frontiers | Frontiers in Environmental Science

TYPE Original Research
PUBLISHED 25 November 2022
DOI 10.3389/fenvs.2022.1046759

OPEN ACCESS

EDITED BY
Xiaobin Liao,
Huaqiao University, China

REVIEWED BY
Jin Li,
Qingdao University, China
Kenji Furukawa,
Kumamoto University, Japan

*CORRESPONDENCE
Shou-Qing Ni,
sqni@sdu.edu.cn

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

SPECIALTY SECTION
This article was submitted to Water and Wastewater Management, a section of the journal Frontiers in Environmental Science

RECEIVED 17 September 2022
ACCEPTED 17 November 2022
PUBLISHED 25 November 2022

CITATION
Wang Z, Liu P, Zhou J, Ismail S, Ahmad S, Awad HM and Ni S-Q (2022), Low-intensity electromagnetic field as a new engineering oriented bioaugmentation strategy for anammox granules. *Front. Environ. Sci.* 10:1046759.
doi: 10.3389/fenvs.2022.1046759

Low-intensity electromagnetic field as a new engineering oriented bioaugmentation strategy for anammox granules

Zhibin Wang[1,2†], Pengpeng Liu[1†], Jing Zhou[1], Sherif Ismail[1,3], Shakeel Ahmad[4], Hanem M. Awad[5] and Shou-Qing Ni[1*]

[1]Shenzhen Research Institute of Shandong University, School of Environmental Science and Engineering, Shandong University, Jinan, China, [2]School of Life Sciences, Shandong University, Qingdao, Shandong, China, [3]Environmental Engineering Department, Zagazig University, Zagazig, Egypt, [4]Department of Soil and Environmental Sciences, Muhammad Nawaz Shareef University of Agriculture, Multan, Pakistan, [5]National Research Centre, Tanning Materials and Proteins Department, Dokki, Giza, Egypt

Improving the relative abundance of bacteria and their activity is still the basis for the efficient operation of anammox process. Here, biomagnetic effect was used to promote anammox granules. Batch test results show that the application of an electromagnetic field (EMF) with a strength of 0.09 μT increased the nitrogen removal performance of anammox by 32.44% while higher strength EMF of 0.20 and 0.25 μT inhibited the activity of anammox bacteria. Long-term experiment indicated that the addition of EMF with a strength of 0.09 μT greatly improved nitrogen removal performance of the granular sludge, especially the total nitrogen removal performance increased by 15.3%. After 120 days of reactor operation, the nitrogen loading rate was increased to 6.4 kg N/m^3/d, and the total nitrogen removal rate of the reactors with and without EMF addition reached 4.92 kg N/m^3/d and 4.25 kg N/m^3/d, respectively. Throughout the experiment, the removal rate of NH_4^+-N and NO_2^--N of anammox reactor with 0.09 μT EMT addition was always higher than that without EMF addition. The high-throughput sequencing analysis showed that the proportion of *Candidatus Brocadia* in reactors with and without EMF addition were 21.3% and 15.8%, respectively. The application of EMF with an intensity of 0.09 μT increased the relative abundance of the main anammox bacteria. 70 kos were enriched under EMF conditions, including ko00780 (Biotin metabolism), ko00540 (Lipopolysaccharide biosynthesis), ko00590 (Arachidonic acid metabolism). 51 kos like ko03030 (DNA replication) decreased after EMF addition. This study demonstrates the feasibility of EMF to promote anammox and expands the application of EMF in wastewater treatment.

KEYWORDS

anammox, EMF, enhancement, high-throughput sequencing, anammox granule

Introduction

Anammox is a new type of biological nitrogen removal process and the potential core technology of mainstream wastewater treatment processes in the future. Due to its huge economic benefits, relevant scholars conducted comprehensive researches to speed up the application of this emerging technology in actual engineering wastewater treatment (Li et al., 2018; Du et al., 2019; Trinh et al., 2021). However, improving the relative abundance of bacteria and their activity is still the basis for the efficient operation of anammox process.

Many physiochemical methods were used to promote the activity of anammox and increase anammox bacteria purity, for example, the anaerobic sludge was activated in an external electric and magnetic field (Qu et al., 2020; Madondo et al., 2022). Anammox activity was increased and anammox start-up time has been shortened with electric field addition (Zhang et al., 2019a; Li et al., 2022) or static magnetic field addition (Yavuz and Çelebi, 2003). Intermediate frequency of pulsed electric field could dramatically enhance anammox activity and granule sludge stability *via* stimulating the secretion of extracellular polymeric substances (Zhang et al., 2019b). After the exposure to suspended MnO_2 nanoparticles, specific anammox activity (SAA) was enhanced (Xu et al., 2019). Graphene oxide concentration in the range of 0.05–0.1 g/L can increase the activity of anammox bacteria and the secretion of extracellular polymers (Wang et al., 2013).

