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Publication date:
2019

Document Version
Publisher's PDF, also known as Version of record

[Link back to DTU Orbit](#)

Citation (APA):
Xu, M., Zhou, S., Li, M., & Zhang, Y. (2019). *Anammox technologies assist in nitrogen removal in bioelectrochemical system*. 1. Poster session presented at Elelromicrobiology 2019, Aarhus, Denmark.

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Anammox technologies assist in nitrogen removal in bioelectrochemical system

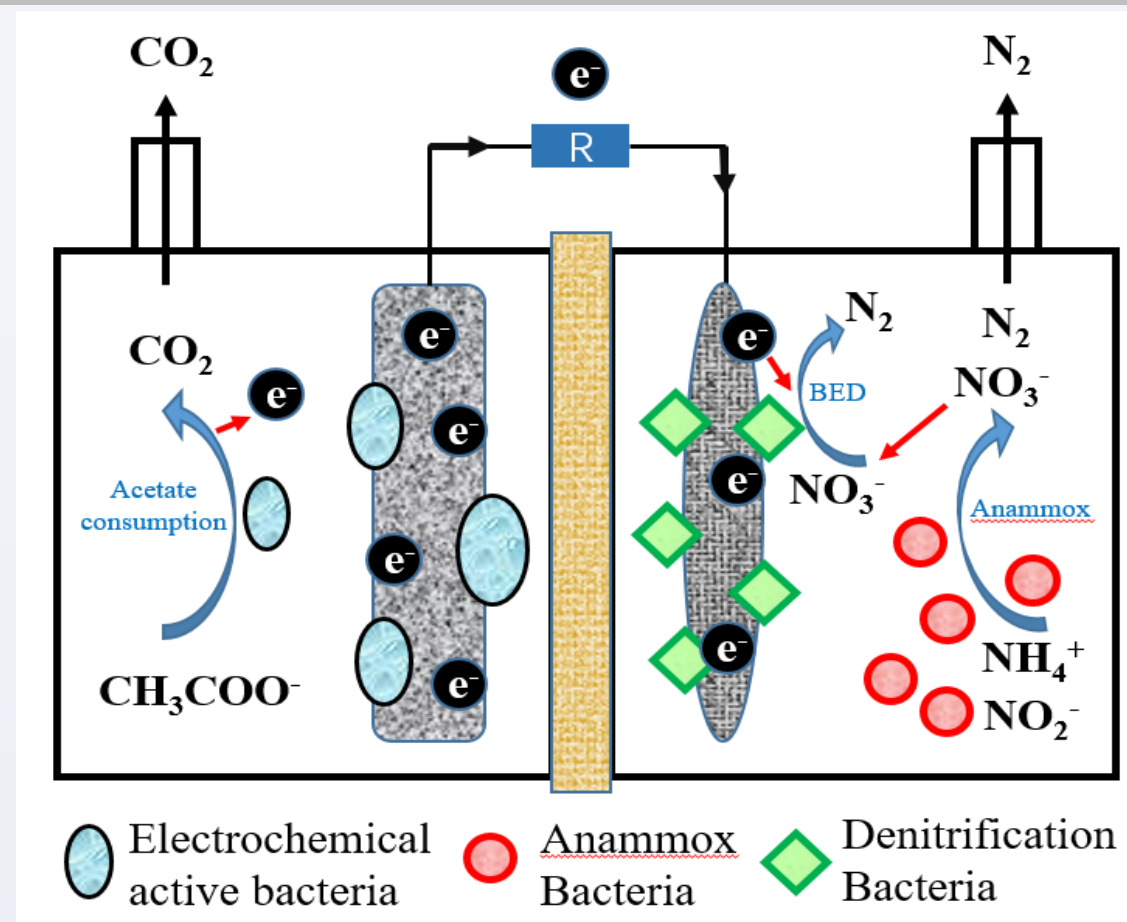
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Introduction

This research coupled Anammox technology with dual chamber microbial fuel cells for a better performance of simultaneous nitrogen removal and power production. The effect of contents of nitrogen in catholyte on reactor's performance was especially studied. Bacterial colonies were analyzed through 16S rRNA technology to determine the reaction mechanism.



Reactor Design

Two identical Plexiglas chambers (4.0 cm long, 3.0 cm diameter, 28 mL volume) were assembled into a dual-chamber MFC reactor separated by a proton exchange membrane. Graphite felts and carbon cloths were individually used as the anode and cathode electrodes, with the PEM sandwiched in the middle.

Two titanium wires were used to connect the anode and cathode electrodes with a spiral rheostat. Disposable syringes were set to take samples and replenish electrolyte, and blue core glass syringes were placed to maintain the pressure and collect N₂ gas from the Anammox process in the cathode chamber. Sealing was ensured by silicone sheet added.

