

Effect of Sewage Load on Microenvironment and Sludge Reduction Efficiency of in Situ Biofilm Sludge Reduction System

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Abstract: The load has significant effect on sludge reduction efficiency and biofilm microenvironment. When the load was 1 ~ 4 kgCOD/ (m³• d), the sludge yield of the system changed little. When the system was greater than 4 kgCOD/ (m³•D), the sludge yield of the system increased doubly and the maximum load of the system was 4 kgMLSS/ (m³•D). At this time, the excess sludge yield is 0.0292 kgMLSS/kgCOD. The ORP aeration and aeration end of the biofilm were respectively (-74.73 ~ 187.89mV) and (-53.21 ~ 260.74mV). The corresponding biofilm aerobic and anoxic environment accounted for 9%, 17% and 91% and 83% respectively, forming a microenvironment of alternating aerobic / anoxic environment. When the load is 4kgCOD/ (m³•D), the change of ATP in sludge in the aeration stage and the shutdown stage is the largest, which is conducive to the dissipation of energy. To promote the sludge reduction, the COD, NH₄⁺-N, TN and SS indexes of the effluent of the system all reached the first grade B standard of the pollutant discharge standard for urban sewage treatment (GB18918-2002).

1. Introduction

With the continuous increase of municipal sewage treatment capacity, the output of surplus sludge has increased dramatically[1]. A large number of excess sludge treatment is facing the problems of high cost, difficulty and two pollution. The investment and operation cost of treatment and disposal accounts for about 25% to 65% of the operation cost of sewage treatment plant, which brings heavy economic burden to the sewage treatment plant [2]. Therefore, minimizing sludge production in the process of sewage treatment has become an important problem to be solved in the field of sewage treatment at home and abroad [3-5].

Because of the existence of the carrier, the sludge concentration of the biofilm can be as high as 6000 ~ 7000mg/L, which is beneficial to the survival of some slow microorganism and the production of sludge in the system. At the same time, a longer food chain can achieve better sewage treatment efficiency [6]. Du Zhenzhong and other multistage AO biofilm reactors were constructed, and spherical porous microorganism carrier was added to the multistage AO reactor, and the sludge yield was reduced by 90% compared with the traditional activated sludge process. Oliveira et al[7]. Uses ASSR + MBR process to reduce sludge reduction to 74%, while reducing biological nutrient removal rate and reducing membrane fouling [8]. Cheng Cheng and other studies found that ASSR-MBR achieved sludge reduction from 6% to 49.7%, and also can efficiently remove COD and NH₄-N[9]. Gu and so on developed a A-2B process. Compared with the traditional activated sludge process, the energy demand of nitrogen removal was reduced by 47%, and the output of excess sludge decreased by 75%. Yao Meng et al [10]. Compared the sludge yield of SBBR and SBR and CASS processes with fillers. The results showed that the sludge yield of SBBR process was the smallest, 0.315kgMLSS/kgCOD, and the best sewage treatment effect. Sludge reduction is also achieved through simple ways of adding metabolic uncoupling agents (MU), but this way is

likely to cause sludge settling performance to deteriorate. Niu et al[11]. Studied the effect of DO concentration on SPRAS sludge reduction and microbial community structure in a microaerobic tank. It was found that when the DO decreased from 2.5 to 0.5 mg / L, the sludge reduction efficiency increased from 42.9% to 68.3%. Biofilm process can achieve better sludge reduction efficiency [12]. The current research focuses on the macro comparison of the reduction effect. However, there is a lack of relevant factors for its stable operation, especially the influence of control factors on the internal environment of biofilm.

In the early stage of this study, a sludge reduction system in situ was built. In this study, the SBBR sludge reduction process was used as the research object, by changing the sewage load of the system. The effects of different load conditions on the reduction efficiency, the efficiency of sewage treatment, the ORP microenvironment in biofilm and the ATP in the sludge were investigated, which laid the foundation for the application of this reduction technology.