The biomagnetic effect refers to the weak biomagnetic field generated by all life activities of organisms (including humans), such as growth, reproduction, and metabolism (Paksy et al., 2000; Cifra et al., 2011; Redlarski et al., 2015). In recent years, biomagnetic effect has been introduced into the field of environmental wastewater treatment. Applying infrared electromagnetic field (EMF) shortened the process startup time by 37%, greatly saved the time cost of domestication, and showed better stability (Wang et al., 2021a). Especially, low-frequency EMFs may affect numerous cell functions at cellular level, such as RNA transcription, DNA synthesis, protein expression, hormone production and metabolic activity (Goodman et al., 1983; Goodman and Henderson, 1988; Litovitz et al., 1994). Certain strength of EMF can stimulate the reproduction and activity of bacteria. For example, static EMFs could improve the NH_4^+-N removal rate of partial nitrification by promote the growth activity of ammonium oxidizing bacteria, and the impact is immediate, reversible, and lasting (Wang et al., 2017; Beretta et al., 2019).

In general, to solve the obstacle of long start-up time of anammox process, this paper adopted EMF to stimulate the enrichment of anammox granules. The rapid enrichment of anammox bacteria was realized; process performance and the stability of the reactor were improved after the addition of electromagnetic waves. This study may provide technical guidance for the feasibility of anammox promotion *via* EMF strategy and expand its application in wastewater treatment.

Materials and methods

Seed sludge and synthetic wastewater

Reactor's inoculation sludge was anammox granules from an UASB that has been operated for 2 years in the laboratory. Particle size of the granular sludge was around 2.0–2.4 mm. The inoculation biomass occupied up to one-third of the reactor's working volume, leading to an initial MLVSS concentration of 8,000 ± 20 mg/L. The medium used in the study contained: $(NH_4)_2SO_4$, $NaNO_2$, $KHCO_3$ (0.5 g/L), KH_2PO_4, $MgSO_4{\cdot}7H_2O$, $CaCl_2{\cdot}2H_2O$ and trace element solution (Wang et al., 2021b). The synthetic low-strength wastewater was fed into the reactor at the pH of 7.5 ± 0.2. The temperature of each reactor was maintained at approximately 28.0 ± 1.0°C. Aeration with high purity nitrogen gas was used 10 min every day to ensure an anaerobic environment. The DO concentration in the reactor was controlled below 0.2 mg/L.

Batch experiment

In order to determine the appropriate EMF strength for anammox granular sludge, six UASB reactors with a working volume of 450 ml were used for anammox culture. The six reactors were treated with 0, 0.06, 0.09, 0.12, 0.20, and 0.25 μT EMF, which were described as r_0, $r_{0.06}$, $r_{0.09}$, $r_{0.12}$, $r_{0.20}$, and $r_{0.25}$, respectively. EMF was provided by a modified specific therapy device with a wavelength of 2–25 μm and a frequency of 50 Hz, which was purchased from Chongqing Aerospace Rocket Electronic Technology Co., Ltd. The concentration of NH_4^+-N and NO_2^--N were 100 mg/L and 130 mg/L, respectively.

Long-term experiment and operational strategy

Long-term experiments were conducted in two parallel laboratory-scale fluidized bed biofilm reactors. R1 was the control reactor, and R2 was treated with 0.09 μT EMF. The maximum working volume of each reactor is 1.2 L. At first, the concentration of NH_4^+-N and NO_2^--N were 50 mg/L and 65 mg/L, respectively. The concentration of NH_4^+-N and NO_2^--N were increased to 100 mg/L and 130 mg/L, respectively at the 60th day. The nitrogen loading rate (NLR) was increased by reducing the hydraulic retention time (HRT) from 3 days to 1.5 and 0.75 days. R2 was irradiated with EMF device for 2 h per day to provide an intensity of 0.09 μT EMF.