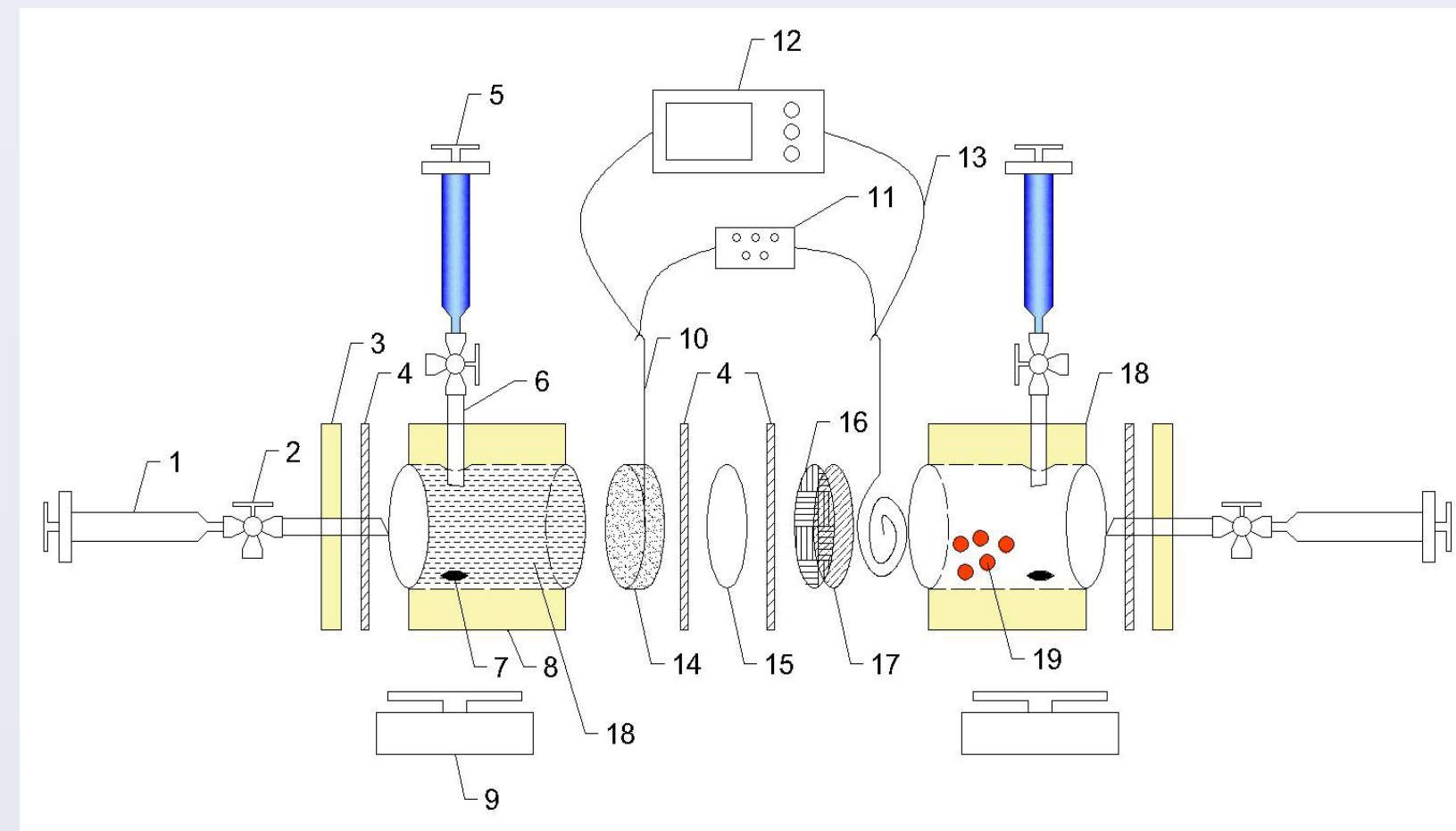


Fig. 1 Schematic of an Anammox-MFC reactor

Bacteria Inoculation

Each anode chamber was inoculated with Electrochemical Active Bacteria (EAB), which was domesticated and screened from samples of Shijing municipal wastewater treatment plant, turbid fluid, anaerobic and heterotrophic, consuming acetate as carbon source.

While each cathode chamber was inoculated with Anammox Bacteria (AnAOB), which was collected from an Anammox-UASB reactor operating for over 10 years, red granules or clusters, anaerobic and autotrophic, consuming carbonate as an inorganic source.

