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Effect of Volume Loading Rate and C/N on Ship Domestic Sewage Treatment by Two Membrane Bioreactors

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Abstract

Design of ship sewage treatment systems that not only satisfy the use of small space on board but also meets International Maritime Organisation (IMO) latest emission standards is still a challenging problem for ship industry. This study provides a comparative disquisition between two different MBR reactors i.e, air-lift multilevel circulation membrane reactor (AMCMBR) and anaerobic/anoxic/aerobic membrane reactor (AOA-MBR) for domestic sewage treatment. The influence of pollutants volume loading rate (VLR) and C/N on effluent chemical oxygen demand (COD), ammonium nitrogen (NH_4^+-N) and TN for marine domestic sewage was analyzed. The results revealed that AMCMBR showed better removal efficiencies for COD and TN than AOA-MBR. The volume of AMCMBR was only half of the AOA-MBR. In addition, high average value of mixed liquor volatile suspended solids (MLVSS)/mixed liquid suspended solids (MLSS) (i.e. 0.75) of AMCMBR indicated high biomass and good pollutants removal achieved by this reactor. An interesting phenomenon was found in the study regarding *Urease* activity for the two reactors. *Urease* activity for AMCMBR in different working conditions all exceeded AOA-MBR and there exist no clear difference of NR activities between AMCMBR and AOA-MBR except for low C/N ratio (i.e. 6 and 4). This phenomenon proved that AMCMBR has a greater performance for treating ship domestic wastewater.

Keywords

air-lift multilevel circulation membrane reactor, anaerobic/anoxic/aerobic membrane reactor, marine domestic sewage, C/N, pollutants volume loading rate, IMO, *Urease*

1 Introduction

Ships discharge black and grey water directly into marine environment, which can bring excessive calamity to the marine ecosystem. The direct discharge of ship sewage can result in nutrient enrichment and algal blooms [1–3]. In recent years, implementation of the latest International Maritime Organization (IMO) discharge standard applicable from 2016 has raised severe emission standards for ship domestic wastewater discharge [4]. Compared with the old IMO emission regulations, like MARPOL73/78 and MEPC.159(55), TN discharge standard is introduced for the first time in new regulation, which should be below 20 mg/L [5, 6]. Therefore, there is an urgent necessity for onboard ships wastewater treatment systems to meet these strict standards.

Nowadays, MBR technology has been applied for wastewater treatment for many advantages, like biomass enrichment, ensured sludge-effluent separation, small footprint, easy manipulation of the hydraulic and sludge retention times (HRT and SRT) and excellent effluent quality [7–9]. Aerobic MBR technology is widely used for domestic wastewater treatment [10, 11]. Khan et al. [12] used a laboratory-scale membrane bioreactor (MBR) for domestic wastewater treatment at two different pollutants volume loading rate (VLR). The results showed that more than 95 % chemical oxygen demand (COD), 89 % ammonium nitrogen (NH_4^+-N) and 34 % total nitrogen (TN) was removed. For the treatment of real domestic waste water, Song et al. [13] employed a novel pilot gravity-driven anoxic/oxic fed-batch membrane bioreactor (AFMBR). The results showed that NH_4^+-N, COD, and TN removals were ranged from 71 % to 97 %, from 78 % to 96 %, and from 20 % to 60 %, respectively. Hussain et al. [14] developed an anaerobic-microaerobic fixed biofilm (AMFB) reactor to study carbon removal simultaneously

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with ammonium oxidation in dilute domestic wastewater. The results demonstrated that AMFB reactor achieved the maximum 99 % COD removal efficiency at HRT of 24 h and almost all biochemical oxygen demand (BOD) was removed. However, most of the studies focus on various aspects of onshore MBRs, while few studies discussed offshore MBRs [7, 15]. In addition, large volume and failure to meet emission standards are found to be existing challenges for ship wastewater treatment systems.

MBR technology is still facing many challenges concerning complex operational requirements and uncontrollability of marine environment. Many influencing factors, like temperature, HRT, pH, influent pollutants loading rate and C/N ratio, were considered to affect the removal efficiency of MBR [16, 17]. In marine pollution aspect, constant pollutants shock and unstable C/N ratio are two significant characteristics of ship domestic sewage [18]. Therefore, it is essential to investigate the influences of pollutants volume loading rate (VLR) and C/N on MBR performance for the new IMO discharge standard.

The activated sludge is an important part of MBR reactor. It directs the treatment efficiency and effects of sewage treatment [19]. Enzyme activity (EA) is an effective way to describe the activity and quantity of microorganisms in the activated sludge which play vital roles in nutrients removal (especially N removal) for MBR [20]. For example, *Urease* (metal enzyme) is a decomposition catalyst of urea which can catalyze urea hydrolysis to generate ammonia and carbon dioxide, as well as the hydrolysis of C-N bond of amide [21]. Therefore, *Urease* can be used as an indicator for aerobic MBR running [22]. *Nitrate Reductase* is the principal limiting step of denitrification. Therefore, taking this enzymatic activity as an indicator can present the denitrification ability of wastewater treatment plants [23]. For pollutant decomposition, microorganisms present in activated sludge play vital role. Moreover, the microbial decomposition rate on organic matter is directly related to the enzyme activity of microorganisms.

To address these existing challenges, comparison of two different MBR systems was performed in this study to analyze the effect of pollutants VLR and C/N on organic degradation and nutrient removal in wastewater treatment. COD removal, TN removal, mixed liquid suspended solids (MLSS), mixed liquor volatile suspended solids (MLVSS), MLVSS/MLVSS (suspended sludge and membrane attached sludge) in the system were examined. Moreover, microbial enzyme activities of activated sludge were studied.

2 Materials and methods

2.1 Sludge and synthetic wastewater

Sludge used in both reactors was acquired from the secondary sedimentation tank of Wenchang municipal wastewater treatment plant at Harbin, Heilongjiang Province, China. Synthetic wastewater containing: NH_4Cl, $MgSO_4$, $CaCl_2$, yeast, $CH_3COONa{\cdot}3H_2O$, glucose, soluble starch, $NaHCO_3$, Trace element liquid. The composition of trace element liquid: $FeCl_3{\cdot}6H_2O$, H_3BO_3, $CuSO_4{\cdot}5H_2O$, KI, $NaCl{\cdot}4H_2O$, $Na_2MnO_4{\cdot}2H_2O$, $ZnSO_4{\cdot}7H_2O$, $CoCl_2{\cdot}6H_2O$, EDTANa. The composition of local blackwater is shown at Table 1. The local blackwater was mixed with synthetic wastewater in some proportions for experimental setup in order to imitate the actual domestic sewage.

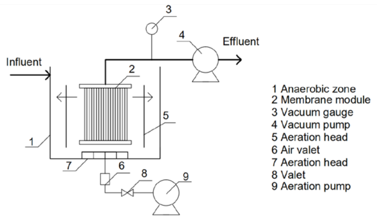
2.2 Bench scale experiment of two different reactors

The schematic diagrams of AMCMBR and AOA-MBR are shown in Fig. 1 (a) and Fig. 1 (b), respectively. Furthermore, two bench-scale MBR systems are shown in Fig. 1 (c). Two reactors were designed by the same processing wastewater flow. The main reactor for both reactors was made of organic glass (Polymethyl methacrylate).