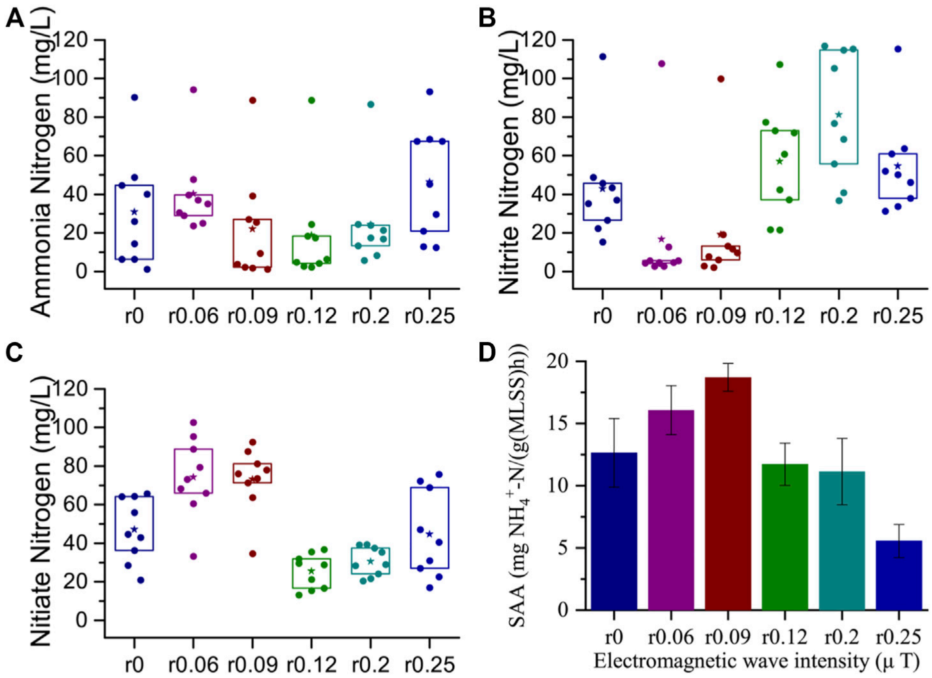

FIGURE 1
Relationships of intensity of EMF and nitrogen concentration during reactor start-up. **(A–C)** Nitrogen concentration detected under different EMF intensity. The dots represent the nine detection values within 18 days, the stars represent the average data of the nine measured values, and the boxes show the data range from 25% to 75%. **(D)** Specific anammox activity under different EMF intensity.

Analytical methods

The concentration of nitrogen compounds (including NH_4^+-N, NO_2^--N and NO_3^--N) and volatile suspended solids (VSS) of the sludge were measured according to the standard methods (Apha Awwa, 2005). The pH and DO were measured using a digital portable pH meter and a digital portable DO instrument (HQ40d, United States). The EMW intensity was measured by an electromagnetic radiation tester (LZT-1160, China).

DNA extraction, high-throughput sequencing and data analysis

In order to verify the long-term effect of low-frequency EMF on anammox process, seed sludge (R0), sludge in reactor with and without additional EMF (described as R1 and R2) were sampled for DNA extraction. Total DNA was extracted from 1 ml of reactor sludge using Power Soil DNA Isolation Kit (MO BIO Laboratories, United States) according to the manufacturer's recommendations. The primers (forward primer: GCCGTAAAC GATGGGCACT, reverse primer: AACGTCTCACGACACGAG CTG) were used to PCR amplify V4 of 16S rRNA from microbial genomic DNA (Tsushima et al., 2007). High-throughput sequencing of samples was performed on the Illumina Miseq platform. The closest bacterial communities (similarities > 97%) were gathered into sequencing reads to form operational taxonomies (OTU).

Results

Determination of preferable electromagnetic field strength for anammox consortia

Six different intensities of EMF were used in the short-term test to determine the optimal EMF intensity for anammox process. As shown in Figure 1, low-intensity EMF could significantly enhance the activity of anammox bacteria. Especially, with 0.09 μT EMF addition, the removal efficiencies of NH_4^+-N and NO_2^--N were significantly increased. When EMF intensity was below 0.12 μT, EMF addition showed higher nitrogen removal performance than the control group and 0.09 μT was the best. However, EMF with intensity of 0.2 and 0.25 μT has negative effect on anammox reaction. This trend was clearer with respect to the SAA analysis. This is similar to Coey's theory of a magnetized water treatment mechanism based on the gradient of the magnetic field intensity rather than the absolute intensity (Fox, 2011).

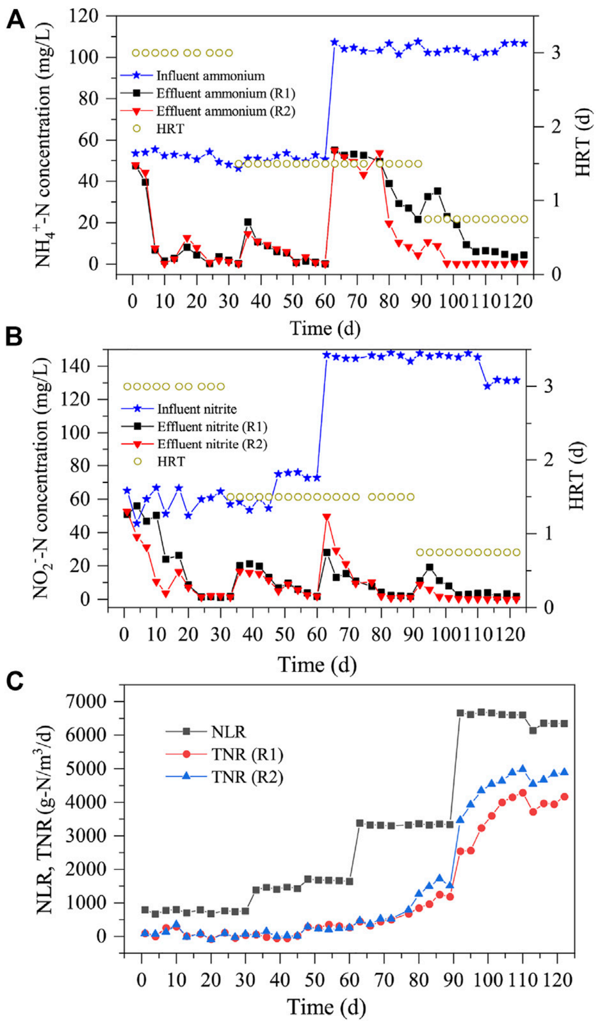

FIGURE 2
Long-term reactors performance. **(A)** NH_4^+-N, **(B)** NO_2^--N, **(C)** Nitrogen loading rate (NLR) and total nitrogen removal rate (TNR).