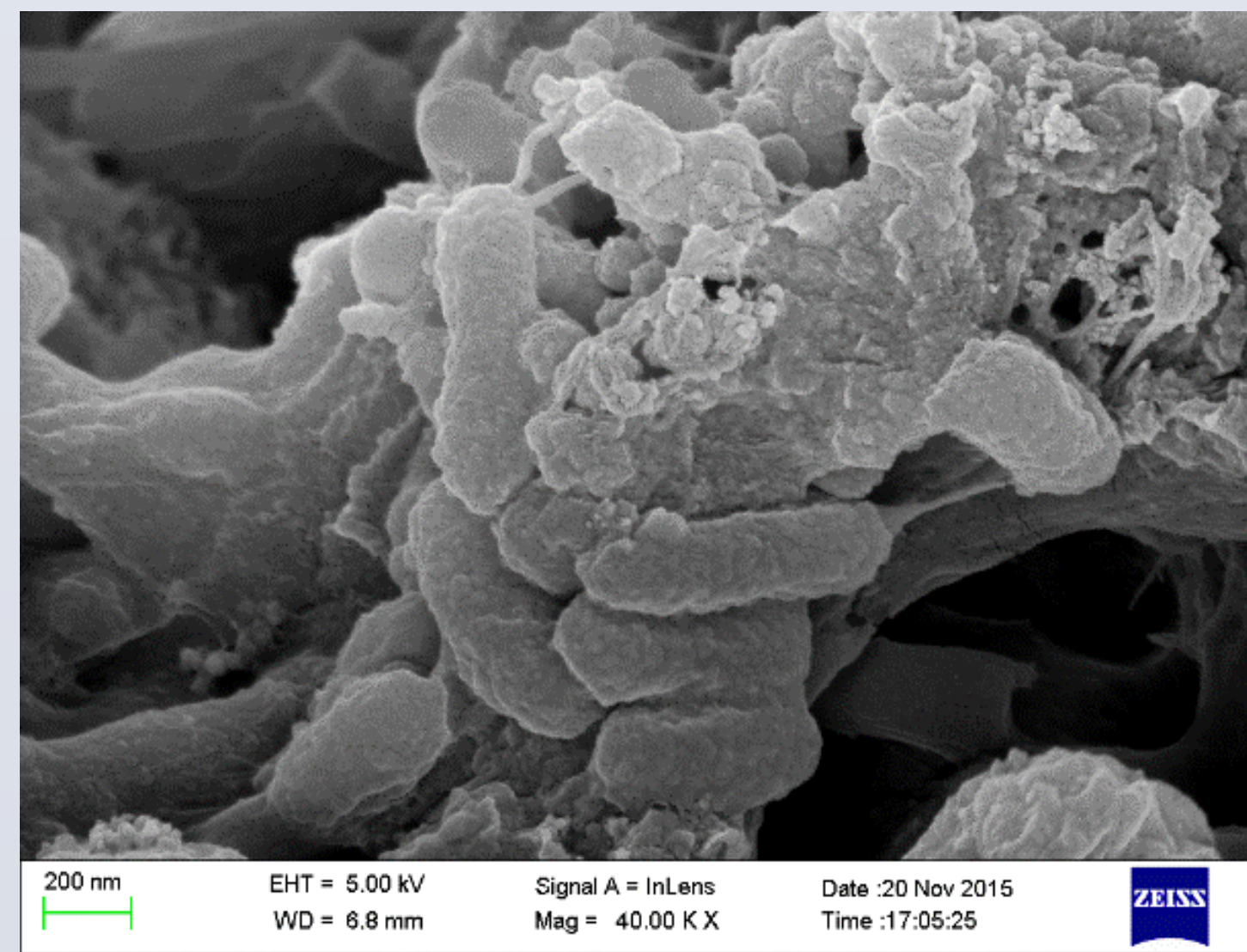
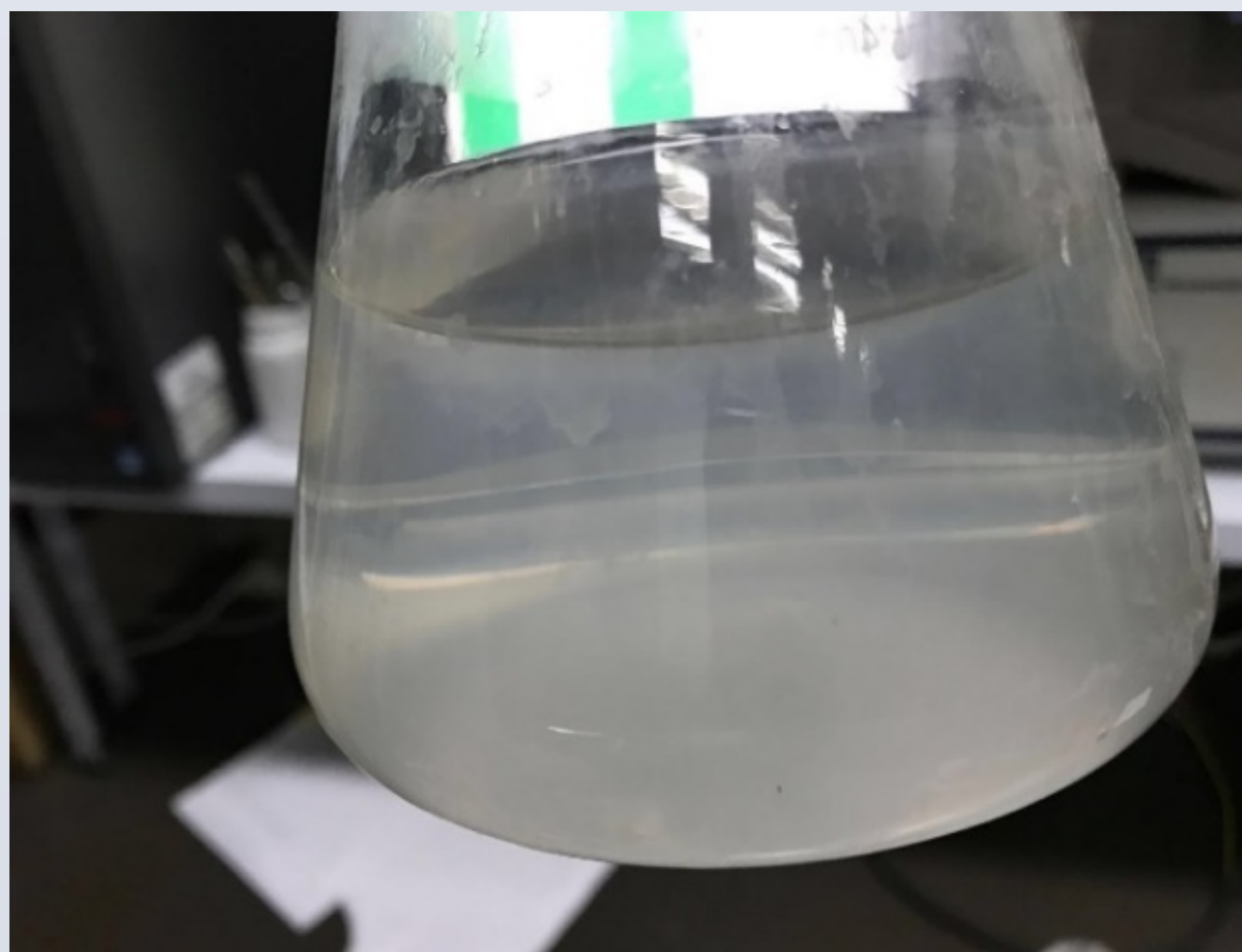