2. Experimental device and method (Materials and methods)

2.1 Experimental device

The experimental device is shown in Figure 1. The SBBR (Sequencing Batch Biofilm Reactor) reactor is used as the reactor and the bottom is equipped with a sludge discharge bucket with an effective volume of 10L. The semi soft composite packing was installed in the reactor, and the film density was 45%. The oxygen supply pump is supplied by the sand head to the reaction zone, and the oxygen charging process is controlled by the time controller. The constant temperature heating rod is adopted to keep the temperature in the reactor stable.

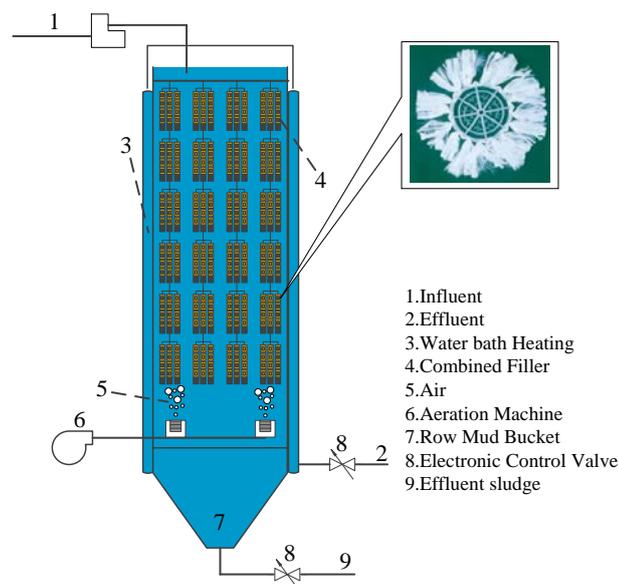


Fig. 1 Experiment equipment

The operating conditions of the reactor are instantaneous influent, reaction 6.5h, precipitation 0.5h, instantaneous drainage, and periodic reaction time of 7.0h. During the reaction, intermittent aeration and 30min/ aeration of 30min were used alternately. The effect of influent load on operational efficiency of SBBR in situ sludge reduction system was investigated by parallel test. The test temperature is 25, the film density is 45%, the gas supply is 150 L / h, and the control inlet load is 1.0kgCOD/(m³·d), 2.0kgCOD/(m³·d), 3.0kgCOD/ (m³·d), 4.0kgCOD/.

2.2 Test water quality (Raw wastewater and physiochemical analysis)

The domestic sewage was treated by adding glucose to control the influent concentration of the reactor so as to form different influent load of the reactor. The test water quality is shown in Table 1.

Table 1 Influent quality of the experiment

| Index (mg/L) | load (kgCOD/(m ³ ·d)) | | | | |
|----------------------------------|----------------------------------|-------------|-------------|--------------|--------------|
| | 1.0 | 2.0 | 3.0 | 4.0 | 5.0 |
| COD | 300 ±100 | 599 ±100 | 880 ±100 | 1193 ±100 | 1482 ±100 |
| NH ₄ ⁺ -N | 49±7 | 57.2±7 | 58.4±7 | 57±7 | 56±7 |
| TN | 62±15 | 72.4±15 | 73.4±15 | 74±15 | 70±15 |
| PO ₄ ³⁻ -P | 4±0.8 | 4±0.8 | 4±0.8 | 4±0.8 | 4±0.8 |
| SS | 75±30 | 87±30 | 92±30 | 96±30 | 100±30 |

2.3 Analysis of surplus sludge yield and water quality

After stable operation of the system, the precipitated sludge in the reactor funnel is used as surplus sludge once a week, and the system does not discharge sludge. At the same time, COD, NH₄⁺-N, TN, PO₄³⁻-P and TP were tested to analyze the sludge reduction effect and sewage treatment efficiency under different influent loading conditions.

2.4 ATP analysis of internal redox potential and energy in biofilms

The biofilm was removed from the stable reactor. The biofilm was placed in the beaker, simulating the reaction conditions in the reactor, and measuring the change of ORP in the biofilm. At the same time, the sludge ATP during the operation of different load reactors was measured to analyze the changes of biofilm microenvironment under different influent loading conditions.