Long-term effects of electromagnetic field on anammox granules

To verify the long-term effects of EMF on granular anammox process, two parallel laboratory-scale UASBs were operated. As shown in Figure 2, from the third day, the concentration of NH_4^+-N and NO_2^--N in the effluent of reactors begin to decrease, and the performance of R2 was better than the control reactor (R1). From the 12th day, the effluent concentration of NH_4^+-N and NO_2^--N in R2 was almost 0 due to the stimulated metabolism of anammox bacteria by EMF. R1 took another 10 days to reach similar NO_2^--N removal. On the 30th day, the total nitrogen removal rates of the two reactors R1 and R2 were 71.9 and 88.9 mg N/m^3/d, respectively. It can be seen that applying 0.09 μT EMF slightly shortened the start-up time and enhanced the total nitrogen removal rate of the reactor.

After 20 days, two anammox reactors were started up successfully. To study the stability of the reactor performance by applying EMFs, HRT was shortened from 3 days to 1.5 days to increase nitrogen load on day 30. Fluctuation of NO_2^--N removal appeared immediately in both reactors and after 20 days, they became stable again.

On the 60th day, the influent NH_4^+-N and NO_2^--N concentrations were adjusted from 50 mg N/L and 65 mg N/L to 100 mg N/L and 130 mg N/L, respectively. Correspondingly, the nitrogen loading rate (NLR) was increased from 1.6 kg N/m^3/d to 3.2 kg N/m^3/d. The adaption performance of R2 was superior to R1, especially NH_4^+-N removal. With the operation of the reactor, the nitrogen removal rate of the two reactors showed a gradually increasing trend, and after the 80th day, R2 exhibited better nitrogen removal performance.

On the 90th day, HRT was adjusted from 1.5 days to 0.75 day, so that the NLR of each reactor was increased from 3.2 kg N/m^3/d to 6.4 kg N/m^3/d. R1 showed dramatic fluctuations in response to the increase of nitrogen load. The sudden increase of nitrogen loading inhibited anammox bacterial activity. The effluent water quality of reactor R1 deteriorated rapidly, and the effluent NH_4^+-N concentration was 31.3 mg N/L, while the effluent NH_4^+-N and NO_2^--N concentration of reactor R2 was less than 15 mg N/L. R1 took about 15 days to fully recover, while R2 only spent 8 days to restore. The stability of reactor R2 to nitrogen loading was better than that of R1. The total nitrogen removal rate (TNR) of R1 and R2 on the 110th day reached 4.25 kg N/m^3/d and 4.92 kg N/m^3/d, respectively.

Effect of electromagnetic field on the content of functional bacteria

To study the effect of EMF on anammox bacteria in anammox granular sludge, the copy number of AnAOB 16S rRNA gene (AMX) in different stages of the two reactors was quantitatively analyzed. The results are shown in Figure 3. As the reactor run, the copy number of AMX gradually increased in the first 60 days, and then stabilized or even decreased slightly. This may be due to the competition with denitrifying bacteria. At the beginning, the copy amount of AMX in R1 and R2 was almost the same, 1.09×10^7 and 1.36×10^7 copies/ng DNA, respectively. After 60 days of cultivation, the copy amount of AMX in R1 and R2 was 2.12×10^7 and 2.87×10^7 copies/ng DNA, respectively. Compared with R1, the copy number of AMX gene in R2 increased by 35.4%. On day 120, the copies of AMX in R1 and R2 were 1.67×10^7 and 1.95×10^7 copies/ng DNA, respectively (Figure 3A). Obviously, EMF can significantly promote the enrichment and reproduction of anammox bacteria in granular sludge.

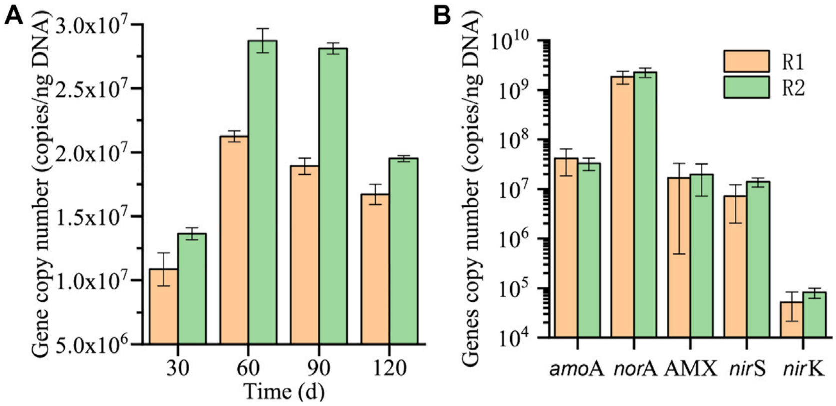

FIGURE 3
The qPCR results. **(A)** AMX copy numbers in different period, **(B)** functional gene copy numbers on day 120.