For AMCMBR, the total volume and effective volume were 60 and 50L, respectively. The two flat-plat membrane module made of polyvinylidene fluoride (PVDF) (Tianjin Motimo Membrane Technology Co., LTD) used in this system with the usable membrane area of $1m^2$, pH range of 2~11 and membrane aperture of 0.02 μm. Therefore, the total membrane area of membrane module in the system is $2m^2$. The system operation was set as 9 min for positive pumping and 30 second for back flushing so as to guarantee the normal membrane flux. A large amount of local ventilation was designed owing to the center-fixed installation of sand head aeration. Meanwhile, it form multilevel circulations through open pore of baffle to mix gas and water adequately, which can increase bubble circulation and meet the requirement of dissolved oxygen (DO). Compared with traditional MBRs, the system has

Table 1 The composition of local blackwater for experiment set-up

Pollutants and working conditions	Value
BOD(mg/L)	90-95
COD (mg/L)	300-400
NH_4^+-N (mg/L)	60-65
TN (mg/L)	65-70
pH	5.5
Salinity (g/L)	0
Suspended solid (mg/L)	300-500



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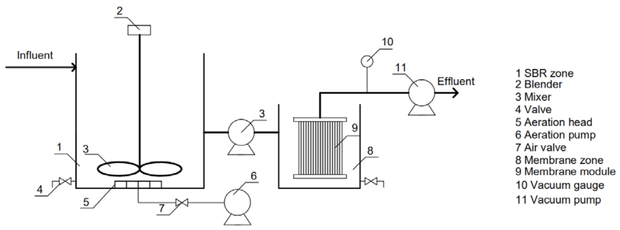

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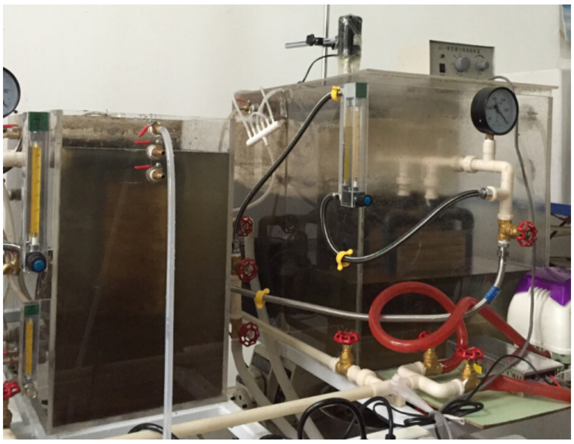


(c)

Fig. 1 Schematic of the two reactors used in this study. (a) AMCMBR; (b) AOA-MBR (c) Two bench-scale MBR reactors (Left: AMCMBR; Right: AOA-MBR)

better exchange and mass transfer, and air bubbles will be distributed more evenly. A parallel connection system of aerobic zones was designed in the pilot-scale AMCMBR. When a membrane in one aerobic zone is under backwashing, the other one can work instead, which will not affect the normal operation of the system.

The AOA-MBR system comprised of a sequencing batch and membrane zone. The total volume of AOA-MBR was 140 L. The same membrane module was used as that of AMCMBR. The wastewater fed into the reactor and through three phases in the sequencing batch zone: anaerobic phase (2 hours), aerobic phase (4 hours) and anoxic phase (2 hours). The phase was changed by on and off control of the air pump. For anaerobic/anoxic phase, air pump was closed, while the blender was open and agitator began to work in order to keep a full mixing of sludge and water. Blender and stirrer were closed and air pump worked again in the aerobic phase. This caused the discharge of water and drop of water level in the reactor. When aerobic phase was finished, the anaerobic phase restarted in the next cycle. The influent wastewater flowed into the membrane zone by peristaltic pump after treating in sequencing batch zone.

2.3 Experimental setup

The sludge and synthetic wastewater were added into the two reactors. The working environment was as follows: dissolved oxygen (DO) was maintained at 2.0–3.0 mg/L, influent COD at 400 mg/L, TN at 25 mg/L. The nutrition solution was added to two reactors by swapping the treated water with untreated synthetic wastewater. When sludge flocculation formed in the reactor and effluent COD went down to 100 mg /L, influent COD and TN were increased to 500 mg/L and 60 mg/L gradually until aerobic activated sludge was fully acclimated. After acclimatization of activated sludge, bioreactor started up. The HRT was maintained at 8 hours and all experiments were conducted under room temperature. The operation was divided into 4 phases according to different working conditions, see Table 2.

2.4 Analytical methods

In all experiments, samples were filtered through 0.45 μm filter paper. The concentrations of COD, MLSS, MLVSS, NH_4^+-N, TN were measured according to the standard methods [24]. The sludge volume index (SVI) was calculated as the biomass volume after 30 min of settling [25]. The pH values of samples were monitored by pH meter (PC-320). Dissolved oxygen (DO) and temperature were monitored via a portable handheld DO meter (WTW, Germany).

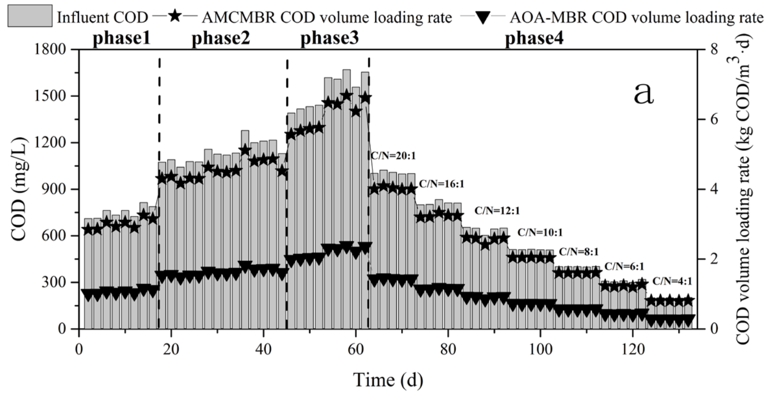
3 Results and discussion

3.1 COD removal efficiency

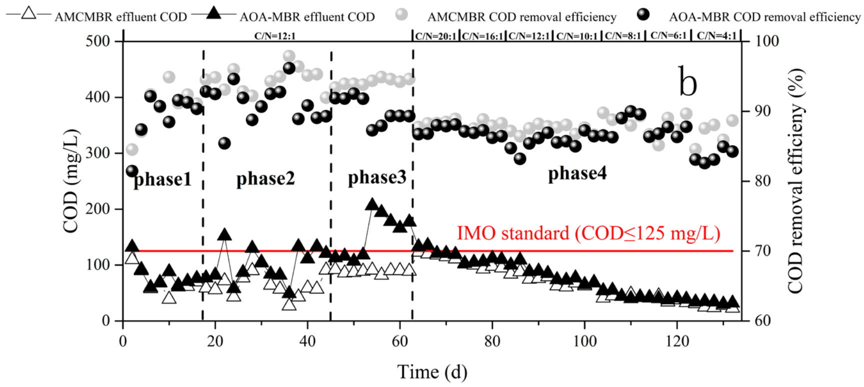
Two different MBR systems were investigated to explored organic carbon and nitrogen removal mechanism in ship wastewater under different pollutants loading. Fig. 2 (a) and (b) illustrates the performance of COD removal efficiency of the two reactors under different COD volume