2.5 Analysis method

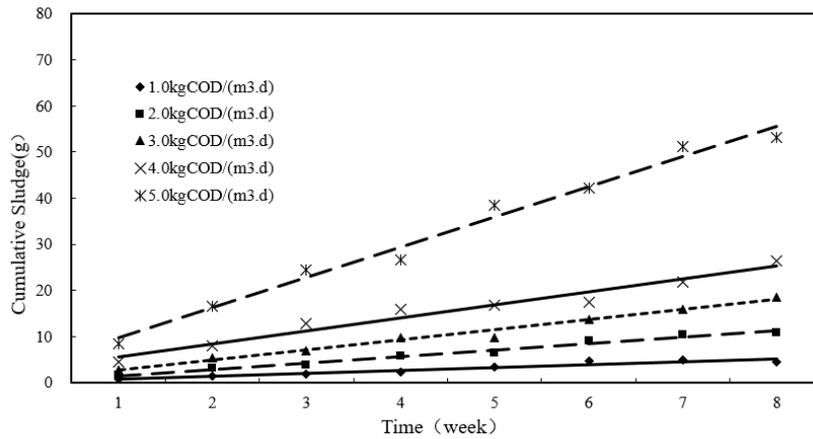
In this study, the ORP in the biofilm was measured by Unisense microelectrode analysis system. The content of ATP in biofilm sludge was measured by luciferase assay to study the energy metabolism of organisms in biofilm. The concentration of various water quality parameters will affect the selection and function of the treatment process (Sun et al). 2016). Therefore, systematic analysis of effluent quality is very important. In water COD, potassium dichromate method (HACH-COD) and NH₄⁺-N content were determined by Nash reagent spectrophotometry. NO₃-N content was determined by 0.45 - M membrane filtration and UV spectrophotometry. TN was treated by alkaline potassium persulfate digestion UV spectrophotometry. The content of PO₄³⁻-P is determined by antimony antimony spectrophotometry. TP was dissolved by potassium persulfate molybdenum antimony spectrophotometric method.

3. Experimental results and discussion

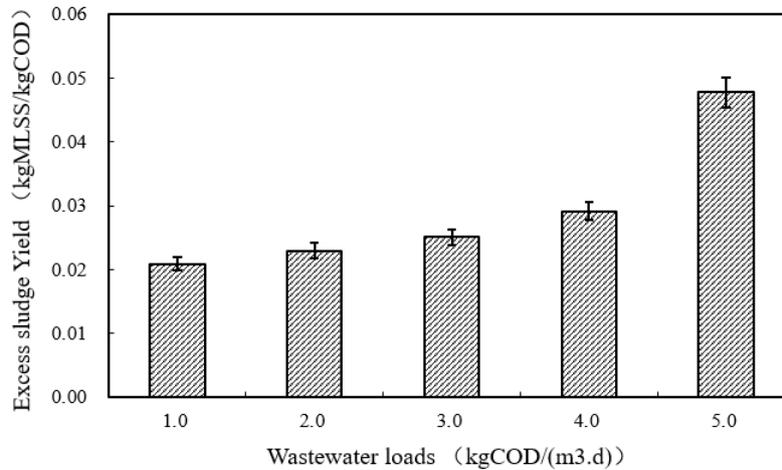
3.1 The effect of load on the sludge reduction efficiency of the system

3.1.1 The effect of load on the sludge yield of the system

Load is an important control parameter in the process of sewage treatment, and has a direct impact on the sludge yield. As shown in Figure 2 (a), with the increase of load, the cumulative surplus sludge yield increased gradually. When the load was 4 kgCOD/ (m³· d), the sludge yield of the system was 0.0292 kgMLSS/kgCOD. When the load is less than 4 kgCOD/(m³· d), the sludge yield of the system has little change, and when the load is more than 4 kgCOD/(m³· d), the sludge yield of the system increases exponentially. Figure 2 (b) showed that when the influent load was 1, 2, 3 and 4 kgCOD/(m³· d), the average sludge yield of the system was 0.0208, 0.0230, 0.0250 and 0.0292 kgMLSS/kgCOD, and the load per 1kgCOD/(m³· d) increased, and the increase of sludge yield was stable in 8% ~ 10%. When the organic load continued to increase to 5 kgCOD/(m³· d), the average sludge yield of the system increased significantly, which was 0.0477kgMLSS/kgCOD, and increased by 78% than that of 4 kgCOD/(m³· d). The test results show that when the load is greater than 4 kgCOD/ (m³· d), it is not conducive to the in situ sludge reduction of the system, that is, the maximum load of the system is 4 kgCOD/(m³· d).



