was isolated from peripheral blood. Targeted gene sequencing was performed using an Agilent SureSelectXT HS custom panel and the Illumina NovaSeq 6000 with 2×150 bp paired-end reads with a minimum coverage of 800×. The custom panel comprised all the exons of 89 genes frequently involved in CHIP, such as DNMT3A, TET2, ASXL1, JAK2, and TP53 (8, 14, 16–18). The detailed process of CHIP variant calling is described in a previous study (19). The unreliable variants, which met any one of the following criteria, were filtered out as sequencing artifacts or germline variants as follows: (1) the number of altered reads on the positive and negative strands was <5, the mapping quality was <30, or the base quality was <30; (2) the variant allele frequency (VAF) was not between 1.5% and 30%; (3) it was among the common germline variants, including those listed in genomAD, 1 k Genome v3, ESP6500, and ExAC; and (4) it was listed in the artifact database with a minor allele frequency of >2% in the internal panel of 1,000 Korean individuals. All reliable non-synonymous variants were annotated as CHIP mutations.

2.3. Statistical analyses

Continuous variables are presented as the mean ± standard deviation or median with interquartile range and were compared using Student's t-test for independent samples or the Mann–Whitney test. Categorical variables are expressed as numbers and percentages and were compared using the $\chi 2$ test or Fisher's exact test. The association between the presence of CHIP and new-onset type 2 diabetes was tested using a Cox proportional hazard model with covariates including age, sex, BMI, and other diabetes risk factors. Diabetes-free survival, estimated using the Kaplan–Meier method, was compared between the groups using the log-rank test. The attributable proportion (AP) due to the interaction between CHIP and high LDL cholesterol was calculated using the epiR package of R statistics (version 4.1.2., R Foundation for Statistical Computing). For this analysis, participants were included in the hyper-LDL cholesterolemia (hyperLDLC) group if their LDL cholesterol level was ≥160 mg/dL (classified as "high level" by the National Cholesterol Education Program) (20, 21) or if they took medication for dyslipidemia. Samples with covariates missing were removed in the risk analysis and interaction analyses. All statistical analyses were performed using Python version 3.7.9 (Python Software Foundation) and its packages Numpy (1.19.4), Scipy (1.5.3), Scikit-learn (0.23.2), and Lifelines (0.25.6).

3. Results

3.1. Baseline characteristics and the prevalence of type 2 diabetes

Of the total 4,927 subjects, the mean age was 55.4 ± 8.1 years, 73% were men, and 790 (16.0%) had CHIP (Supplementary Table S1). The prevalence of subjects with CHIP (CHIP carrier) increased with age, and gene mutations in DNMT3A and TET2 were the most frequently identified (Supplementary Figure S2). Compared with subjects without CHIP, CHIP carriers were significantly older (58.7 ± 8.5 years old vs. 54.7 ± 7.9 years old, $p < 0.001$) and had lower levels of LDL cholesterol (120.9 ± 31.7 mg/dL vs. 125.4 ± 32.8 mg/dL, $p < 0.001$). However, sex, BMI, waist circumference, and the prevalence of hypertension and dyslipidemia were not different between subjects with and without CHIP (Supplementary Table S1). Of the total 4,927 subjects, 696 (14.1%) had type 2 diabetes. The prevalence of type 2 diabetes in subjects with CHIP (15.2%) was higher than that in subjects without CHIP (13.9%), but it was statistically insignificant.

3.2. Impact of CHIP on the incidence of new-onset type 2 diabetes

Among the 4,231 subjects without type 2 diabetes, 4,047 (96%) had at least one follow-up examination. The baseline blood levels of glucose and HbA1C were slightly higher in subjects with CHIP than in subjects without CHIP (Table 1). The median follow-up duration was 5.1 years (interquartile range 3.3–6.1 years), and 385 (9.5%) subjects were newly diagnosed with type 2 diabetes. The median time to new-onset type 2 diabetes was 3.1 years (interquartile range 2.0–4.9 years). The incidence of type 2 diabetes in CHIP carriers (11.8%) was significantly higher than that in subjects without CHIP (9.1%) ($p = 0.039$). The Kaplan–Meier curve demonstrated that CHIP carriers had a higher risk of new-onset type 2 diabetes (log-rank $p = 0.033$; Figure 1A). To investigate the effect of CHIP on new-onset type 2 diabetes, a Cox regression analysis was performed. In a univariate analysis, CHIP significantly increased the risk of new-onset type 2 diabetes (hazard ratio [HR] 1.32, 95% confidence interval [CI] 1.02–1.70, $p = 0.034$). However, in a multivariate analysis adjusting for age, sex, BMI, and family history of diabetes, it was not significant (HR 1.19, 95% CI 0.92–1.54, $p = 0.194$) (Supplementary Table S2).