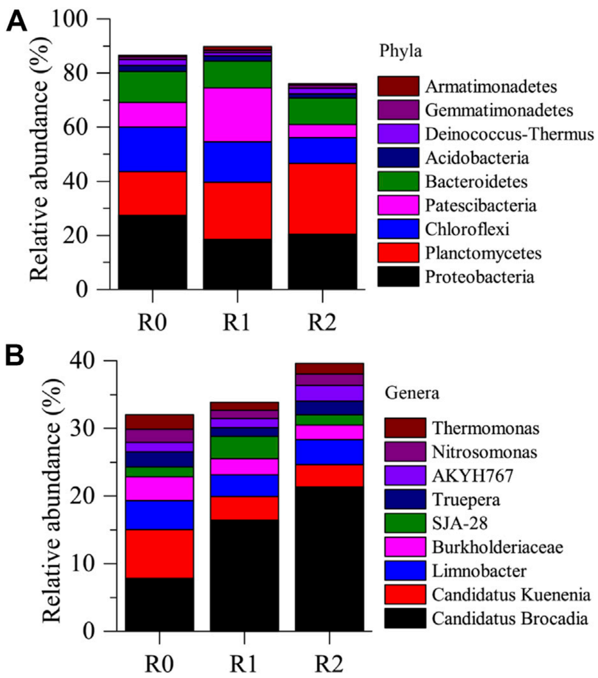

FIGURE 4
Taxonomic classification of the bacterial communities in R1 and R2 at phylum **(A)** and genus **(B)** levels.

In addition, effect of EMF on the content of nitrogen metabolic genes was studied. On the 120th day, the content of each functional gene of *amo*A, *nor*A, *nir*S and *nir*K was quantitatively analyzed. As shown in Figure 3B, the gene copy numbers of *nor*A in R1 and R2 are 1.85×10^9 and 2.27×10^9 copies/ng DNA, respectively, and the *nor*A gene content in R2 was significantly higher than that in R1. It is inferred that EMF may promote the reproduction of NOB to a certain extent, and even the degree of influence is higher than that of anammox bacteria (Liu et al., 2008; Benyoucef et al., 2021; Ni et al., 2021).

Effect of electromagnetic field on the bacterial community of anammox granules

According to the Chao1 index, the bacterial species richness of R1 and R2 were 832.93, and 801.21, respectively. The Shannon index of diversity also indicated that the application of EMF decreased bacterial diversity from 6.33 for R1 to 5.49 for R2. Figure 4 show the changes of granular sludge community under different treatment conditions at phylum and genus levels. At phylum level, the relative abundance of Proteobacteria in R1 and R2 are 18.6% and 20.5%, respectively. The relative abundance of Planctomycetes, where anammox bacteria are located, increased from 21.1% in R1 to 26.1% in R2. Planctomycetes is the dominated phylum in R2 thanks to the EMF. The irradiation of EMF significantly increased the relative abundance of Planctomycetes in the community. This reveals that electromagnetic fields directly affected the balance of microbial communities (Ratushnyak et al., 2008).

At genus level, *Candidatus Brocadia* dominated the composition of anammox bacteria in two sludge samples. The proportion of *C. Brocadia* in reactor R2 is 21.3%, while the proportions in R1 is 16.4%. Obviously, the application of EMF increased the main anammox genus *C. Brocadia* in R2.

The PICRUSt technique was used to predict the effect of EMF on community function (KEGG Orthology, KO). There is a total of 121 KOs with an average relative abundance above 1%, and

70 of these 121 KO pathways were enriched under EMF conditions, including ko00780 Biotin metabolism, ko00540 Lipopolysaccharide biosynthesis, ko00590 Arachidonic acid metabolism, ko00730 Thiamine metabolism, ko02030 Bacterial chemotaxis, ko00785 Lipoic acid metabolism, ko00020 Citrate cycle et al. The relative abundance of 51 KOs decreased under EMF conditions, including ko00290 Valine, leucine and isoleucine biosynthesis, ko03030 DNA replication, ko00740 Riboflavin metabolism, ko03430 Mismatch repair, ko00030 Pentose phosphate pathway et al. (Supplementary Table S1).

Discussion

Electromagnetic field stimulated anammox activities and anammox abundance of anammox granules

Under proper strength, EMF can increase the activity of anammox bacteria. Otherwise, the performance of anammox bacteria may be reduced. In this study, the best EMF strength is 0.09 μT. Within a certain range, the activity of anammox granular sludge increased with the increase of the EMF strength, and the intensity beyond the best strength will inhibit anammox activity.

The q-PCR experiment showed that the copy number of AMX gene after 30 days of EMF irradiation was 35.4% higher than that of the control. The EMF can significantly promote the enrichment and reproduction of anammox bacteria in anammox granular sludge. On the 120th day, the number of *nor*A gene copies in the two reactors reached 10^9, indicating that the number of nitrite-oxidizing bacteria (NOB) was much higher than that of anammox bacteria. It is clear that the EMF may also promote the reproduction of NOB, even the degree of impact is higher than that of anammox bacteria (Wang et al., 2021a).