(a) The effect of load on the amount of accumulated excess sludge



(b) Effect of load on average sludge yield

Fig.2 Effect of loading on efficiency of sludge reduction in the reactor

3.1.2 Effect of load on ORP microenvironment in biofilm

The experimental study found that the biofilm formed under different load conditions is different. With the increase of load, the color of biofilm gradually changed from brown to black. This is due to the different oxygen consumption rates of biofilms under different load conditions. From Fig. 3, we can see that under the condition of 150L/h supply volume, the DO of liquid phase in different load reactors will increase during the aeration stage. The trend of decreasing during the shutdown stage is regular. However, the thickness of biofilm in reactor increased with the increase of load. When the load increased from 1 kgCOD/(m³·d) to 3 kgCOD/(m³·d). The thickness of biofilm is increased from 0.8cm to 1.2cm, and the high load influent reactor provides sufficient nutrition for the growth of biofilm, which is beneficial to the growth and accumulation of biofilm.

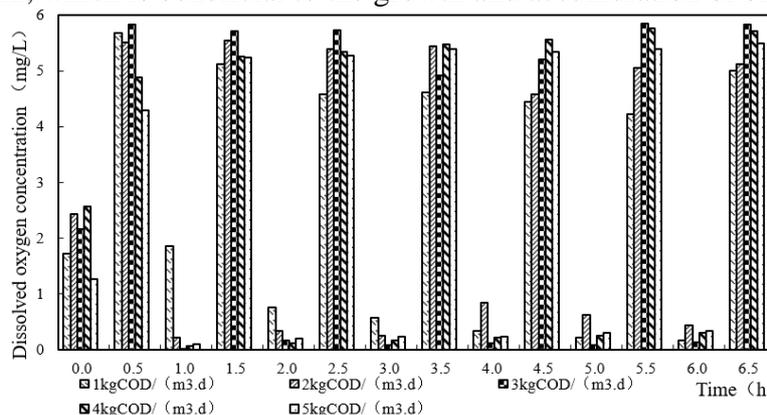
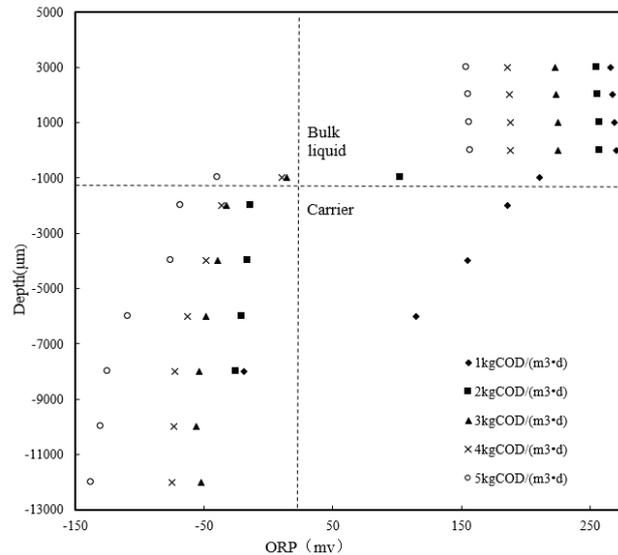
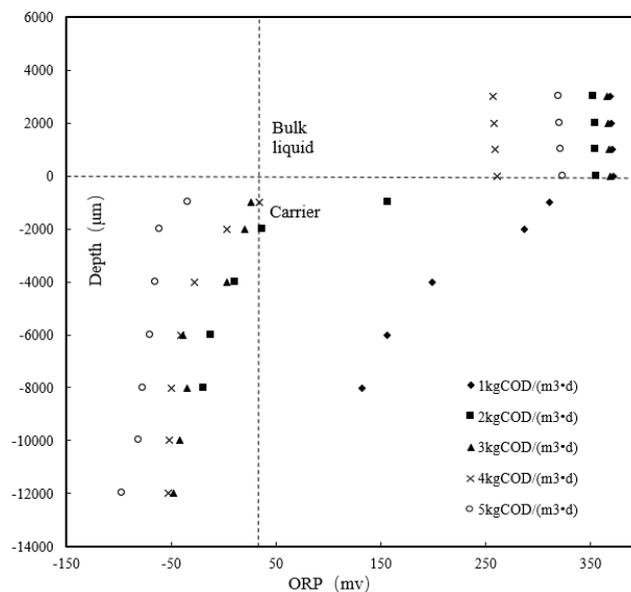