In addition, it was investigated whether CHIP clone size or specific CHIP mutation contributed more to the increased risk of new-onset type 2 diabetes. Among CHIP carriers, 14.4% (92/635) had large CHIP clones defined as VAF ≥10%. Large CHIP carriers had a higher risk of new-onset type 2 diabetes compared with small CHIP carriers or subjects without CHIP in the Kaplan–Meier curve (Supplementary Figure S3). When examining individual CHIP mutations, there was no single mutation that significantly increased the risk of new-onset type 2 diabetes (Supplementary Figure S3).

3.3. Impact of CHIP on new-onset type 2 diabetes and its interaction with LDL cholesterol

To identify any subgroups that had a greater impact on CHIP, an interaction analysis between CHIP and clinical factors was conducted. Among the clinical factors, only LDL cholesterol showed a significant interaction with CHIP for new-onset type 2 diabetes ($p = 0.030$, Supplementary Table S3). Therefore, the subjects were divided into the non-hyperLDLC group (LDL cholesterol <160 mg/dL and no medication for dyslipidemia) and the hyperLDLC group (LDL cholesterol ≥160 mg/dL and/or medication for dyslipidemia). The Kaplan–Meier curve demonstrated that CHIP carriers had a higher risk of new-onset type 2 diabetes only in the hyperLDLC group (log-rank $p = 0.004$; Figures 1B, C). A multivariate Cox regression analysis also showed that CHIP significantly increased the risk of new-onset type 2 diabetes in the hyperLDLC group (adjusted HR 1.64, 95% CI 1.09–2.47, $p = 0.018$) but not in the non-hyperLDLC group (adjusted HR 0.98, 95% CI 0.69–1.38, $p = 0.894$) (Figure 1D). Furthermore, CHIP and high LDL cholesterol demonstrated a significant synergistic effect on the development of new-onset type 2 diabetes (Table 2). The subgroup analysis showed that CHIP carriers in the hyperLDLC group had an ∼2-fold higher risk of developing new-onset type 2 diabetes than non-CHIP carriers in the non-hyperLDLC group (HR 2.05, 95% CI 1.40–3.00, $p < 0.001$). This interaction between LDL cholesterol and CHIP may significantly contribute to the development of new-onset type 2 diabetes, at a rate of 34% (Supplementary Table S3). It suggested the synergistic effect of CHIP and high LDL cholesterol on new-onset type 2 diabetes. To exclude the effects of statins on the risk of new-onset type 2 diabetes, a subgroup analysis was conducted only in those who did not take medication for dyslipidemia, and the results were similar (Supplementary Table S4).

4. Discussion

The present study demonstrated that CHIP increased the risk of new-onset type 2 diabetes in subjects with high LDL cholesterol. CHIP had a significant interaction with LDL cholesterol, and they showed synergism in increasing the risk for new-onset type 2 diabetes. The risk for new-onset type 2 diabetes in CHIP carriers in the hyperLDLC group was twice that of non-CHIP carriers in the non-hyperLDLC group.

4.1. Association between CHIP and type 2 diabetes

To the best of our knowledge, this is the first cohort study demonstrating a significant association between CHIP and the development of new-onset type 2 diabetes. Although Jaiswal et al. reported the association between CHIP and type 2 diabetes for the first time, it was a cross-sectional study (14). Additionally, its association was only significant in European and South Asian populations (14). In the present study of an East Asian population,

TABLE 1 Clinical characteristics of study subjects according to CHIP status.