Fig. 2 Pictures and SEM Images of anodic Electrochemical Active Bacteria

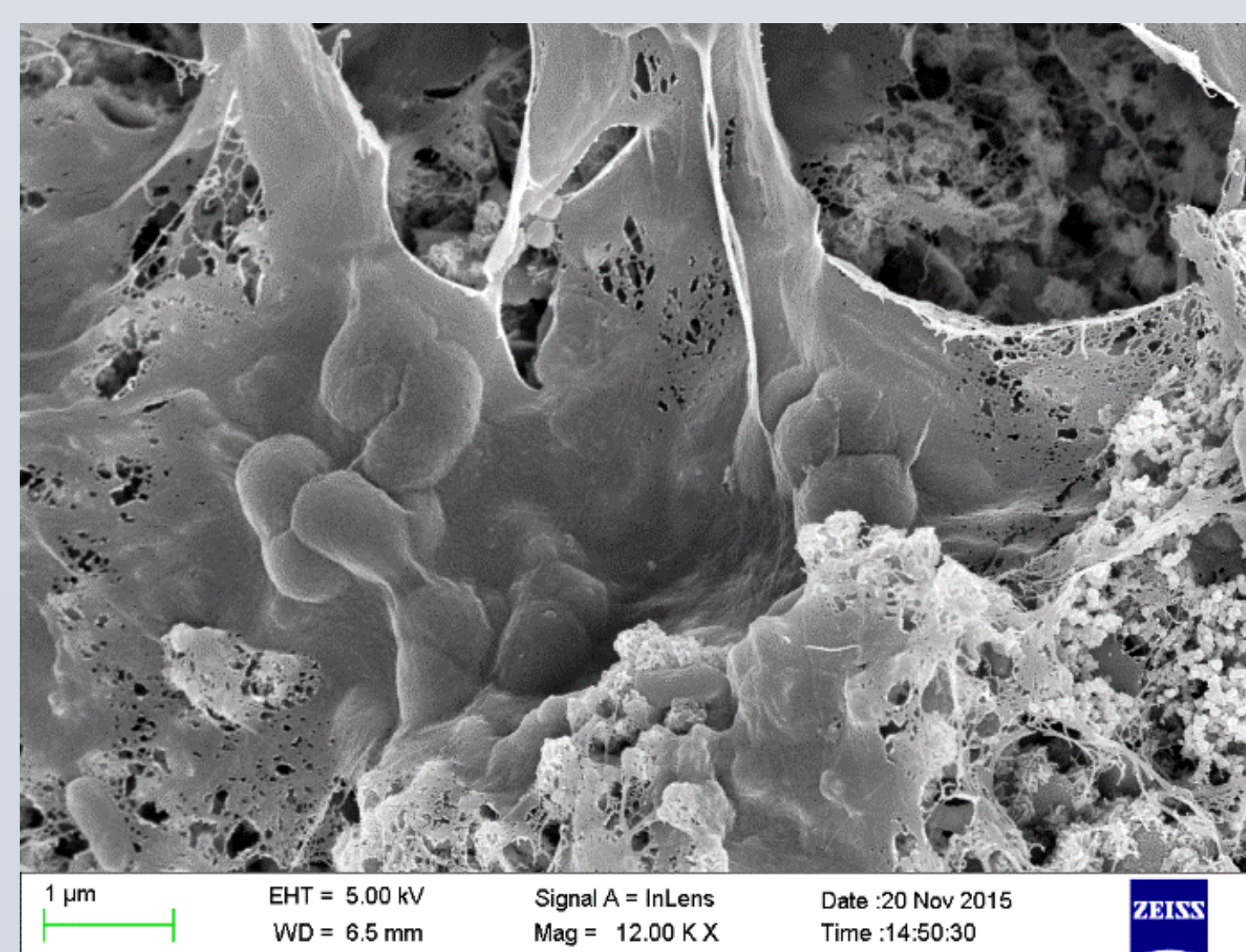


Fig. 3 Pictures and SEM Images of cathodic Anammox Bacteria

Nutrients concentration gradients

Three NaAc concentrations (0.01, 0.02, 0.03 mol/L) were applied respectively to the anolyte of A1, A2, A3-MFC to investigate the effects of COD level on MFC performance. Four mixed nitrogenous solutions containing N-NH₄⁺ concentrations (200, 300, 400, 500 mg·L⁻¹) with corresponding N-NO₂⁻ concentrations (264, 396, 528, 660 mg·L⁻¹) were applied respectively to the catholyte of C1, C2, C3, C4-MFC to study the influence of nitrogen content on performance. In each reactor, the initial pH of anolyte was adjusted to 8.0 ± 0.2, while catholyte was 7.0 ± 0.5.

MFC Performance

V_{max} output of A1, A2, A3-MFC were 369.86 mV, 348.00 mV, and 330.90 mV, and the cycle periods were 2900 min, 4800 min and 8200 min respectively. V_{max} output of C1, C2, C3, C4-MFC were 322.90 mV, 351.53 mV, 360.84 mV, and 247.41 mV individually.