Fig. 3 Effect of loading on DO in the running period of reactor

To further investigate the effect of load on the internal microenvironment of biofilms, microelectrodes were used to monitor ORP in biofilms during aeration and shutdown. As shown in Fig. 4, load has a significant effect on the ORP microenvironment inside the biofilm system. When the load is $1.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the biofilm thickness is 0.8cm , and the ORP inside the biofilm is -19.24 to 270.55mV when aeration starts. The thickness of biofilm aerobic layer is 0.6cm , the thickness of the anoxic layer is 0.2cm , and the aerobic anoxic environment accounts for 75% and 25% respectively. At the end of aeration, the ORP in the biofilm was 132.14 to 372.07mV , and the biofilm was completely aerobic.



(a) The internal ORP of the biofilm during the reaction of 0h (aeration)



(b) The internal ORP of the biofilm at the reaction 0.5h (end of aeration)

Fig. 4 Effect of loading on ORP inside the biofilm

When the load is $2.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the biofilm thickness is 0.8cm , and the ORP inside the biofilm starts from -24.99 to 257.24mV at the beginning of aeration. The thickness of the aerobic layer is 0.1cm , the thickness of the anoxic layer is 0.7cm , and the aerobic anoxic environment accounts for 12.5% and 87.5% respectively. At the end of aeration, the ORP of the biofilm was -18.43 to 356.06mV , the thickness of the aerobic layer was 0.4cm , the thickness of the anoxic layer was 0.4cm , and the aerobic anoxic environment accounted for 50% . When the load is $3.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the thickness of the biofilm is 1.2cm . The internal ORP of the biofilm is -52.21 to 224.60mV at the beginning of aeration, the thickness of the aerobic layer is 0.1cm , the thickness of the anoxic layer is 1.1cm , and the good anoxic environment is 9% and 91% , respectively. At the

end of aeration, the ORP of the biofilm was -48.21 to 368.27mV, the thickness of the aerobic layer was 0.4cm, the thickness of the anoxic layer was 0.8cm, and the aerobic anoxic environment accounted for 33% and 67% respectively. When the load is 4.0kgCOD/(m³• d), the thickness of the biofilm is 1.2cm. The internal ORP of the biofilm is -74.73 to 187.89mV at the beginning of aeration, the thickness of the aerobic layer is 0.1 cm, the thickness of the anoxic layer is 1.1 cm, and the good anoxic environment is 9% and 91%, respectively. At the end of aeration, the ORP of the biofilm was -53.21 to 260.74mV, the thickness of the aerobic layer was 0.2cm, the thickness of the anoxic layer was 1.0cm, and the aerobic anoxic environment accounted for 17% and 83% respectively. When the load increased to 5.0kgCOD/(m³• d), the internal membrane of the biofilm was both anoxic at the beginning and end of aeration, and the anoxic environment accounted for 100%. From the above analysis, with the increase of load and the beginning and end of aeration, the interior of the biofilm showed a gradual decrease in aerobic environment and an increasing trend in the anoxic environment. At the same time, when the load is 1.0kgCOD/(m³• d) to 4.0kgCOD/(m³• d), the aerobic environment of the organism increases at the beginning of aeration at the end of aeration, forming a microenvironment of aerobic / anoxic alternation.