Table 2 Three different stages of influent parameters

Phase	Duration (d)	COD (mg/L)	NH_4^+-N (mg/L)	TN (mg/L)	C/N
Phase 1	1-18				
Phase 2	19-46	711-1654	46-108	47-110	12:1
Phase 3	47-64				
Phase 4	65-132	1000-200	50	50	20:1-4:1



(a)



(b)

Fig. 2 COD removal of two different reactors: (a) COD volume loading rate; (b) COD removal efficiency

loading rate (VLR). Two reactors showed clear difference in effluent contaminants. During the whole experiment, the average COD removal efficiency of AMCMBR and AOA-MBR was 93.30 % and 90.35 %, respectively, indicating that both reactors performed well in COD removal. This result is consistent with the previous findings by Li et al. [26]. The authors used sequencing batch reactor (SBR) to study the simultaneous removal of organic carbon, nitrogen and phosphorus. The results showed that the reactor achieved around 90 % COD removal rate at a COD loading of 1.00 kg COD/m^3 per day. It should be noticed that the effluent COD rise rapidly at first and then fall in the next few days when OLR increases. More pollutants were degraded and the effluent quality was thus improved due to the well adaptation of bacteria to stable DO environment during this period [27]. As can be seen from the Fig. 2, the COD removal rate of AMCMBR showed an upward trend throughout the three phases, from 90.52 % in phase 1 to 94.30 % in phase 4, respectively. However, AOA-MBR performs differently as compared to AMCMBR. An increase of OLR to around 1 kg-COD/ $m^3 \times d$ increased the removal efficiencies of COD of AOA-MBR from 89.17 % to 91.15 %. Finally, the COD removal efficiency fall at OLR of 1.6 kg-COD/ $m^3 \times d$. This suggests that the optimal OLR of AOA-MBR is 1 kg-COD/ $m^3 \times d$ in domestic ship sewage treatment. During 4th stage, the influent C/N is slightly reduced from 20:1 to 4:1. It can be clearly observed that for the two reactors, effluent COD declined in this stage and COD removal rate was kept at around 87 % throughout the whole experiment.

More than 90 % COD was removed throughout the experiment. This indicates combined removal mechanism of microbial degradation and membrane filtration for the removal of organic pollutants by both MBR systems [28, 29]. It should be noticed that COD removal efficiency of the AOA-MBR was lower than AMCMBR, which demonstrated that microorganisms in the AMCMBR have a higher capacity of organic carbon removal and better endurance of shock loading than single activated sludge in the AOA-MBR. Moreover, membrane retention in AMCMBR promotes biomass concentration in the MBR system to achieve perfect removal efficiency of pollutants [30, 31]. The IMO specifies that the maximum allowable mass concentration of COD in ship wastewater is

125 mg/L. However, discharge standard for water pollutants from ships in China prescribes that wastewater discharged COD concentration should be below 60mg/L in passenger ships after January 1, 2021 [32]. The presented result indicates that AMCMBR can serve as a feasible and practical reactor to treat ship sewage for former new Chinese emission standard.

3.2 Nitrogen removal efficiency

Fig. 3 illustrates the changes in NH_4-N and TN concentrations and their overall removal efficiencies for both reactors. It can be seen from Fig. 3 (a) and (b) that the two reactors performed well on NH_4^+-N removal under different influent COD concentration and C/N. The average NH_4^+-N removal efficiencies of AMCMBR for phases I, II, III and IV were 94.88 %, 90.43 %, 88.24 % and 95.00 %, while AOA-MBR achieved 95.44 %, 88.74 %, 79.82 % and 97.24 % NH_4^+-N removal rate, respectively. It is worth noting that the effluent NH_4^+-N of AOA-MBR has increased significantly and remained at a high level in phase 3 when N volume loading rate increased to 0.35-0.4 Kg NH_4^+-N/m^3•d. In this phase, the effluent NH_4^+-N increased to around 20 mg/L and NH4-N removal rate lowered to 80 %, which indicats that the maximum nitrogen loading of AOA-MBR is 100 mg/L in domestic ship wastewater treatment. However, as nitrogen volume loading rate increase during phase 3, the NH_4^+-N removal efficiency of AMCMBR kept at 88.24 % and the average effluent NH_4^+-N concentration was 11.36mg/L, which is far lower than AOA-MBR (20.23 mg/L). This suggested that AMCMBR had higher nitrogen loading capacity than AOA-MBR. The average NH_4^+-N removal efficiencies of AMCMBR and AOA-MBR exceed 95 % in phase 4. Results indicated that change of C/N ratio had no great impact on NH_4^+-N removal efficiency. This observation is in line with previous study, which states that autotrophic bacteria could grow easily in aerobic environment, resulting in a good performance on NH_4^+-N removal [33].

The TN removal by AMCMBR and AOA-MBR are shown in Fig. 3 (c). In phase 1, despite some fluctuation, the TN removal efficiency of AMCMBR and AOA-MBR were averaged at 93.78 % and 95.09 and the average effluent TN concentration was 3.48 mg/L and 2.69 mg/L, respectively. With increase in nitrogen volume loading rate, TN removal rate of two reactors showed downward trend in phase 2 to phase 3, nevertheless AOA-MBR seems to be more affected. During these two phases, in AOA-MBR, TN removal efficiency fell from 85.86 % to 77.39 %, while TN removal efficiency of AMCMBR kept at around 88 %. The results suggested that nitrogen volume loading have significant impact on AOA-MBR than AMCMBR. It is obvious from Fig. 3 (c) that when influent C/N ratio dropped from 20:1 to 8:1 in phase 4, the average removal rate of total nitrogen is higher than 88 %. However, as C/N ratio of influent wastewater declined to 6:1, the effluent TN of AMCMBR increased greatly. The corresponding average effluent TN concentration and removal efficiency was 17 mg/L and 66 %. While TN concentration of AOA-MBR was 11 mg/L and TN removal efficiency was 75 %. This indicates that the biological nutrient removal (BNR) system of AMCMBR is on the verge of collapse. Furthermore, it should be noted that the C/N ratio of the two reactors were switched to 4:1 with the corresponding average effluent TN concentrations of 26.47 and 20.52 mg/L showing removal efficiencies of 63 % and 55 %, respectively. The main reason for this phenomenon is the presence of insufficient carbon source for microorganisms under low C/N ratio, which inhibited the growth of microorganisms. Therefore, it leads to a significant reduction in denitrification efficiency [34, 35].