Biological mechanism of electromagnetic field on anammox granular sludge

The target of EMF can be water, cell plasma membrane and genome. Under the EMF irradiation, the structure of the soft clusters of water molecules was changed and activated, thereby enhancing the metabolic function of the organism and improving the function of the organism (Tekutskaya et al., 2015). The effect of EMF on ion migration is mediated by direct or indirect effects on cell membranes (Dihel et al., 1985). A molecular dynamics simulation was carried out to study the influence of external EMF on the NaCl electrolyte aqueous solution at a temperature of 25°C. When the electromagnetic wave changed at a frequency of 50, 100 or 200 GHz and the intensity of the EMF changed within the range of 0.1–0.3 V/Å, the diffusion coefficients of water molecules and ions increased with the increase of the frequency and intensity of the EMF (Chang and Weng, 2008).

Studies have shown that through vertically polarized magnetic media, surface biomagnetism can affect bacterial adhesion and protein synthesis, thereby achieving the purpose of improving bacterial activity (Chua and Yeo, 2005). After 15 min of exposure to EMF the specific growth rate of *Rhodobacter sphaeroides* grown under anaerobic conditions increased by 1.2 times (Soghomonyan et al., 2016). This difference is caused by the direct effect of radiation on membrane transfer and bacterial enzymes. Trushin attributed the EMF to microbial biological communication (Trushin, 2003). The EMF acted on bacterial populations during the formation and display of biofilms, and *Helicobacter pylori* showed obvious differences in cell viability (Di Campli et al., 2010). The biofilm secreted by bacteria has a protective function for bacteria. Biofilms provide a place for bacteria to resist drastic changes in habitat and facilitate the formation of communities. (Adams et al., 2003; Jain et al., 2007).

To study the biological mechanism of EMF on anammox granular sludge, this article focuses on the cultivation of anammox bacteria, the evolution of microbial communities and the expression of functional genes. Genes related to Biotin metabolism, Lipopolysaccharide biosynthesis, Arachidonic acid metabolism, Thiamine metabolism, Bacterial chemotaxis, Lipoic acid metabolism, Citrate cycle et al. were enriched under EMF conditions; and genes related to Valine, leucine and isoleucine biosynthesis, DNA replication, Riboflavin metabolism, Mismatch repair, Pentose phosphate pathway et al. were decreased under EMF conditions. This may be the reason why EMF irradiation shortened the start-up time of anammox process.

Practical application of electromagnetic field-assisted anammox system

In this study, the total nitrogen removal rate of anammox reactor after EMW irradiation was increased by 15.8%. And the relative concentration of anammox bacteria was increased from 16.4% to 21.3% under EMW environment. Therefore, adding EMW to promote anammox process is an effective method. In order to better apply EMW to the actual anammox reactor, EMW permeable pipes need to be pre-installed inside the reactor, and EMW generators need pre-installed inside the pipes. By adjusting the number of EMW generators, electromagnetic wave intensity, aeration intensity and sludge stirring speed, the anammox sludge can be uniformly and effectively exposed to EMW. Since low EMW intensities can significantly increase the efficiency of the anammox process, and only 2–3 h of EMW addition per day can increase the anammox efficiency, the increase of operating costs is limited. And the construction and operation cost of EMW system is relatively low, therefore, adding EMW is an economical method to promote anammox process.

Conclusion

Both reactor performance and SAA indicated that proper EMF (0.09 μT) promoted the activity of anammox granules, while high EMF density (i.e., 0.25 μT) deteriorated anammox reaction. The nitrogen removal performance of reactor with 0.09 μT EMF addition has been greatly improved. When the nitrogen loading rate was increased to 6.4 kg $N/m^3/d$, the total nitrogen removal rate reactors with and without EMF addition reached 4.25 kg $N/m^3/d$ and 4.92 kg $N/m^3/d$, respectively. Environmental diversity sequencing analysis showed that EMF can promote the enrichment of anammox bacteria, especially the relative abundance of *C. Brocadia* and *Candidatus Kuenenia*. In particular, the proportion of *C. Brocadia* in R2 was 21.3%, and the proportion in R1 was 16.4%. According to the prediction results of microbial community function, EMF can directly enhance the level of physiological metabolism. In summary, low-intensity EMF provides a new engineering oriented bioaugmentation strategy for anammox granules.

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.

Author contributions

ZW and PL performed the majority of the experiments and data interpretation. JZ and SI helped in experiments. ZW wrote the manuscript. SA and HA revised the manuscript. S-QN designed and supervised the study.

Acknowledgments

The authors gratefully acknowledge the support from the Shenzhen Science and Technology Program (JCYJ20220530141203007), National Natural Science Foundation of China (22076100), Young Scholars Program of Shandong University (2020QNQT012), Taishan Scholar Youth Expert Program of Shandong Province (tsqn201909005), Key Research and Developmental Program of Shandong Province (2021CXGC011202, 2022TZXD0044, 2022CXGC021002), Qingdao Science and Technology Huimin Demonstration Guide Project (20-3-4-4-nsh), Jinan Science and Technology Research leader studio Project (2020GXRC054) and thanks to Shandong Province Postdoctoral Innovative Talents Support Program (202101003).