The analysis indicates that the mass transfer capacity of dissolved oxygen in biofilm determines the proportion of oxygen, anoxia and anaerobic zone. The microenvironment formed is an important factor affecting the degree of dominance of microbial populations in biofilms. In the system, there are DO and organic matter transported in the outer layer of the biofilm, which can carry out aerobic biodegradation. With the depletion of organic matter and DO in the outer layer of biofilm, the inner layer of biofilm formed a region where some of the undepleted DO had no organic matter. In this area, microorganism degrades the hydrolysate of dead cells as substrate for biodegradation. As DO was consumed in the process of transmission, an anaerobic zone appeared in the innermost layer of the biofilm. The microorganisms in this area are mainly inert bacteria, and the hydrolysis process of the dead bacteria is carried out. The organic matter produced by hydrolysis can spread to the outer layer of the biofilm for biological oxidation. Therefore, when the organic loading is low, the DO consumption of organic film degradation on the outer layer of biofilm is less. As shown in Figure 4, the remaining DO can be diffused deeply, and the redox potential in the biofilm is higher, and a better aerobic microenvironment is formed when the load is 1 kgCOD/(m³• d). With the increase of organic load, the organic matter adsorbed on the outer layer of the biofilm increased, and the DO transferred from the liquid phase was almost consumed by the degradation of the outer organic matter. The depth diffusion of DO is limited, so when the load is controlled at 2 kgCOD/(m³• d) to 5 kgCOD/(m³• d), the anoxic region within the biofilm increases gradually and even appears in the whole anoxic region.

3.1.3 The effect of load on the internal ATP of biofilm

The change rule of ATP in periodic operation under different load conditions is shown in Figure 5.

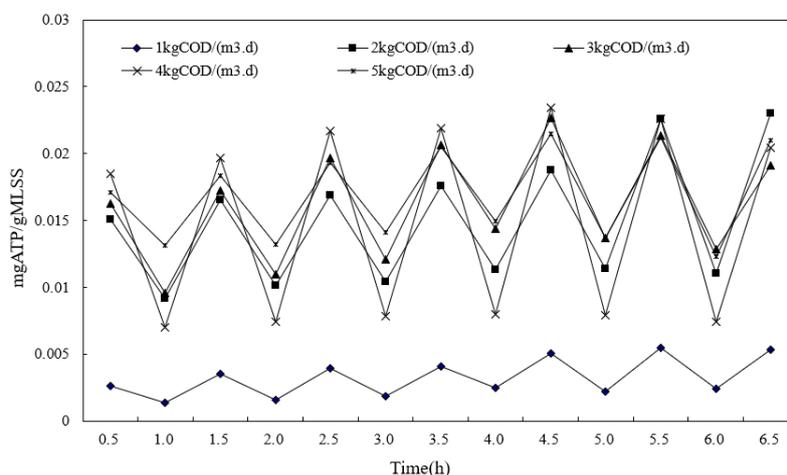


Fig.5 Effect of loading on ATP inside the biofilm

According to figure 5, the sludge ATP in the system varies regularly under different loads. When the load is from 1 to 5kgCOD/(m³• d), the content of ATP is 0.0026~0.0055mgATP/gMLSS, 0.0151~0.0226mgATP/gMLSS, 0.0162~ 0.0227mgATP/gMLSS, 0.0185~0.0234mgATP/ gMLSS and 0.0171~0.0215mgATP/gMLSS, respectively. The levels of ATP were 0.0014~0.0025mgATP/gMLSS, 0.0092~0.0114mgATP/gMLSS, 0.0096~0.0143mgATP/gMLSS, 0.0071~0.0081mgATP/gMLSS and 0.0131~0.0149mgATP/gMLSS, respectively.