Yang et al. [33] reported that the TN removal mainly rely on simultaneous nitrification and denitrification (SND) performance in SBR, which on account of DO concentration gradients arising from diffusional limitations. Besides, total nitrogen loss in the supernatant of aerobic zone may be caused by SND and assimilation during the continuous aeration membrane bioreactor processes [25, 36, 37]. Therefore, it is important to analyze the pollutants removal efficiency of these two reactors based on SND performance. Fig. 3 (d) showed the percentage of assimilation (AS) and SND of AMCMBR and AOA-MBR of each phase. Equations (1) and (2) represent the SND performance of nitrogen removal [38]:

$$\mu_{\text{assimilation}} = \frac{Y_{\text{obs}} \times \left(\text{COD}_{\text{in}} - \text{COD}_{\text{ef}}\right) \times 0.075}{\text{TN}_{\text{in}} - \text{TN}_{\text{ef}}}, \quad (1)$$

$$\mu_{\text{SND}} = \left(1 - \frac{\eta_{\text{assimilation}}}{100}\right) \times 100\%, \quad (2)$$

where represents N removal assimilation rate, Y_{obs} represents the observed biomass yield coefficient, COD_{in} and COD_{ef} represents the average influent COD and effluent COD, TN_{in} and TN_{ef} represents the influent TN and the effluent TN, μ_{SND} represents the nitrogen removal rate in SND process.

Fig. 3 (d) shows that SND performance of AMCMBR was higher than AOA-SBR during former three phase.

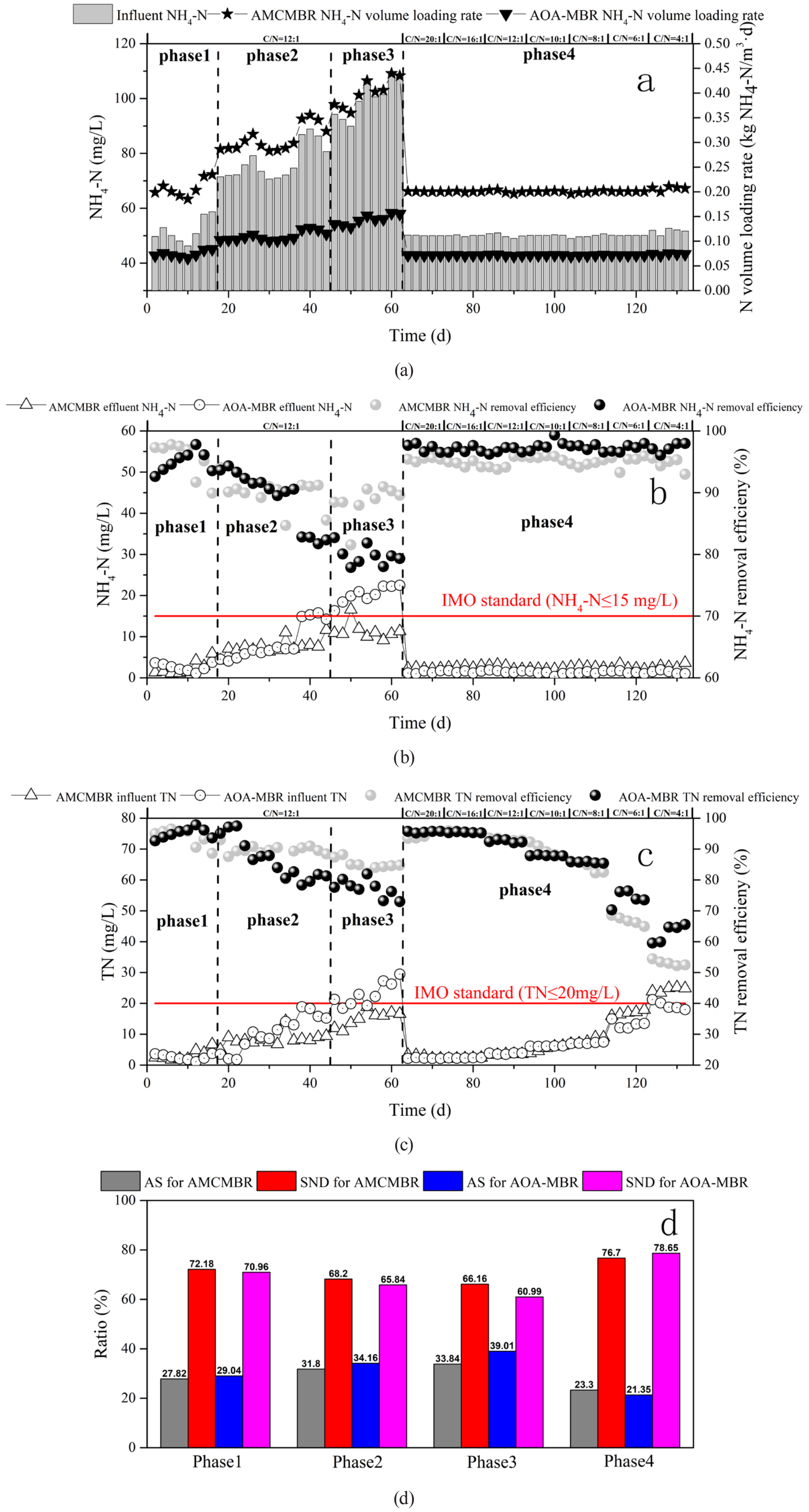


Fig. 3 N removal: (a) NH_4-N volume loading rate of two different reactors; (b) The comparison of NH_4-N removal efficiency of AMCMBR and AOA-MBR; (c) The comparison of TN removal efficiency of AMCMBR and AOA-MBR; (d) Assimilation (AS) and Simultaneous nitrification and denitrification (SND) of AMCMBR and AOA-MBR

This conclusion confirms that the capacity of nitrogen removal in the AMCMBR was much higher than in the AOA-MBR. The main reason for this is the existence of multi-stage loop condition in AMCMBR which is beneficial to the enrichment of nitrifiers and heterotrophic bacteria. The enrichment of bacteria resulted in the formation of large granular sludge, anaerobic/anoxic conditions exist in inner granular sludge whereas the outer one is aerobic [39]. Besides, compared with AOA-MBR, the inner design of membrane module can maintained biomass concentration to enhance the nitrogen removal capacity [40]. Therefore, AMCMBR achieved higher nitrogen removal efficiency than AOA-MBR under different nitrogen volume loading rate. As can be seen from Fig. 3 (c), the nitrogen removal efficiency of the AOA-MBR still higher to 75 % under low C/N ratio (i.e. C/N=6) during fourth phase, which is in consistent with the previous research [11]. Besides, the SND performance of AOA-MBR exceeds than AMCMBR in this phase which indicated that AOA-MBR has advantage for treating ship wastewater with low C/N ratio (i.e. C/N=4 and 6). To sum up, AMCMBR perform well in high nitrogen volume loading rate while AOA-MBR showed good performance under low C/N ratio.

It should be noted that both reactors have equal performance of average COD, NH_4^+-N and TN removal rate. However, in these three aspects: the total equipment volume, effective volume of reaction zone and expected total equipment volume of pilot-scale equipment, AOA-MBR is nearly three times as large as AMCMBR. This means that the AMCMBR has a higher pollutant loading rate, which is presented at Table 3. Therefore, AMCMBR is more practical mean for handling ship domestic wastewater.