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/fenvs.2022.1046759/full#supplementary-material

References

Adams, B., Bates, T., and Oliver, J. (2003). Survival of *Helicobacter pylori* in a natural freshwater environment. *Appl. Environ. Microbiol.* 69 (12), 7462–7466. doi:10.1128/aem.69.12.7462-7466.2003

APHA, AWWA, WEF. (2005). Standard methods for the examination of water and wastewater. American Public Health Association: Washington, DC.

Benyoucef, N., Pauss, A., Abdi, N., Sarde, C.-O., Grib, H., and Mameri, N. (2021). Enhancement of the denitrification performance of an activated sludge using an electromagnetic field in batch mode. *Chemosphere* 262, 127698. doi:10.1016/j.chemosphere.2020.127698

Beretta, G., Mastorgio, A. F., Pedrali, L., Saponaro, S., and Sezenna, E. (2019). The effects of electric, magnetic and electromagnetic fields on microorganisms in the perspective of bioremediation. *Rev. Environ. Sci. Biotechnol.* 18 (1), 29–75. doi:10.1007/s11157-018-09491-9

Chang, K.-T., and Weng, C.-I. (2008). Properties of aqueous NaCl solutions in external electromagnetic fields. *Mol. Phys.* 106 (21-23), 2515–2524. doi:10.1080/00268970802577826

Chua, L.-Y., and Yeo, S.-H. (2005). Surface bio-magnetism on bacterial cells adhesion and surface proteins secretion. *Colloids Surfaces B Biointerfaces* 40 (1), 45–49. doi:10.1016/j.colsurfb.2004.10.004

Cifra, M., Fields, J. Z., and Farhadi, A. (2011). Electromagnetic cellular interactions. *Prog. biophysics Mol. Biol.* 105 (3), 223–246. doi:10.1016/j.pbiomolbio.2010.07.003

Di Campli, E., Di Bartolomeo, S., Grande, R., Di Giulio, M., and Cellini, L. (2010). Effects of extremely low-frequency electromagnetic fields on *Helicobacter pylori* biofilm. *Curr. Microbiol.* 60 (6), 412–418. doi:10.1007/s00284-009-9558-9

Dihel, L. E., Smith-Sonneborn, J., and Middaugh, C. R. (1985). Effects of an extremely low frequency electromagnetic field on the cell division rate and plasma membrane of Paramecium tetraurelia. *Bioelectromagnetics* 6 (1), 61–71. doi:10.1002/bem.2250060107

Du, R., Peng, Y., Ji, J., Shi, L., Gao, R., and Li, X. (2019). Partial denitrification providing nitrite: Opportunities of extending application for anammox. *Environ. Int.* 131, 105001. doi:10.1016/j.envint.2019.105001

Fox, A. (2011). "Magnetism and magnetic materials, by JMD Coey: Scope: Handbook," in *Level: Advanced undergraduates, postgraduates and researchers* (Taylor & Francis).

Goodman, R., Bassett, C. A. L., and Henderson, A. S. (1983). Pulsing electromagnetic fields induce cellular transcription. *Science* 220 (4603), 1283–1285. doi:10.1126/science.6857248

Goodman, R., and Henderson, A. S. (1988). Exposure of salivary gland cells to low-frequency electromagnetic fields alters polypeptide synthesis. *Proc. Natl. Acad. Sci. U. S. A.* 85 (11), 3928–3932. doi:10.1073/pnas.85.11.3928

Jain, A., Gupta, Y., Agrawal, R., Jain, S. K., and Khare, P. (2007). Biofilms—a microbial life perspective: A critical review. *Crit. Rev. Ther. Drug Carr. Syst.* 24 (5), 393–443. doi:10.1615/critrevtherdrugcarriersyst.v24.i5.10

Li, D., Dang, Z., and Zhang, J. (2022). Novel strategy for rapid start-up and stable operation of anammox: Negative pressure coupled with the direct-current electric field. *J. Environ. Manag.* 315, 115167. doi:10.1016/j.jenvman.2022.115167

Li, J., Li, J., Gao, R., Wang, M., Yang, L., Wang, X., et al. (2018). A critical review of one-stage anammox processes for treating industrial wastewater: Optimization strategies based on key functional microorganisms. *Bioresour. Technol.* 265, 498–505. doi:10.1016/j.biortech.2018.07.013

Litovitz, T., Krause, D., Montrose, C., and Mullins, J. (1994). Temporally incoherent magnetic fields mitigate the response of biological systems to temporally coherent magnetic fields. *Bioelectromagnetics* 15 (5), 399–409. doi:10.1002/bem.2250150504

Liu, S., Yang, F., Meng, F., Chen, H., and Gong, Z. (2008). Enhanced anammox consortium activity for nitrogen removal: Impacts of static magnetic field. *J. Biotechnol.* 138 (3), 96–102. doi:10.1016/j.jbiotec.2008.08.002

Madondo, N. I., Kweinor Tetteh, E., Rathilal, S., and Bakare, B. F. (2022). Effect of an electromagnetic field on anaerobic digestion: Comparing an electromagnetic system (ES), a microbial electrolysis system (MEC), and a control with No external force. *Molecules* 27 (11), 3372. doi:10.3390/molecules27113372