It is known that when the load is 1 ~ 4kgCOD/(m³• d), the ATP content in the sludge increases gradually with the increase of the load. When the load increased to 4kgCOD/(m³• d), the change of ATP in sludge in the aeration and shutdown stage was the largest. When the load continued to increase to 5kgCOD/(m³• d), the ATP in the aeration stage and the stopping stage of the sludge decreased slightly, and showed a trend of reducing the degree of change of ATP in different oxygen filling stages.

The analysis shows that the SBBR operation mode of intermittent oxygen supply is adopted in this experiment. It can be seen from the analysis of the change of ORP microenvironment in the biofilm mentioned above. The alternate operation of aeration and stopping aeration changes the oxygen environment conditions of microorganism alternately, and the biomembrane forms a good aerobic and anoxic alternation microenvironment, and the ATP content in the microbial cells depends on the environmental conditions. Therefore, the change of ATP shows the regular fluctuation of the aeration stage and the decrease of the exposure stage. ATP is an important parameter for controlling bacterial activity in activated sludge. As a substance storing energy in living cells, when cells die quickly, it can well characterize the activity of cells in the system. The increase of organic loading increased the matrix of carrier, resulting in the increase of substrate concentration and metabolic activity. Therefore, when the load increased from 1 kgCOD/(m³• d) to 4kgCOD/(m³• d), the ATP content in the sludge increased gradually during aeration stage. But when the thickness of the biofilm thickness is the best, with the increase of the thickness of the biofilm, the biofilm attached to the most inner layer of the fiber filler is difficult to obtain enough nutrients and DO, and the activity of the biofilm is poor. Therefore, when the load continued to rise to 5kgCOD/(m³• d), the ATP content in the biofilm decreased. In different aeration stages, the change of oxygen environment is not obvious, resulting in the reduction of ATP.

According to the above research, intermittent aeration system has achieved a good reduction effect. According to the analysis, aerobic environment is formed during aeration stage, so that the energy generated by microorganism can be stored in ATP through oxidative phosphorylation process, resulting in the increase of ATP. It provides energy for anabolism. When the aeration stops, an anoxic environment is formed. The change of environment makes the process of oxidative phosphorylation of cells inhibited. So that the energy generated by catabolism can be transformed into heat, and can not be converted into ATP necessary for anabolism. As a result, ATP decreases, and this alternately running environment makes microbial biosynthesis and catabolism no longer coincide. Thus, the sludge yield of the system is reduced, and in situ sludge reduction is achieved.

3.2 Effect of load on the efficiency of sewage treatment

Table 2 The testing effect of loading on wastewater treatment

| Indicators (mg / L) | load(kgCOD/(m ³ .d)) | | | | | | | | | |
|----------------------------------|---------------------------------|------------------|----------|------------------|----------|------------------|----------|------------------|----------|------------------|
| | 1.0 | | 2.0 | | 3.0 | | 4.0 | | 5.0 | |
| | Effluent | Removal rate (%) | Effluent | Removal rate (%) | Effluent | Removal rate (%) | Effluent | Removal rate (%) | Effluent | Removal rate (%) |
| COD | 37 | 87.7 | 37 | 93.8 | 39 | 95.6 | 41 | 96.6 | 57 | 96.2 |
| NH ₄ ⁺ -N | 3.8 | 92.2 | 3.7 | 93.5 | 3.8 | 93.5 | 6.4 | 88.8 | 17.7 | 68.7 |
| TN | 19.2 | 69.0 | 14.6 | 79.8 | 13.9 | 81.1 | 17.3 | 76.8 | 33.5 | 52.1 |
| PO ₄ ³⁻ -P | 1.8 | 55.0 | 1.7 | 57.5 | 1.7 | 57.5 | 1.6 | 60.0 | 2.5 | 37.5 |
| SS | 4.3 | 94.3 | 7.2 | 91.7 | 5.4 | 94.1 | 6.9 | 92.8 | 13.1 | 86.9 |