	Total (n = 4,047)	CHIP carrier (n = 635)	Non-carrier (n = 3,412)	p
Age, years	54.8 ± 8.0	58.3 ± 8.6	54.1 ± 7.7	<0.001
Male	2,915 (72.0)	454 (71.5)	2,461 (72.1)	1.000
BMI, kg/m^2	24.0 ± 2.8	24.0 ± 2.5	24.0 ± 2.8	0.793
BMI ≥ 25 kg/m^2	1,356 (33.5)	224 (35.3)	1,132 (33.2)	0.314
WC, cm	85.5 ± 7.7	86.0 ± 7.3	85.4 ± 7.7	0.096
Systolic BP, mmHg	118.2 ± 13.2	118.8 ± 13.2	118.1 ± 13.2	0.263
Diastolic BP, mmHg	78.6 ± 10.1	77.9 ± 9.5	78.7 ± 10.2	0.071
Fasting glucose, mg/dL	98.0 ± 9.5	98.7 ± 9.8	97.9 ± 9.5	0.060
HbA1C, %	5.6 ± 0.3	5.7 ± 0.3	5.6 ± 0.3	0.005
Total cholesterol, mg/dL	198.9 ± 36.9	194.7 ± 38.9	199.6 ± 36.5	0.002
Triglycerides, mg/dL	106.0 (74.0–150.0)	102.0 (73.0–145.0)	107.0 (75.0–151.0)	0.067
HDL cholesterol, mg/dL	52.0 ± 12.4	51.8 ± 12.3	52.0 ± 12.4	0.728
LDL cholesterol, mg/dL	127.3 ± 32.0	123.5 ± 31.1	128.0 ± 32.1	0.001
Hypertension	1,425 (35.2)	226 (35.6)	1,199 (35.1)	0.821
Dyslipidemia	2,178 (53.8)	323 (50.9)	1,855 (54.4)	0.109
Medication for dyslipidemia	540 (13.3)	93 (14.6)	447 (13.1)	0.309
Family history of diabetes	980 (24.2)	148 (23.3)	832 (24.4)	0.579
Follow-up duration, years	5.1 (3.3–6.1)	5.1 (3.0–6.1)	5.1 (3.3–6.1)	0.323
New-onset type 2 diabetes	385 (9.5)	75 (11.8)	310 (9.1)	0.039

Values are mean ± standard deviation, median (interquartile range), or n (%).
CHIP, clonal hematopoiesis of indeterminate potential; BMI, body mass index; WC, waist circumference; BP, blood pressure.

the prevalence of type 2 diabetes was not different between subjects with and without CHIP, but CHIP increased the risk of new-onset type 2 diabetes. In this study, we accepted a broader definition of CHIP mutations than the previous study (14). CHIP mutations are classified into putative drivers (PD-CHIP) and non-putative drivers (non-PD-CHIP) (13). While Jaiswal et al. defined only hematologic cancer driver mutations as CHIP mutations (PD-CHIP), we included both PD and non-PD-CHIP mutations that cause any of the non-synonymous amino acid alterations in this study. Clonal expansions can be induced by various genetic alterations, including copy number variants (CNVs), structural variations, and epigenetic changes, not just somatic "driver" mutations (PD-CHIP). Hence, non-PD-CHIP mutations could be considered "passengers" in clonal expansion caused by other alterations that could not be detected using our targeted panels but may still have clinical consequences. Recently, it is reported that a large CHIP clone defined as VAF ≥10% was associated with a greater risk of cardiovascular disease and all-cause mortality (22). In this study, the Kaplan–Meier curve showed that large CHIP was associated with a higher risk of new-onset type 2 diabetes compared with small CHIP (Supplementary Figure S3). However, it was insignificant in a multivariate analysis adjusting for age, sex, BMI, and family history of diabetes (HR 1.62, 95% CI 0.96–2.72, p = 0.069), and this may be because of the small number of large CHIP carriers. Further large-scale longitudinal studies on the association between CHIP and new-onset type 2 diabetes are needed to validate our results.

4.2. Plausible mechanism underlying the effect of CHIP on the development of type 2 diabetes

Among genes mutated in CHIP, DNMT3A and TET2 were the most frequent (9, 14, 23). Consistent with these results, DNMT3A and TET2 were the most commonly mutated genes in both our total subjects and our subjects with new-onset type 2 diabetes (Supplementary Figure S2).

CHIP may affect insulin resistance and contribute to the development of type 2 diabetes. Fuster et al. generated mice with CHIP to investigate the effect of CHIP on atherosclerosis and insulin resistance (24, 25). Tet2 loss-of-function-driven clonal hematopoiesis showed a progressive aggravation of insulin resistance and an increase in fasting blood glucose levels in aged, obese mice (25). It was accompanied by increased proinflammatory cytokine interleukin (IL)-1β expression in white adipose tissues. These results suggest that inflammation is an important mechanism underlying the effect of CHIP on insulin resistance.