3.3 Sludge characteristic of different reactors

The MLSS and MLVSS in suspended sludge of AMCMBR and AOA-MBR are illustrated in Fig. 4 (a) and (b). During the former 54 days, MLSS and MLVSS of two reactors gradually increased when COD and TN VLR rose. On 54th day, the concentration of MLSS and MLVSS of suspended sludge in AMCMBR increased to around 6799 mg/L and 5315 mg/L. The corresponding MLVSS/MLSS value was nearly 0.78 in this period. However, the value of MLSS, MLVSS and MLVSS/MLSS of AOA-MBR was 6022 mg/L, 4400mg/L and 0.73, respectively. Despite fluctuation, the value of MLSS and MLVSS showed a downward trend when C/N ratio reduced from 20:1 to 4:1. The corresponding average value of MLSS, MLVSS and MLVSS/MLSS in AMCMBR were 5038 mg/L, 3802 mg/L and 0.76, respectively, while 4957 mg/L, 3480 mg/L and 0.69 achieved by AOA-MBR. It can be concluded that high pollutants VLR can have a positive effect on the biomass growth, resulting in the increase of suspended sludge in two reactors, while low C/N ratio has a negative influence on the biomass growth.

Previous study reported that MLVSS/MLSS can be effectively used to analyze sludge activity [41]. In this study, as can be seen from Fig. 4 that the value of MLVSS/MLSS in suspend sludge of AMCMBR was higher than AOA-MBR. This interesting phenomenon indicated high activity of microorganism and greater removal efficiencies achieved by lab-scale AMCMBR. This phenomenon confirms the previous conclusion that AMCMBR showed better removal efficiencies for COD and TN than AOA-MBR, which presented in Subsections 3.1 and 3.2. In addition, the average value of MLVSS/MLSS in the attached sludge of AMCMBR exceeded 0.70, revealing that the biomass in the pilot-scale AMCMBR was healthy. The findings are in agreement with Chen [42] and Ng's study [43], who found that the biomass kept healthy when the MLVSS/MLSS ratio was 0.8 ± 0.05. These conclusions demonstrated that AMCMBR has good pollutants removal in comparison with AOA-MBR under different working conditions.

3.4 Enzyme activities

The microorganisms in activated sludge play important role in pollutants decomposition. Previous study reported that the enzyme activity of microorganisms have close

Table 3 Comparison of overall performance of AMCMBR and AOA-MBR

Parameters/Reactor	AMCMBR	AOA-MBR
Total equipment volume(m^3)	0.06	0.15
Effective volume of reaction zone(m^3)	0.05	0.14
Expected total equipment volume of pilot-scale equipment(m^3)	1.06	2.98
Stable flow(m^3/h)	0.01	0.01
Average COD removal efficiency (%)	91.35	88.92
Average NH_4-N removal efficiency (%)	92.13	90.31
Average TN removal (%)	87.54	85.80

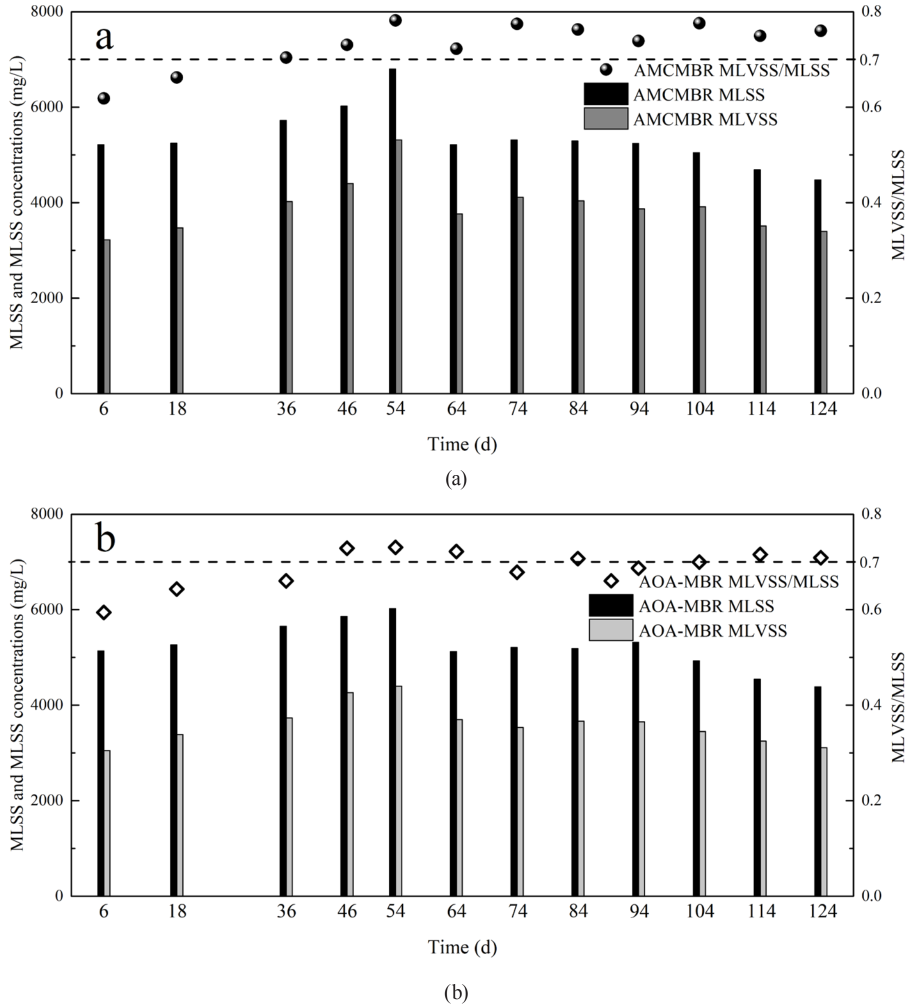
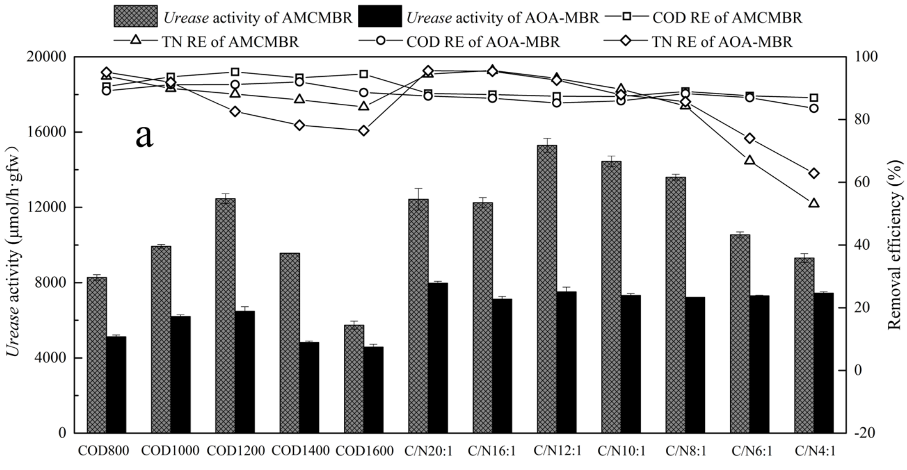


Fig. 4 MLSS and MLVSS in suspend sludge of (a) AMCMBR and (b) AOA-MBR

relationship with the microbial decomposition rate on pollutants [44]. Better enzymatic activities and more microbial species in the MBR system are the main reason of excellent biological COD and TN removal [45]. Therefore, microbial enzyme activities were evaluated to make clear the influence and pollutants removal efficiency on ship wastewater treatment. The sludge samples were taken in different working conditions to analyze enzyme activities, and measured enzyme activities data were averaged to reduce the experimental error.