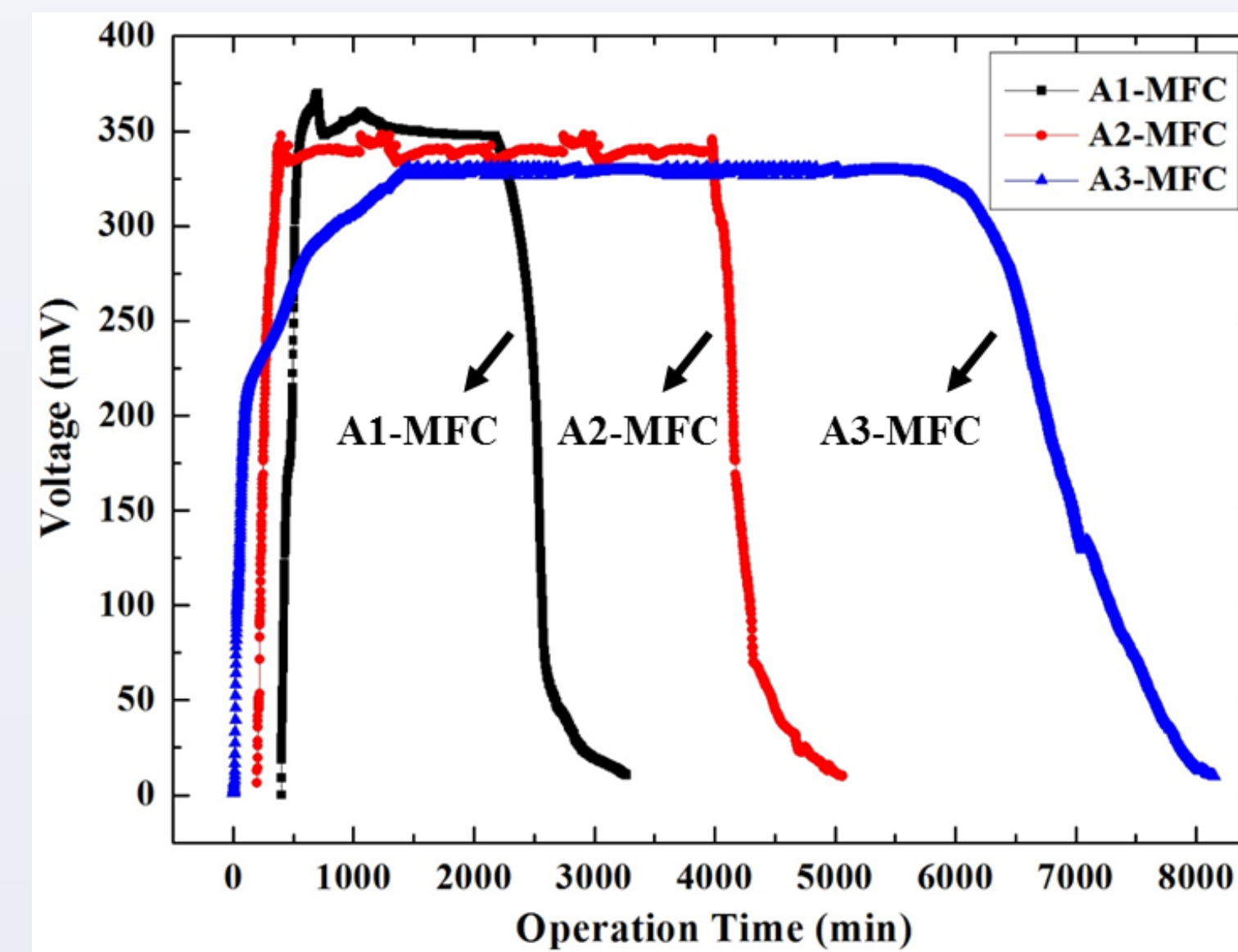


Fig. 4 Power production of A1 ~ A3-MFC in one period

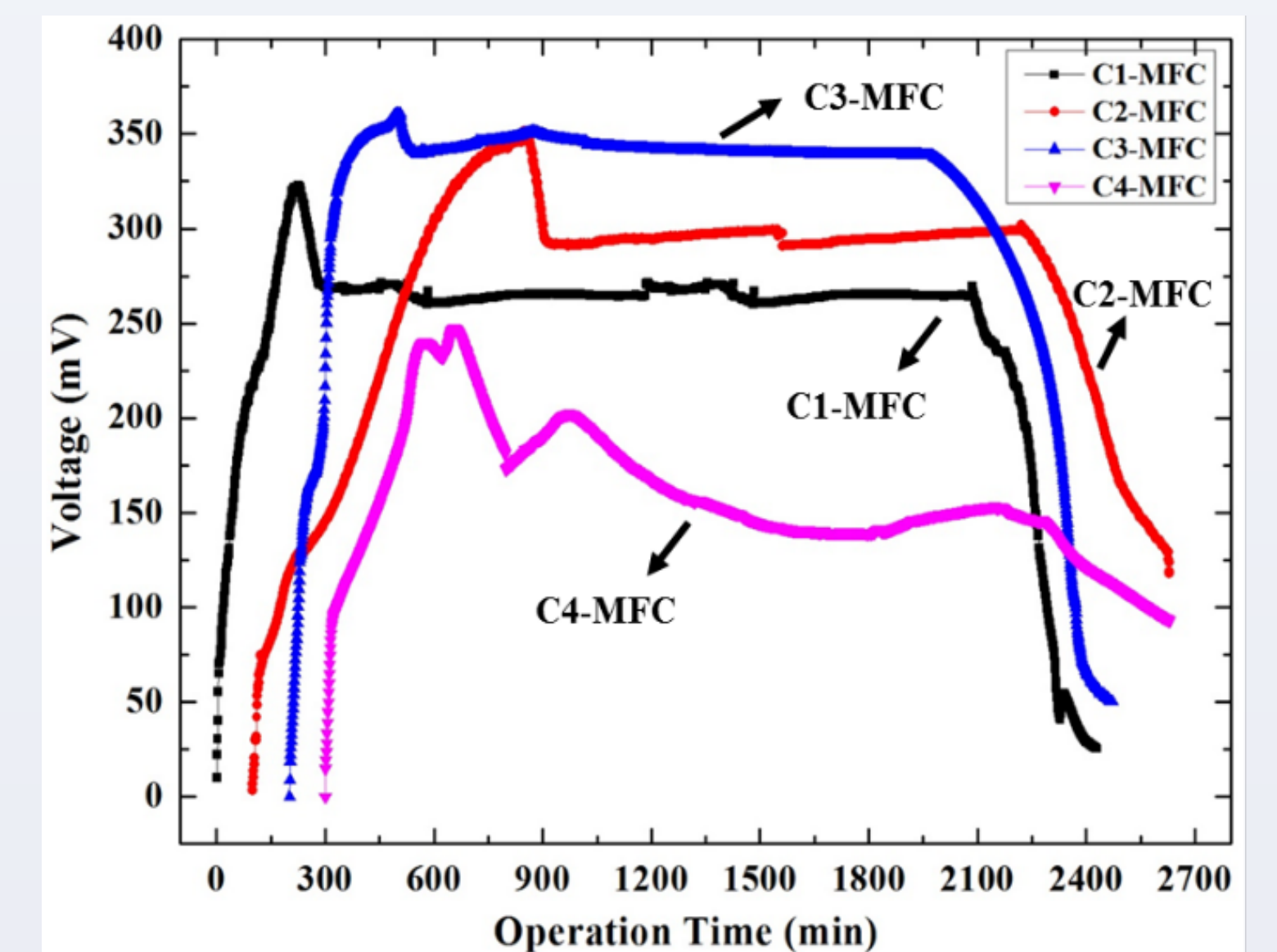


Fig. 5 Power production of C1 ~ C4-MFC in one period

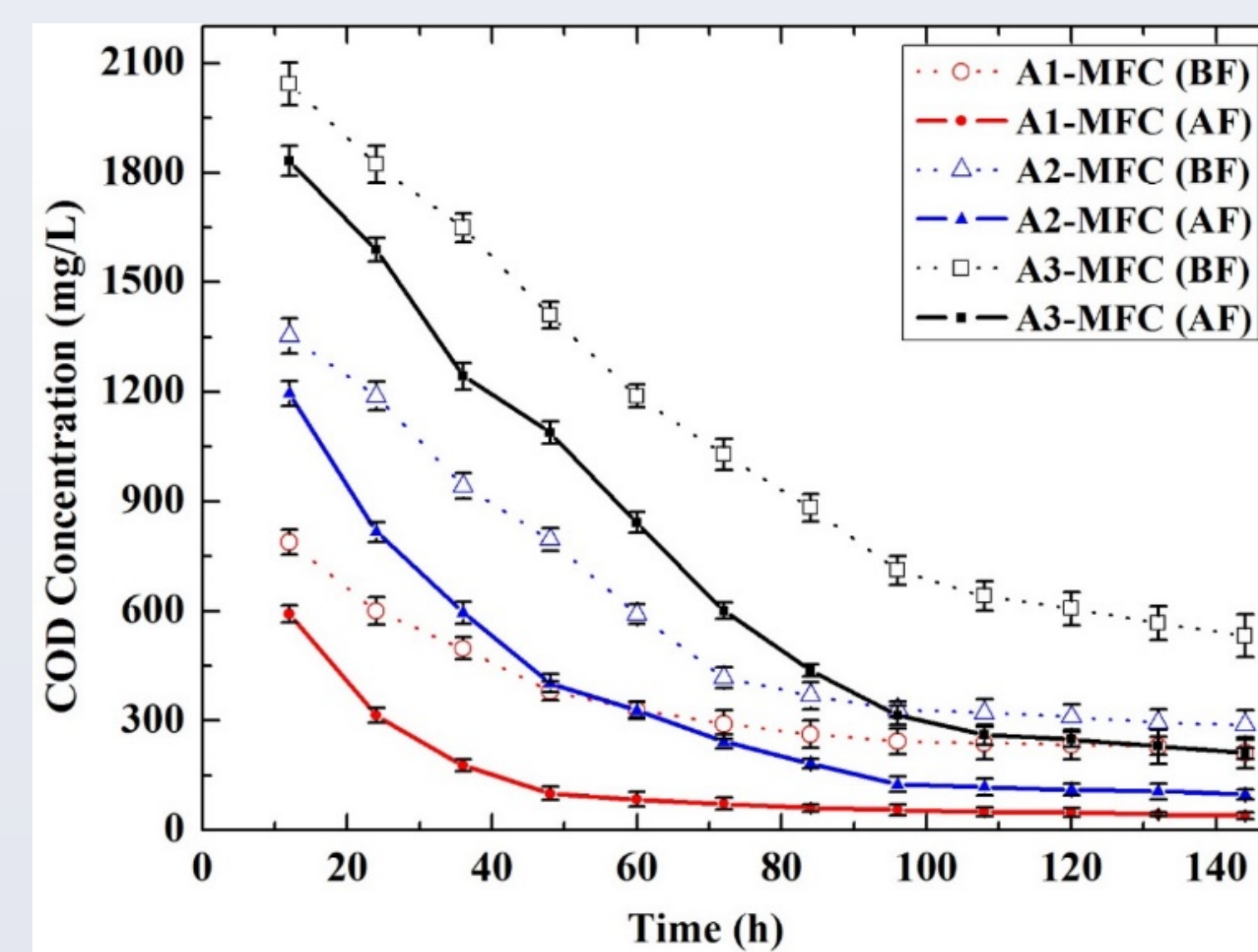


Fig. 6 Comparisons of the time courses of COD concentration change in A1, A2, A3-MFC (BF: Before filtration, AF: After filtration)

	1-MFC	2-MFC	3-MFC	4-MFC	
Influent (mg·L ⁻¹)	N-NH ₄ ⁺	207.69	320.06	411.3	503.92