CHIP shifts macrophages to a more inflammatory state. Dnmt3a or Tet2-deficient J774.1 myeloid cells created using a lentivirus/CRISPR system showed increased expression of inflammatory cytokines and chemokines, such as IL-1β, IL-6, and CCL5 (26). Inflammatory cytokines, including IL-1β, were upregulated in macrophages isolated from mice with

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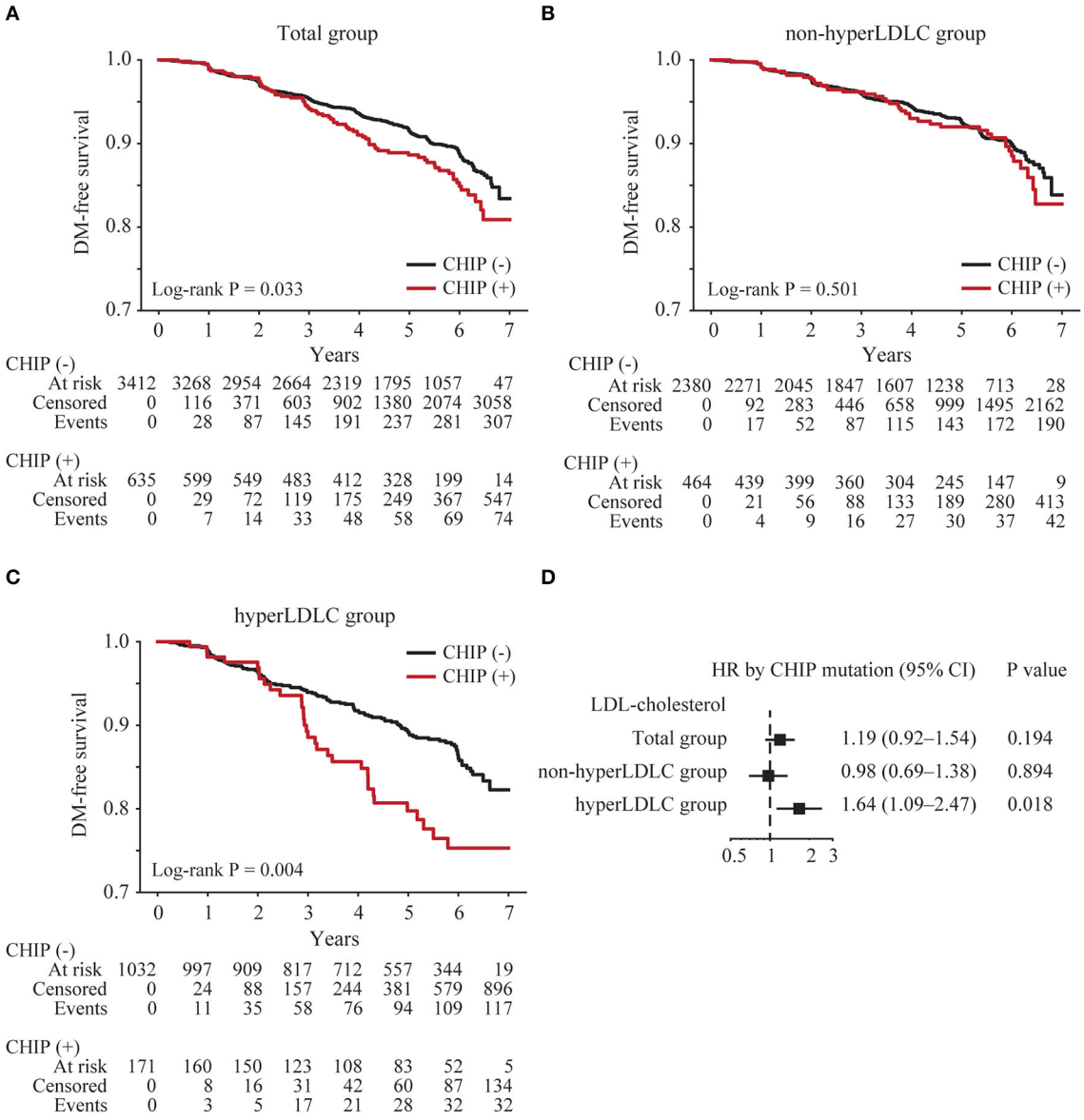

FIGURE 1
Risk of new-onset type 2 diabetes according to the presence of CHIP. Kaplan–Meier curves were plotted for all subjects (total group) (n = 4,047) **(A)** subjects with low LDL cholesterol levels (non-hyperLDLC group) **(B)**, and subjects with high LDL cholesterol levels (hyperLDLC group) **(C)**. The multivariate Cox regression model was analyzed adjusting for age, sex, BMI, and family history of diabetes in the total group, non-hyperLDLC group, and hyperLDLC group **(D)**.

Tet2 deficiency restricted to myeloid cells (Mye-Tet2-KO mice) compared with macrophages from control mice (24). In patients with heart failure, peripheral blood monocytes of patients carrying DNMT3A mutations have demonstrated a significant upregulation of inflammatory genes compared with those of patients without DNMT3A mutations (27). Type 2 diabetes is a chronic inflammatory disease (28). The infiltration and expansion of macrophage and the production of proinflammatory cytokines in several tissues, including the pancreatic islets, adipose tissue, muscle, and liver, contribute to the development of type 2 diabetes. Therefore, more inflammatory macrophages related to CHIP may be an underlying mechanism of the increased risk of new-onset type 2 diabetes.