Average *Urease* activities on different working conditions of two reactors are shown in Fig. 5 (a). Two enzymes were observed, indicating that activities of enzymes were get affected by different working condition. Both the average activities of two enzymes showed similar trend when COD concentrations increased from 800 mg/L to 1600 mg/L, first increased and then decreased. When the influent COD reached 1200 mg/L, the *Urease* activities of AMCMBR reached a maximum of 12465 μmol × h^{-1} × gfw^{-1}, while AOA-MBR achieved 6474 μmol × h^{-1} × gfw^{-1} at this time. The corresponding COD removal efficiencies of AMCMBR also reached a maximum to more than 95 %. With decrement of C/N ratio, despite fluctuation, the *Urease* activities of AMCMBR showed an upward trend at first and decreased afterward as C/N ratio declined from 20:1 to 4:1, while the *Urease* activities of AMCMBR showed some fluctuation during this period. When the C/N ratio was switched to 4, *Urease* activity, COD removal efficiencies of AMCMBR and AOA-MBR were 9310 and 7442 μmol × h^{-1} × gfw^{-1}, 87 % and 84 %, respectively. It can be concluded that increasing pollutant VLR or variation of C/N ratio did not significantly affect *Urease* activities of AOA-MBR.

Average activities of *Nitrate Reductase* on different working conditions of two reactors were demonstrated in Fig. 5 (b). The *Nitrate Reductase* activities showed a downward trend as influent COD concentration increased. When influent COD concentration rose to 1600 mg/L, the *Nitrate Reductase* activities of AMCMBR and AOA-MBR



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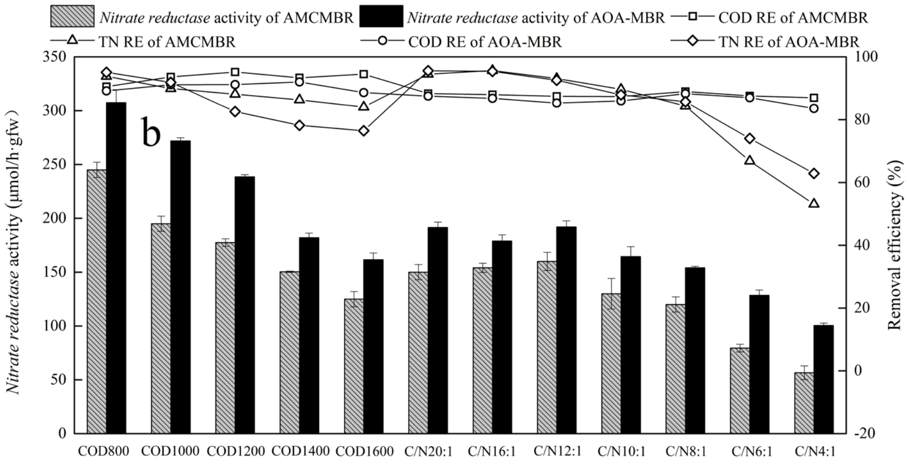

(b)

Fig. 5 Average activities of (a) Urease and (b) NR on different working conditions of two reactors

fell to only 125 and 162 μmol × h^{-1} × gfw^{-1}, respectively, while corresponding TN removal efficiencies reduced from 94 % and 95 % to 84 % and 76 %, respectively. Similarly, the *Nitrate Reductase* activities of two reactors first dropped rapidly and then grew gradually. When C/N ratio reduced to 4, *Nitrate Reductase* activity of AMCMBR and AOA-MBR were 57 and 100 μmol × h^{-1} × gfw^{-1}, respectively. Meanwhile, the TN removal efficiencies reached the lowest point of 53 % and 63 %, respectively.

In summary, the *Urease* activity seems more sensible to the COD removal while NR activity has more close relationship with the TN removal. Li et al. [45] reported that *Urease* is closely related to the number of matrix microorganisms and organic matter content. In addition, the high *Urease* activities indicated that the environment is more beneficial to the growth and generation of microorganisms, and better for the purification effect of pollutants [45]. Wang et al. [46] thought that *Nitrate Reductase* is an essential enzyme in the assimilation process of the exogenous nitrate. Therefore, it has close relationship with denitrification process [18]. In this study, it can be clearly observed that *Urease* activity of AMCMBR in different working conditions all exceeded AOA-MBR, confirming that AMCMBR achieved better COD removal efficiency than AOA-MBR. However, except for low C/N ratio (i.e. 6 and 4), there are no clear difference of *Nitrate Reductase* activities between AMCMBR and AOA-MBR. Therefore, enzyme activities analysis indicated that the AMCMBR has a greater performance on treating ship domestic wastewater.

4 Conclusions

In this study, two reactors were developed to compare the removal efficiency of COD and TN in treating ship domestic sewage under different COD concentration and C/N. Several results were listed as follows: (1) Throughout the experiment, the AMCMBR and AOA-MBR achieved good removal efficiency of COD and TN (Results indicated that the satisfactory removal efficiencies of COD and TN was achieved in the former stages (Re(COD)=91.35 % and 88.92 %; Re(TN)=87.54 % and 85.80 %). (2) Compared with AOA-MBR, the volume of AMCMBR is smaller than AOA-MBR; however AMCMBR performed more stable on COD and TN removal on ship wastewater treatment. (3) The MLVSS/MLSS of AMCMBR was higher than AOA-MBR in each stage, revealing that AMCMBR achieved high activity of microorganism and greater removal efficiencies in comparison with AOA-MBR. (4) AMCMBR has higher *Urease* activities, which also proved that AMCMBR has better pollutants removal capacity. All these conclusions indicated that AMCMBR is a practical system to treat ship sewage.

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References

[1] Sun, C., Leiknes, T., Weitzenböck, J., Thorstensen, B. "Development of an integrated shipboard wastewater treatment system using biofilm-MBR", Separation and Purification Technology, 75(1), pp. 22–31, 2010.
https://doi.org/10.1016/j.seppur.2010.07.005

[2] Feng, R., Zaidi, A. A., Zhang, K., Shi, Y. "Optimisation of Microwave Pretreatment for Biogas Enhancement through Anaerobic Digestion of Microalgal Biomass", Periodica Polytechnica Chemical Engineering, 63(1), pp. 65–72, 2019.
https://doi.org/10.3311/PPch.12334

[3] Zaidi, A. A., Feng, R., Shi, Y., Khan, S. Z., Mushtaq, K. "Nanoparticles augmentation on biogas yield from microalgal biomass anaerobic digestion", International Journal of Hydrogen Energy, 43(31), pp. 14202–14213, 2018.
https://doi.org/10.1016/j.ijhydene.2018.05.132

[4] IMO-MEPC "Revised guidelines on Implementation of Effluent Standards and Performance Tests for Sewage Treatment Plants", Committee MEP, I.M. Organization, 2006..