	N-NO ₂ ⁻	269.67	400.21	537.79	638.8
	N-NO ₃ ⁻	0.57	1.16	1.09	0.72
	TN	477.93	721.43	950.18	1143.44
Effluent (mg·L ⁻¹)	N-NH ₄ ⁺	13.74	16.87	7.47	121.56
	N-NO ₂ ⁻	45.32	61.25	57.02	181.25
	N-NO ₃ ⁻	46.15	58.16	50.04	82.74
	TN	105.21	146.28	114.52	385.55
Removal rate (%)	N-NH ₄ ⁺	93.38	94.73	98.18	75.88
	N-NO ₂ ⁻	83.19	84.70	89.40	71.63
	N-NO ₃ ⁻	83.19	84.70	89.40	71.63
	TN	77.99	81.11	87.95	66.28

Table 1 Comparisons of ammonium, nitrite and nitrate concentration change in C1 ~ C4-MFC

Bacterial Analysis

Sample ID	Raw reads (PE)	Clean Reads (PE)	Clean Contigs	Final Contigs
A0	62563	60099	41206	38414
A1	83189	78965	53177	48951
A2	95479	90619	52548	47143
C0	108958	104123	59547	54690
C1	31357	30232	20112	18568
C2	73010	69251	42582	38677
C3	62695	59950	42453	39566
C4	56845	54347	33085	30523

Table 2 Richness and diversity estimation of the 16S rRNA sequencing libraries from the MiSeq sequencing analysis