4.3. The effect of the interaction between CHIP and high LDL cholesterol levels on the development of new-onset type 2 diabetes

In this study, the interaction between CHIP and high LDL cholesterol levels showed a synergistic effect on the development of new-onset type 2 diabetes. CHIP significantly increased the risk of new-onset type 2 diabetes in the hyperLDLC group (adjusted HR 1.64) but not in the non-hyperLDLC group (adjusted HR 0.98). The hyperLDLC group included 45% (540/1203) statin users. Statin use can increase the risk of diabetes, and a meta-analysis reported an odds ratio of 1.09 for new-onset type 2 diabetes (29, 30). Even after excluding statin users, CHIP still increased

TABLE 2 Subgroup analysis of the risk of new-onset type 2 diabetes based on CHIP status and LDL cholesterol level.

	New-onset type 2 diabetes (n = 377)	No event (n = 3,644)	Adjusted HR (95% CI)	p
Non-hyperLDLC group and non-carrier	188 (49.9)	2184 (59.9)	1.00 (reference)	
Non-hyperLDLC group and CHIP carrier	41 (10.9)	414 (11.4)	1.03 (0.73–1.45)	0.880
HyperLDLC group and non-carrier	116 (30.8)	908 (24.9)	1.32 (1.05–1.67)	0.020
HyperLDLC group and CHIP carrier	32 (8.5)	138 (3.8)	2.05 (1.40–3.00)	<0.001

CHIP, clonal hematopoiesis of indeterminate potential; HR, hazard ratio; CI, confidence interval.

the risk of new-onset type 2 diabetes (Supplementary Table S4). This result suggests that the synergistic effect of CHIP and LDL cholesterol on the development of new-onset type 2 diabetes was independent of statin use. The underlying hypothesis for the synergism of CHIP and high LDL cholesterol is complex. First, LDL cholesterol itself may have exacerbated inflammation (31), leading to the development of type 2 diabetes. In particular, oxidized LDL and small dense LDL are more proinflammatory and proatherogenic than naïve LDL (32, 33), and they have also been reported to be associated with new-onset type 2 diabetes (34). Oxidized LDL cholesterol activates the nucleotide-binding domain, leucine-rich-containing family, and pyrin domain-containing-3 (NLRP3) inflammasome in the cytoplasm of macrophages, and NLRP3 inflammasome leads to proinflammatory cytokines IL-1β release (35). Therefore, LDL cholesterol and CHIP may boost macrophage activation through the inflammasome, resulting in a synergistic effect on the development of type 2 diabetes. Second, high LDL cholesterol levels may stimulate the proliferation of hematopoietic stem cells with CHIP mutations and increase the number of inflammatory myeloid cells in peripheral blood. Increased cholesterol levels can promote the proliferation and mobilization of hematopoietic stem cells (36, 37). APOE-deficient ($Apoe^{-/-}$) or LDL receptor-deficient ($Ldlr^{-/-}$) mice fed high-fat diets showed proliferation of hematopoietic stem cells as well as leukocytosis in peripheral blood (38–40). These results suggest that high LDL cholesterol levels may potentiate the effects of CHIP on type 2 diabetes.

4.4. Limitations

This study has some limitations. Since our study did not evaluate inflammatory markers/cytokines, such as IL-6, oxidized LDL, small dense LDL, or insulin resistance, it was difficult to elucidate the mechanism by which CHIP interacts with LDL cholesterol to increase the risk of type 2 diabetes. Further studies are needed to determine the underlying mechanism of the association between CHIP and type 2 diabetes. The time and number of follow-up examinations were different for each subject due to the nature of this retrospective cohort study.

5. Conclusion

This is the first study to demonstrate that the presence of CHIP was significantly associated with the development of type 2 diabetes. In particular, CHIP and high LDL cholesterol levels synergistically increased the risk of new-onset type 2 diabetes. CHIP may be a hidden risk factor for type 2 diabetes.

Data availability statement

The datasets presented in this article are not readily available because of patent issues. Requests to access the datasets should be directed to HS (shany00@gmail.com).

Ethics statement

The studies involving human participants were reviewed and approved by Seoul National University Hospital Institutional Review Board. The patients/participants provided their written informed consent to participate in this study.