[5] IMO "International Convention for the Prevention of Pollution from Ships, 1973, as Modified by the Protocol of 1978 Relating Thereto (Marpol), I.M. Organization, 1973 modified 1978", 1978.

[6] IMO-MEPC "Revised Guidelines on Implementation of Effluent Standards and Performance Tests for Sewage Treatment Plants", Committee MEP, I.M. Organization, 2010.

[7] Sun, F. Y., Wang, X. M., Li, X. Y. "An innovative membrane bioreactor (MBR) system for simultaneous nitrogen and phosphorus removal", Process Biochemistry, 48(11), pp. 1749–1756, 2013.
https://doi.org/10.1016/j.procbio.2013.08.009

[8] Su, X., Wang, Y., Xue, B., Hashmi, M. Z., Lin, H., Chen, J., Wang, Z., Mei, R., Sun, F. "Impact of resuscitation promoting factor (Rpf) in membrane bioreactor treating high-saline phenolic wastewater: Performance robustness and Rpf-responsive bacterial populations", Chemical Engineering Journal, 357, pp. 715–723, 2019.
https://doi.org/10.1016/j.cej.2018.09.197

[9] Hu, D., Li, X., Chen, Z., Cui, Y., Gu, F., Jia, F., Xiao, T., Su, H., Xu, J., Wang, H., Wu, P., Zhang, Y. "Performance and extracellular polymers substance analysis of a pilot scale anaerobic membrane bioreactor for treating tetrahydrofuran pharmaceutical wastewater at different HRTs", Journal of Hazardous Materials, 342, pp. 383–391, 2018.
https://doi.org/10.1016/j.jhazmat.2017.08.028

[10] Habib, R., Asif, M. B., Iftekhar, S., Khan, Z., Gurung, K., Srivastava, V., Sillanpää, M. "Influence of relaxation modes on membrane fouling in submerged membrane bioreactor for domestic wastewater treatment", Chemosphere, 181, pp. 19–25, 2017.
https://doi.org/10.1016/j.chemosphere.2017.04.048

[11] Stazi, V., Tomei, M. C. "Enhancing anaerobic treatment of domestic wastewater: State of the art, innovative technologies and future perspectives", Science of The Total Environment, 635, pp. 78–91, 2018.
https://doi.org/10.1016/j.scitotenv.2018.04.071

[12] Khan, S. J., Ilyas, S., Ur-Rehman, Z. "Impact of nitrogen loading rates on treatment performance of domestic wastewater and fouling propensity in submerged membrane bioreactor (MBR)", Bioresource Technology, 141, pp. 46–49, 2013.
https://doi.org/10.1016/j.biortech.2013.03.196

[13] Song, D., Zhang, W., Liu, C., Wang, P., Sun, Z., Zhao, L., Zhai, X., Qi, J., Ma, J. "Development of a novel anoxic/oxic fed-batch membrane bioreactor (AFMBR) based on gravity-driven and partial aeration modes: A pilot scale study", Bioresource Technology, 270, pp. 255–262, 2018.
https://doi.org/10.1016/j.biortech.2018.08.049

[14] Hussain, A., Lee, J., Reid, R., Lee, H. S. "Upflow anaerobic-microaerobic fixed biofilm reactor integrating methanogenesis with partial nitrification", Chemical Engineering Journal, 348, pp. 281–291, 2018.
https://doi.org/10.1016/j.cej.2018.05.013

[15] Xia, S., Guo, J., Wang, R. "Performance of a pilot-scale submerged membrane bioreactor (MBR) in treating bathing wastewater", Bioresource Technology, 99(15), pp. 6834–6843, 2008.
https://doi.org/10.1016/j.biortech.2008.01.044

[16] Cai, Y. H., Ben T., Zaidi, A. A., Shi, Y., Zhang, K., Lin, A. Q., Liu, C. "Effect of pH on Pollutants Removal of Ship Sewage Treatment in an Innovative Aerobic-Anaerobic Micro-Sludge MBR System", Water, Air, & Soil Pollution, 230, Article number: 163, 2019.
https://doi.org/10.1007/s11270-019-4211-0

[17] Gao, M., Zhang, L., Florentino, A. P., Liu, Y. "Performance of anaerobic treatment of blackwater collected from different toilet flushing systems: Can we achieve both energy recovery and water conservation?", Journal of Hazardous Materials, 365, pp. 44–52, 2019.
https://doi.org/10.1016/j.jhazmat.2018.10.055

[18] Cai, Y., Li, X., Zaidi, A. A., Shi, Y., Zhang, K. Feng, R., Lin, A., Liu, C. "Effect of hydraulic retention time on pollutants removal from real ship sewage treatment via a pilot-scale air-lift multilevel circulation membrane bioreactor", Chemosphere, 236, Article number: 124338, 2019.
https://doi.org/10.1016/j.chemosphere.2019.07.069

[19] McNicholl, B. P., McGrath, J. W., Quinn, J. P. "Development and application of a resazurin-based biomass activity test for activated sludge plant management", Water Research, 41(1), pp. 127–133, 2007.
https://doi.org/10.1016/j.watres.2006.10.002

[20] Cai, Y., Ben, T., Zaidi, A. A., Shi, Y., Zhang, K. "Nitrogen removal augmentation of ship sewage by an innovative aerobic-anaerobic micro-sludge MBR technology", Process Biochemistry, 82, pp. 123–134, 2019.
https://doi.org/10.1016/j.procbio.2019.04.020

[21] Follmer, C. "Insights into the role and structure of plant ureases", Phytochemistry, 69(1), pp. 18–28, 2008.
https://doi.org/10.1016/j.phytochem.2007.06.034

[22] Unda-Calvo, J., Martínez-Santos, M., Ruiz-Romera, E., Lechuga-Crespo, J. L. "Implications of denitrification in the ecological status of an urban river using enzymatic activities in sediments as an indicator", Journal of Environmental Sciences, 75, pp. 255–268, 2019.
https://doi.org/10.1016/j.jes.2018.03.037

[23] Cai, Y., Li, X., Zaidi, A., Shi, Y., Zhang, K., Sun, P., Lu, Z. "Processing Efficiency, Simulation and Enzyme Activities Analysis of an Air-Lift Multilevel Circulation Membrane Bioreactor (AMCMBR) on Marine Domestic Sewage Treatment", Periodica Polytechnica Chemical Engineering, 63(3) pp. 448–458, 2019.
https://doi.org/10.3311/PPch.13389

[24] American Public Health Association Inc. "Standard Methods for the Examination of Water and Wastewater", APHA, New York, NY, USA, 1995.