Ni, S.-Q., Wang, Y., Wang, Z., Cui, Z., Zhang, J., Ahmad, S., et al. (2021). Utilizing infrared electromagnetic fields to increase anammox activity and growth rates. *ACS Es. Throughput Water* 1 (5), 1220–1228. doi:10.1021/acsestwater.0c00295

Paksy, K., Thuróczy, G., Forgács, Z., Lázár, P., and Gaáti, I. (2000). Influence of sinusoidal 50-Hz magnetic field on cultured human ovarian granulosa cells. *Electro-Magnetobiology* 19 (1), 91–97. doi:10.1081/jbc-100100299

Qu, G., Lv, P., Cai, Y., Tu, C., Ma, X., and Ning, P. (2020). Enhanced anaerobic fermentation of dairy manure by microelectrolysis in electric and magnetic fields. *Renew. Energy* 146, 2758–2765. doi:10.1016/j.renene.2019.06.050

Ratushnyak, A., Andreeva, M., Morozova, O., Morozov, G., and Trushin, M. (2008). Effect of extremely high frequency electromagnetic fields on the microbiological community in rhizosphere of plants. *Int. Agrophysics* 22 (1), 71.

Redlarski, G., Lewczuk, B., Żak, A., Koncicki, A., Krawczuk, M., Piechocki, J., et al. (2015). The influence of electromagnetic pollution on living organisms: Historical trends and forecasting changes. *BioMed Res. Int.* 2015, 1–18. doi:10.1155/2015/234098

Soghomonyan, D., Trchounian, K., and Trchounian, A. (2016). Millimeter waves or extremely high frequency electromagnetic fields in the environment: What are their effects on bacteria? *Appl. Microbiol. Biotechnol.* 100 (11), 4761–4771. doi:10.1007/s00253-016-7538-0

Tekutskaya, E., Barishev, M., and Ilchenko, G. (2015). Influence of low-frequency electromagnetic field on DNA molecules in water solutions. *Biofizika* 60 (6), 1099–1103.

Trinh, H. P., Lee, S.-H., Jeong, G., Yoon, H., and Park, H.-D. (2021). Recent developments of the mainstream anammox processes: Challenges and opportunities. *J. Environ. Chem. Eng.* 9 (4), 105583. doi:10.1016/j.jece.2021.105583

Trushin, M. V. (2003). Studies on distant regulation of bacterial growth and light emission. *Microbiology* 149 (2), 363–368. doi:10.1099/mic.0.25825-0

Tsushima, I., Kindaichi, T., and Okabe, S. (2007). Quantification of anaerobic ammonium-oxidizing bacteria in enrichment cultures by real-time PCR. *Water Res.* 41 (4), 785–794. doi:10.1016/j.watres.2006.11.024

Wang, D., Wang, G., Zhang, G., Xu, X., and Yang, F. (2013). Using graphene oxide to enhance the activity of anammox bacteria for nitrogen removal. *Bioresour. Technol.* 131, 527–530. doi:10.1016/j.biortech.2013.01.099

Wang, Z., Liu, P., Ni, S.-Q., Lee, T., and Ahmad, S. (2021a). Low-Frequency Infrared electromagnetic wave promotes partial nitrification by affecting the community signal system. *Chem. Eng. J.* 425, 131636. doi:10.1016/j.cej.2021.131636

Wang, Z., Liu, X., Ni, S.-Q., Zhang, J., Zhang, X., Ahmad, H. A., et al. (2017). Weak magnetic field: A powerful strategy to enhance partial nitrification. *Water Res.* 120, 190–198. doi:10.1016/j.watres.2017.04.058

Wang, Z., Liu, X., Ni, S.-Q., Zhuang, X., and Lee, T. (2021b). Nano zero-valent iron improves anammox activity by promoting the activity of quorum sensing system. *Water Res.* 202, 117491. doi:10.1016/j.watres.2021.117491

Xu, J.-J., Cheng, Y.-F., Zhu, X.-L., Zhu, W.-Q., and Jin, R.-C. (2019). The performance and microbial community in response to MnO2 nanoparticles in anammox granular sludge. *Chemosphere* 233, 625–632. doi:10.1016/j.chemosphere.2019.06.006

Yavuz, H., and Çelebi, S. S. (2003). A typical application of magnetic field inwastewater treatment with fluidized bed biofilm reactor. *Chem. Eng. Commun.* 190 (5-8), 599–609. doi:10.1080/00986440302112

Zhang, C., Li, L., Hu, X., Wang, F., Qian, G., Qi, N., et al. (2019a). Effects of a pulsed electric field on nitrogen removal through the ANAMMOX process at room temperature. *Bioresour. Technol.* 275, 225–231. doi:10.1016/j.biortech.2018.12.037

Zhang, C., Li, L., Wang, Y., and Hu, X. (2019b). Enhancement of the ANAMMOX bacteria activity and granule stability through pulsed electric field at a lower temperature (16±1° C). *Bioresour. Technol.* 292, 121960. doi:10.1016/j.biortech.2019.121960