Author contributions

YK and S-YC designed the study. MK, HS, YK, HL, HP, JY, and S-YC collected, analyzed, and interpreted the data. MK and HS wrote the manuscript. YK, HL, HP, SC, JY, and S-YC contributed to the discussion and reviewed and edited the manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of interest

HS and YK are shareholders of Genome Opinion, Inc. The content of this study has been applied as a patent by HS and S-YC.

The remaining 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.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fpubh.2023.1181879/full#supplementary-material

References

1. American Diabetes Association Professional Practice C, American Diabetes Association Professional Practice C, Draznin B, Aroda VR, Bakris G, Benson G, et al. 2. Classification and diagnosis of diabetes: standards of medical care in diabetes-2022. *Diabetes Care.* (2022) 45:S17–38. doi: 10.2337/dc22-S002

2. Stern M, Williams K, Eddy D, Kahn R. Validation of prediction of diabetes by the Archimedes model and comparison with other predicting models. *Diabetes Care.* (2008) 31:1670–1. doi: 10.2337/dc08-0521

3. Abdul-Ghani MA, Abdul-Ghani T, Stern MP, Karavic J, Tuomi T, Bo I, et al. Two-step approach for the prediction of future type 2 diabetes risk. *Diabetes Care.* (2011) 34:2108–12. doi: 10.2337/dc10-2201

4. Raynor LA, Pankow JS, Duncan BB, Schmidt MI, Hoogeveen RC, Pereira MA, et al. Novel risk factors and the prediction of type 2 diabetes in the atherosclerosis risk in communities (ARIC) study. *Diabetes Care.* (2013) 36:70–6. doi: 10.2337/dc12-0609

5. Fuchsberger C, Flannick J, Teslovich TM, Mahajan A, Agarwala V, Gaulton KJ, et al. The genetic architecture of type 2 diabetes. *Nature.* (2016) 536:41–7. doi: 10.1038/nature18642

6. Mahajan A, Taliun D, Thurner M, Robertson NR, Torres JM, Rayner NW, et al. Fine-mapping type 2 diabetes loci to single-variant resolution using high-density imputation and islet-specific epigenome maps. *Nat Genet.* (2018) 50:1505–13. doi: 10.1038/s41588-018-0241-6

7. Jaiswal S, Ebert BL. Clonal hematopoiesis in human aging and disease. *Science.* (2019) 366:eaan4673. doi: 10.1126./science.aan4673

8. Jaiswal S, Natarajan P, Silver AJ, Gibson CJ, Bick AG, Shvartz E, et al. Clonal Hematopoiesis and Risk of Atherosclerotic Cardiovascular Disease. *N Engl J Med.* (2017) 377:111–21. doi: 10.1056/NEJMoa1701719

9. Bhattacharya R, Zekavat SM, Haessler J, Fornage M, Raffield L, Uddin MM, et al. Clonal hematopoiesis is associated with higher risk of stroke. *Stroke.* (2022) 53:788–97. doi: 10.1161/STROKEAHA.121.037388

10. Libby P, Buring JE, Badimon L, Hansson GK, Deanfield J, Bittencourt MS, et al. Atherosclerosis. *Nat Rev Dis Primers.* (2019) 5:56. doi: 10.1038/s41572-019-0106-z

11. Amoros-Perez M, Fuster JJ. Clonal hematopoiesis driven by somatic mutations: A new player in atherosclerotic cardiovascular disease. *Atherosclerosis.* (2020) 297:120–6. doi: 10.1016/j.atherosclerosis.02, 008.

12. Castillo JJ, Mull N, Reagan JL, Nemr S, Mitri J. Increased incidence of non-Hodgkin lymphoma, leukemia, and myeloma in patients with diabetes mellitus type 2: a meta-analysis of observational studies. *Blood.* (2012) 119:4845–50. doi: 10.1182/blood-2011-06-362830

13. Bonnefond A, Skrobek B, Lobbens S, Eury E, Thuillier D, Cauchi S, et al. Association between large detectable clonal mosaicism and type 2 diabetes with vascular complications. *Nat Genet.* (2013) 45:1040–3. doi: 10.1038/ng.2700

14. Jaiswal S, Fontanillas P, Flannick J, Manning A, Grauman PV, Mar BG, et al. Age-related clonal hematopoiesis associated with adverse outcomes. *N Engl J Med.* (2014) 371:2488–98. doi: 10.1056/NEJMoa1408617

15. Lee C, Choe EK, Choi JM, Hwang Y, Lee Y, Park B, et al. Health and Prevention Enhancement (H-PEACE): a retrospective, population-based cohort study conducted at the Seoul National University Hospital Gangnam Center, Korea. *BMJ Open.* (2018) 8:e019327. doi: 10.1136/bmjopen-2017-019327