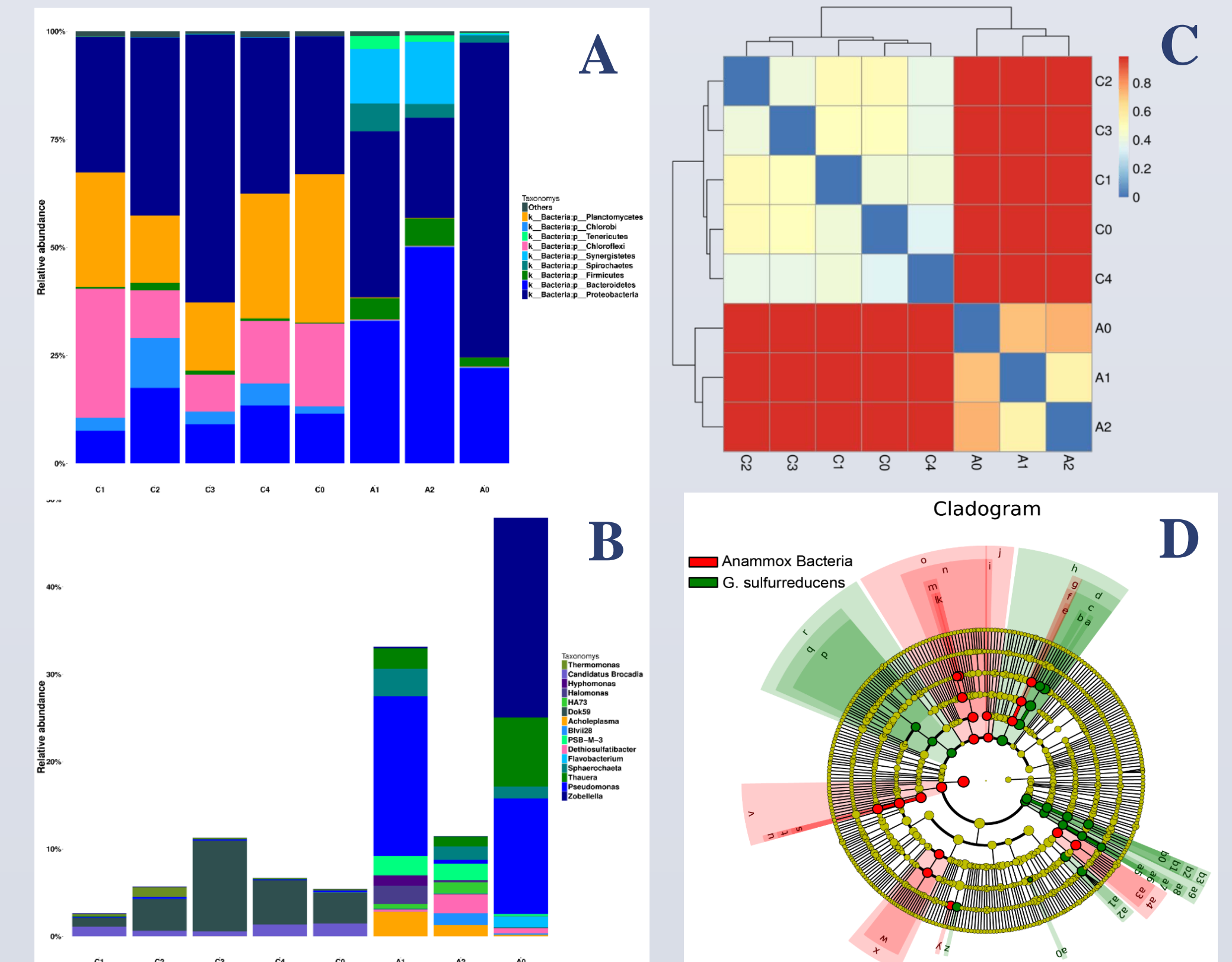


Fig. 7-A, B Relative abundance of different microbial colony composition on level of phylum and genus. Fig. 7-C Bray-Curtis distance heatmap of each sample. Fig. 7-D Cladogram analysis of EAB and AnAOB.

Conclusions

- ◆ Integrated Anammox-MFCs were operated successfully, and the performance of simultaneous nitrogen removal, COD consumption and power production was satisfactory.
- ◆ A higher acetate concentration did not increase the power production apparently, but extended the reaction cycle obviously. 0.01 mol/L was the optimum value.
- ◆ As the content of nitrogen increased, the capacity of MFC was enhanced, but inhibited obviously while excessive. 400 mg·L⁻¹ (N-NH₄⁺) and 528 mg·L⁻¹ (N-NO₂⁻) was optimum.
- ◆ The removal rates of COD, ammonium, nitrite and total nitrogen were 93.2%, 98.2%, 89.4% and 87.9% under the optimum circumstance.
- ◆ The dominant genus of anodic EAB was *Pseudomonas*. And the dominant cathodic genus included *Candidatus Brocadia* (Anammox) and *Dok59* (Bioelectrical-Denitrification).

Acknowledgments

Financial supports from the Ministry of Science and Technology of China for State Key Research and Development Project (2016YFC0400708) is gratefully acknowledged.