[25] Zhang, F., Li, P., Chen, M., Wu, J., Zhu, N., Wu, P., Chiang, P., Hu, Z. "Effect of operational modes on nitrogen removal and nitrous oxide emission in the process of simultaneous nitrification and denitrification", Chemical Engineering Journal, 280, pp. 549–557, 2015.
https://doi.org/10.1016/j.cej.2015.06.016

[26] Li, J., Xing, X. H., Wang, B. Z. "Characteristics of phosphorus removal from wastewater by biofilm sequencing batch reactor (SBR)", Biochemical Engineering Journal, 16(3), pp. 279–285, 2003.
https://doi.org/10.1016/S1369-703X(03)00071-8

[27] Chen, Z., Hu, D., Ren, N., Zhang, Z. P. "Simultaneous removal of organic substances and nitrogen in pilot-scale submerged membrane bioreactors treating digested traditional Chinese medicine wastewater", International Biodeterioration & Biodegradation, 62(3), pp. 250–256, 2008.
https://doi.org/10.1016/j.ibiod.2008.01.010

[28] Huang, Z., Ong, S. L., Ng, H. Y. "Submerged anaerobic membrane bioreactor for low-strength wastewater treatment: Effect of HRT and SRT on treatment performance and membrane fouling", Water Research, 45(2), pp. 705–713. 2011.
https://doi.org/10.1016/j.watres.2010.08.035

[29] Zhang, S., Yang, F., Liu, Y., Zhang, X., Yamada, Y., Furukawa, K. "Performance of a metallic membrane bioreactor treating simulated distillery wastewater at temperatures of 30 to 45°C", Desalination, 194(1-3), pp. 146–155, 2006.
https://doi.org/10.1016/j.desal.2005.10.029

[30] Chae, S. R., Shin, H. S. "Characteristics of simultaneous organic and nutrient removal in a pilot-scale vertical submerged membrane bioreactor (VSMBR) treating municipal wastewater at various temperatures", Process Biochemistry, 42(2), pp. 193–198, 2007.
https://doi.org/10.1016/j.procbio.2006.07.033

[31] Monti, A., Hall, E. R., Dawson, R. N., Husain, H., Kelly, H. G. "Comparative study of biological nutrient removal (BNR) processes with sedimentation and membrane-based separation", Biotechnology and Bioengineering, 94(4), pp. 740–752, 2006.
https://doi.org/10.1002/bit.20896

[32] China National Standard "Discharge Standard for Water Pollutants from Ships, (GB 3552-2018)", Ministry of Ecology and Environment of the People's Republic of China, Taiwan, Republic of China, 2018.

[33] Yang, S., Yang, F., Fu, Z., Wang, T., Lei, R. "Simultaneous nitrogen and phosphorus removal by a novel sequencing batch moving bed membrane bioreactor for wastewater treatment", Journal of Hazardous Materials, 175(1-3), pp. 551–557, 2010.
https://doi.org/10.1016/j.jhazmat.2009.10.040

[34] Wang, X., Wang, S., Xue, T., Li, B., Dai, X., Peng, Y. "Treating low carbon/nitrogen (C/N) wastewater in simultaneous nitrification-endogenous denitrification and phosphorous removal (SNDPR) systems by strengthening anaerobic intracellular carbon storage", Water Research, 77, pp. 191–200, 2015.
https://doi.org/10.1016/j.watres.2015.03.019

[35] Yu, T., Qi, R., Li, D., Zhang, Y., Yang, M. "Nitrifier characteristics in submerged membrane bioreactors under different sludge retention times", Water Research, 44(9), pp. 2823–2830, 2010.
https://doi.org/10.1016/j.watres.2010.02.021

[36] Ma, W., Han, Y., Ma, W., Han, H., Zhu, H., Xu, C., Li, K., Wang, D. "Enhanced nitrogen removal from coal gasification wastewater by simultaneous nitrification and denitrification (SND) in an oxygen-limited aeration sequencing batch biofilm reactor", Bioresource Technology, 244, pp. 84–91, 2017.
https://doi.org/10.1016/j.biortech.2017.07.083

[37] Kinh, C. T., Suenaga, T., Hori, T., Riya, S., Hosomi, M., Smets, B. F., Terada, A. "Counter-diffusion biofilms have lower N_2O emissions than co-diffusion biofilms during simultaneous nitrification and denitrification: Insights from depth-profile analysis", Water Research, 124, pp. 363–371, 2017.
https://doi.org/10.1016/j.watres.2017.07.058

[38] Yang, S., Yang, F., Fu, Z., Lei, R. "Comparison between a moving bed membrane bioreactor and a conventional membrane bioreactor on organic carbon and nitrogen removal", Bioresource Technology, 100(8), pp. 2369–2374, 2009.
https://doi.org/10.1016/j.biortech.2008.11.022

[39] Chen, Z., Wang, H., Ren, N., Chen, Z., Cui, M., Nie, S. "Performance and model of a novel membrane bioreactor to treat the low-strengthen complex wastewater", Bioresource Technology, 114, pp. 33–45, 2012.
https://doi.org/10.1016/j.biortech.2012.02.026

[40] Meng, F., Chae, S. R., Drews, A., Kraume, M., Shin, H. S., Yang, F. "Recent advances in membrane bioreactors (MBRs): Membrane fouling and membrane material", Water Research, 43(6), pp. 1489–1512, 2009.
https://doi.org/10.1016/j.watres.2008.12.044

[41] Fan, J., Ji, F., Xu, X., Wang, Y., Yan, D., Xu, X., Chen, Q., Xiong, J., He, Q. "Prediction of the effect of fine grit on the MLVSS/MLSS ratio of activated sludge", Bioresource Technology, 190, pp. 51–56, 2015.
https://doi.org/10.1016/j.biortech.2015.04.035

[42] Chen, Z., Su, H., Hu, D., Jia, F., Li, Z., Cui, Y., Ran, C., Wang, X., Xu, J., Xiao, T., Li, X., Wang, H. "Effect of organic loading rate on the removal of DMF, MC and IPA by a pilot-scale AnMBR for treating chemical synthesis-based antibiotic solvent wastewater", Chemosphere, 198, pp. 49–58, 2018.
https://doi.org/10.1016/j.chemosphere.2018.01.091

[43] Ng, K. K., Shi, X., Yao, Y., Ng, H. Y. "Bio-entrapped membrane reactor and salt marsh sediment membrane bioreactor for the treatment of pharmaceutical wastewater: Treatment performance and microbial communities", Bioresource Technology, 171, pp. 265–273, 2014.
https://doi.org/10.1016/j.biortech.2014.08.078

[44] Wu, Z., Xu, F., Yang, C., Su, X., Guo, F., Xu, Q., Peng, G., He, Q., Chen, Y. "Highly efficient nitrate removal in a heterotrophic denitrification system amended with redox-active biochar: A molecular and electrochemical mechanism", Bioresource Technology, 275, pp. 297–306, 2019.
https://doi.org/10.1016/j.biortech.2018.12.058

[45] Li, M., Liang, Z., Callier, M. D., Roque d'Orbcastel, E., Ma, X., Sun, L., Li, X., Wang, S., Song, X., Liu, Y. "Nitrogen and organic matter removal and enzyme activities in constructed wetlands operated under different hydraulic operating regimes", Aquaculture, 496, pp. 247–254, 2018.
https://doi.org/10.1016/j.aquaculture.2018.06.016

[46] Wang, D., Li, T., Huang, K., He, X., Zhang, X. X. "Roles and correlations of functional bacteria and genes in the start-up of simultaneous anammox and denitrification system for enhanced nitrogen removal", Science of The Total Environment, 655, pp. 1355–1363, 2019.
https://doi.org/10.1016/j.scitotenv.2018.11.321