16. Coombs CC, Zehir A, Devlin SM, Kishtagari A, Syed A, Jonsson P, et al. Therapy-related clonal hematopoiesis in patients with non-hematologic cancers is common and associated with adverse clinical outcomes. *Cell Stem Cell.* (2017) 21:374–382. doi: 10.1016/j.stem.07.010

17. Gillis NK, Ball M, Zhang Q, Ma Z, Zhao Y, Yoder SJ, et al. Clonal haemopoiesis and therapy-related myeloid malignancies in elderly patients: a proof-of-concept, case-control study. *Lancet Oncol.* (2017) 18:112–21. doi: 10.1016/S1470-2045(16)30627-1

18. Takahashi K, Wang F, Kantarjian H, Doss D, Khanna K, Thompson E, et al. Preleukaemic clonal haemopoiesis and risk of therapy-related myeloid neoplasms: a case-control study. *Lancet Oncol.* (2017) 18:100–11. doi: 10.1016/S1470-2045(16)30626-X

19. Bolton KL, Koh Y, Foote MB, Im H, Jee J, Sun CH, et al. Clonal hematopoiesis is associated with risk of severe Covid-19. *Nat Commun.* (2021) 12:5975. doi: 10.1038/s41467-021-26138-6

20. Grundy SM, Stone NJ, Bailey AL, Beam C, Birtcher KK, Blumenthal RS. 2018 AHA/ACC/AACVPR/AAPA/ABC/ACPM/ADA/AGS/APhA/ASPC/NLA/PCNA guideline on the management of blood cholesterol: executive summary: a report of the american college of cardiology/american heart association task force on clinical practice guidelines. *Circulation.* (2019) 139:e1046–81. doi: 10.1161/CIR.0000000000000624

21. National Cholesterol Education Program (NCEP). Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. *Circulation.* (2002) 106:3143–421. doi: 10.1161/circ.106.25.3143

22. Gumuser ED, Schuermans A, Cho SMJ, Sporn ZA, Uddin MM, Paruchuri K, et al. Clonal hematopoiesis of indeterminate potential predicts adverse outcomes in patients with atherosclerotic cardiovascular disease. *J Am Coll Cardiol.* (2023) 81:1996–2009. doi: 10.1016/j.jacc.03401

23. Bick AG, Weinstock JS, Nandakumar SK, Fulco CP, Bao EL, Zekavat SM, et al. Inherited causes of clonal haematopoiesis in 97,691 whole genomes. *Nature.* (2020) 586:763–8. doi: 10.1038/s41586-020-2819-2

24. Fuster JJ, Maclauchlan S, Zuriaga MA, Polackal MN, Ostriker AC, Chakraborty R, et al. Clonal hematopoiesis associated with TET2 deficiency accelerates atherosclerosis development in mice. *Science.* (2017) 355:842–7. doi: 10.1126/science.aag1381

25. Fuster JJ, Zuriaga MA, Zorita V, Maclauchlan S, Polackal MN, Viana-Huete V, et al. TET2-loss-of-function-driven clonal hematopoiesis exacerbates experimental insulin resistance in aging and obesity. *Cell Rep.* (2020) 33:108326. doi: 10.1016/j.celrep.2020.108326

26. Sano S, Oshima K, Wang Y, Katanasaka Y, Sano M, Walsh K, et al. CRISPR-mediated gene editing to assess the roles of Tet2 and Dnmt3a in clonal hematopoiesis and cardiovascular disease. *Circ Res.* (2018) 123:335–41. doi: 10.1161/CIRCRESAHA.118.313225

27. Abplanalp WT, Cremer S, John D, Hoffmann J, Schuhmacher B, Merten M, et al. Clonal hematopoiesis-driver DNMT3A mutations alter immune cells in heart failure. *Circ Res.* (2021) 128:216–28. doi: 10.1161/CIRCRESAHA.120.317104

28. Donath MY, Shoelson SE. Type 2 diabetes as an inflammatory disease. *Nat Rev Immunol.* (2011) 11:98–107. doi: 10.1038/nri2925

29. Rajpathak SN, Kumbhani DJ, Crandall J, Barzilai N, Alderman M, Ridker PM, et al. Statin therapy and risk of developing type 2 diabetes: a meta-analysis. *Diabetes Care.* (2009) 32:1924–9. doi: 10.2337/dc09-0738

30. Sattar N, Preiss D, Murray HM, Welsh P, Buckley BM, de Craen AJ, et al. Statins and risk of incident diabetes: a collaborative meta-analysis of randomised statin trials. *Lancet.* (2010) 375:735–742.doi: 10.1016/S0140-6736(09)61965-6

