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# Abstract of thesis

Anaerobic digestion (AD) of livestock waste is an attractive practice as it can solve the problem of waste contamination and produce renewable energy. However, the ammonia which is rich in livestock waste or produced as a metabolic end product during the biodegradation of high nitrogenous compounds, is toxic to methanogens and often causes failure of the whole biological process at high concentration. The author also reported that the several methods, such as ammonia stripping, struvite precipitation or adding ammonia adsorption material, have been developed to alleviate ammonia inhibition. However, all these methods have certain limitations like energy intensive and large effluent discharge that are challenging for their practical application (Chapter 1). The author introduced that bedding material fixed-bed reactor by adding absorbent to immobilize microorganism in bioreactor for anaerobic digestion of ammonium rich waste can be used to minimize these limitations. Zeolite could adsorb ammonia and dissociate trace elements like $Ca^{2+}$ and $Mg^{2+}$ (via ion-exchange) which provide favorable environment for microbial growth. Anaerobes immobilize on bed materials could create a high cell density and activity for resistance of high ammonia environment. Nevertheless, very few research investigated the combination effects of suitable bed material and zeolite to mitigate ammonia inhibition in AD process. Meanwhile, raw zeolite containing less metal cations showed limited ion-exchange capacity with poor ammonium adsorption ability. Modification works on zeolite designed specifically for high ammonium adsorption capacity and nutritional cations supplement are required. Furthermore, it has been found that intermittent illumination could relieve ammonium inhibition and enhance methane production. Therefore, the main objective of the author's research was to develop a novel bioprocess incorporated with optimal bed material fixed modified-zeolite bioreactor and intermittent illumination for alleviating ammonia inhibition to improve the efficiency of ammonium-rich AD.

In chapter 2, the author firstly explored the optimal bed material to develop the fixed bed zeolite bioreactor for mitigating ammonia inhibition during ammonium-rich AD process. Three typical materials including polymer foaming sponge (PFS), chlorinated polyethylene (CPE) and porous nylon (PN) were investigated. Series of batch experiments were carried with 3 bed materials at ammonium-rich condition ($NH_4^+$-N: 7511 mg/l). The CPE fixed bioreactor showed the shortest start-up period (2 days) and the highest methane production (96 times higher than that of the Control). In addition, the author carried the CPE bioreactor under long-term (100 days) operation successfully. The author illustrated that the synergy of ammonia adsorption and microorganism immobilization on CPE fixed zeolite contributed to the enhanced performance. Zeolite adsorbed ammonia and dissociated trace

elements such as $Ca^{2+}$ and $Mg^{2+}$, which provided microbes a favorable environment for growth. Meanwhile, large amount of anaerobes aggregated on the CPE fixed zeolite system exhibited high resistance to ammonia inhibition environment.

Oyster shell with high $Ca^{2+}$ content for ammonium ion exchange, and lignite contains abundant mineral elements could be a good source for cations supplement in AD. Hence, in chapter 3, oyster shell and lignite were used as modification material for zeolite optimization by the author. In addition, the author further adopted intermittent illumination for constructing a high-efficient AD bioprocess. After modification, the author evaluated the workability of the newly synthesized OLMZ (oyster shell-lignite modified zeolite). The results showed that the content of metal ions in OLMZ increased significantly, and ammonia adsorption capability of OLMZ was improved by 1.3-fold compared with raw zeolite (UMZ). Adsorption kinetics and isotherm results verified ammonia adsorption on OLMZ was dominated by ion exchange with high affinity for ammonia uptake. Then, the author investigated the feasibility and possible benefits of incorporating the CPE fixed OLMZ bioreactor with intermittent illumination strategy. The illuminated CPE fixed OLMZ bioreactor (OLMZ-I) resulted in the highest methane yield of 372 mL/g-$DOC_{removed}$, which showed 300% increase comparing to the Control (124 mL/g-$DOC_{removed}$). Correspondingly, OLMZ-I showed increased ATP and coenzyme $F_{420}$ value implying high activation of methanogens. The large microbe immobilized (Biomass quantity and SEM observation) in the OLMZ-I further supported the enhanced performance of the reactor. Meanwhile, the sludge conductance in OLMZ-I increased about 3-fold compared to the control, reflecting the electrical communication between anaerobes was strengthened. Furthermore, the author proposed the underlying mechanisms of the integrated OLMZ-I bioprocess. The synergetic effects of ammonia removal, microbe immobilization, metal cations supplement and accelerated electrical communication between methanogenic system combined with light stimulation on methanogen activation facilitated the enhancement of methanogenesis in OLMZ-I bioreactor.

In summary, the author has shown in this thesis that CPE is the optimal material for developing high efficiency bed material fixed zeolite bioreactor for ammonium-rich AD. The modified zeolite (OLMZ) illustrated advanced ammonium adsorption capacity and abundant metal cations content, effectively mitigated the negative impacts of ammonia and improved the microorganism growth. Moreover, light stimulation further enhanced the activity of methanogens in the CPE fixed OLMZ bioreactor. The novel illuminated CPE fixed OLMZ bioprocess shows great potential for biomethane recovery from ammonium-rich waste.

## Abstract of assessment result

【Review】

Anaerobic digestion (AD) of livestock waste is an attractive practice as it can solve the problem of waste contamination and produce renewable energy. However, the inhibition of high concentration ammonia poses a great challenge to AD of ammonium-rich livestock wastes. In this regard, the author successfully developed a novel bioreactor constructed by identifying the optimal bed material (CPE), modifying raw zeolite with high cation exchange (OLMZ), investigating the feasibility and possible benefits of integrating the CPE fixed OLMZ bioreactor with intermittent illumination (OLMZ-I) for high-efficient ammonium-rich anaerobic digestion. The OLMZ-I bioprocess further accelerate all the ammonium-rich waste treatment and improve the biomethane conversion. Moreover, the author illustrated the mechanism involved in the newly developed OLMZ-I bioprocess. This present study provides a valuable platform for improving the biogas production efficiency and advance anaerobic digestion engineering.

【Result】

The final examination committee conducted a meeting as a final examination on 24 July, 2020. The applicant provided an overview of dissertation, addressed questions and comments raised during Q&A session. All of the committee members reached a final decision that the applicant has passed the final examination.

【Conclusion】

Therefore, the final examination committee approved that the applicant is qualified to be awarded the degree of (Doctor of Philosophy in Biotechnology).