31. Jukema RA, Ahmed TA, Tardif JC. Does low-density lipoprotein cholesterol induce inflammation? If so, does it matter? Current insights and future perspectives for novel therapies. *BMC Med*. (2019) 17:197.doi: 10.1186/s12916-019-1433-3

32. Steinberg D. Low density lipoprotein oxidation and its pathobiological significance. *J Biol Chem*. (1997) 272:20963–6. doi: 10.1074/jbc.272.34.20963

33. Ikezaki H, Lim E, Cupples LA, Liu CT, Asztalos BF, Schaefer EJ, et al. Small dense low-density lipoprotein cholesterol is the most atherogenic lipoprotein parameter in the prospective framingham offspring study. *J Am Heart Assoc*. (2021) 10:e019140. doi: 10.1161/JAHA.120.019140

34. Hoogeveen RC, Ballantyne CM, Bang H, Heiss G, Duncan BB, Folsom AR, et al. Circulating oxidised low-density lipoprotein and intercellular adhesion molecule-1 and risk of type 2 diabetes mellitus: the atherosclerosis risk in communities study. *Diabetologia*. (2007) 50:36–42. doi: 10.1007/s00125-006-0533-8

35. Duewell P, Kono H, Rayner KJ, Sirois CM, Vladimer G, Bauernfeind FG, et al. NLRP3 inflammasomes are required for atherogenesis and activated by cholesterol crystals. *Nature*. (2010) 464:1357–61. doi: 10.1038/nature08938

36. Oguro H. The roles of cholesterol and its metabolites in normal and malignant hematopoiesis. *Front Endocrinol*. (2019) 10:204. doi: 10.3389/fendo.2019.00204

37. Morgan PK, Fang L, Lancaster GI, Murphy AJ. Hematopoiesis is regulated by cholesterol efflux pathways and lipid rafts: connections with cardiovascular diseases. *J Lipid Res*. (2020) 61:667–75. doi: 10.1194/jlr.TR119000267

38. Murphy AJ, Akhtari M, Tolani S, Pagler T, Bijl N, Kuo CL, et al. ApoE regulates hematopoietic stem cell proliferation, monocytosis, and monocyte accumulation in atherosclerotic lesions in mice. *J Clin Invest*. (2011) 121:4138–49. doi: 10.1172/JCI57559

39. Feng Y, Schouteden S, Geenens R, Van Duppen V, Herijgers P, Holvoet P, et al. Hematopoietic stem/progenitor cell proliferation and differentiation is differentially regulated by high-density and low-density lipoproteins in mice. *PLoS One*. (2012) 7:e47286. doi: 10.1371/journal.pone.0047286

40. Seijkens T, Hoeksema MA, Beckers L, Smeets E, Meiler S, Levels J, et al. Hypercholesterolemia-induced priming of hematopoietic stem and progenitor cells aggravates atherosclerosis. *FASEB J*. (2014) 28:2202–13. doi: 10.1096/fj.13-243105

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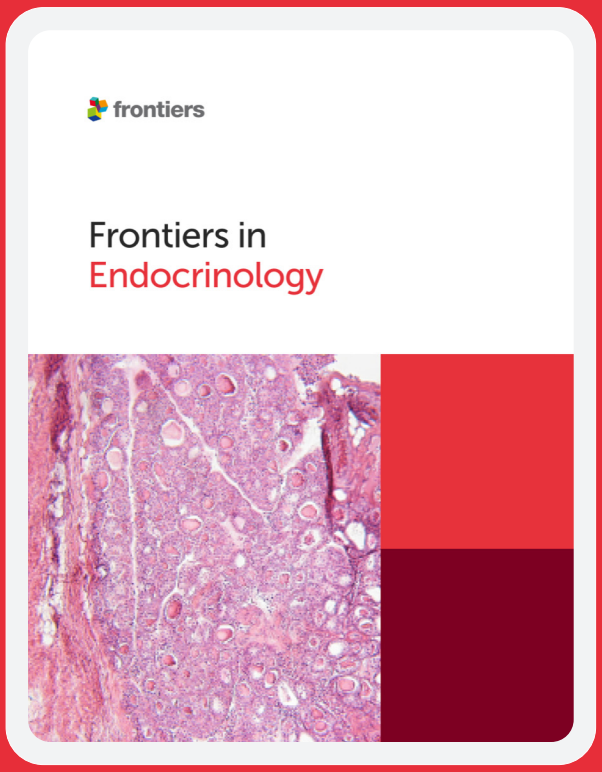

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