It can be seen from table 2 that when the load is less than 4.0kgCOD/(m³• d), COD, NH₄⁺-N, TN and SS have reached the first class B standard of the pollutant discharge standard of urban sewage treatment (GB18918-2002), and the load has little effect on the removal of COD. When the load is

greater than $4.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the removal of $\text{NH}_4^+\text{-N}$, TN and $\text{PO}_4^{3-}\text{-P}$ is significantly affected. When the load was increased from $1.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$ to $4.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the removal rate of COD, TN and $\text{PO}_4^{3-}\text{-P}$ increased by 8.9%, 7.8% and 5% respectively, and the removal rate of $\text{NH}_4^+\text{-N}$ decreased by 3.4%. When the load was $5\text{ kgCOD}/(\text{m}^3 \cdot \text{d})$, the COD removal changed little when the load was $4.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, and the removal rate of $\text{NH}_4^+\text{-N}$, TN and $\text{PO}_4^{3-}\text{-P}$ decreased by 19.9%, 24.7% and 22.5% respectively.

It is believed that the process of nitrogen removal in this system may be accomplished through the following ways: (1) the environment exists simultaneously in the aerobic and anoxic environment of the biofilm, and the removal of nitrogen by simultaneous nitrification and denitrification at the stage of aeration is realized. (2) during the aeration stage, nitrification and aeration ceasing stage will further intensify denitrification and denitrification. Therefore, the removal rate of $\text{NH}_4^+\text{-N}$ and TN is higher.

The cyclic operation of PO_4^{3-} in the reactor under various load conditions is shown in Figure 6. When the load is $1 \sim 5.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the PO_4^{3-} concentration in the system is in continuous decline during the whole cycle operation. But when the load was $5.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the degradation rate of PO_4^{3-} was slower than that of $1.0\text{kgCOD}/(\text{m}^3 \cdot \text{d}) \sim 4.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, and the PO_4^{3-} concentration of effluent was higher, and no phosphorus release was found under different load conditions. It is considered that the special oxygen supply method makes the system a certain anaerobic and anoxic environment, facultative anaerobes can convert inorganic phosphorus into PH_3 , and new biological phosphorus removal can be found in the system. Therefore, the effect of organic loading on phosphorus removal in the system needs further study.

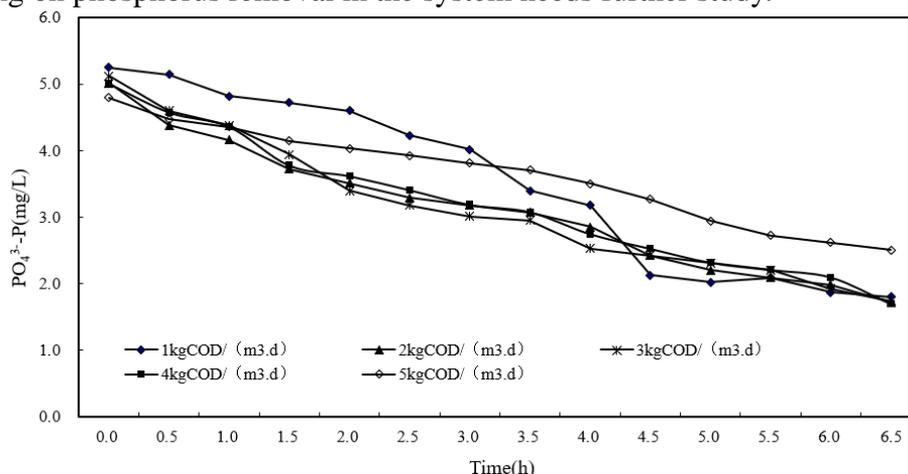


Fig. 6 Effect of loading on $\text{PO}_4^{3-}\text{-P}$ removal in the reactor

4. Conclusion

(1) The effect of load on biofilm sludge reduction system in situ is significant. When the load was $4\text{ kgCOD}/(\text{m}^3 \cdot \text{d})$, the sludge yield of the system was $0.0292\text{ kgMLSS}/\text{kgCOD}$. When the load is less than $4\text{ kgCOD}/(\text{m}^3 \cdot \text{d})$, the sludge yield of the system has little change, and when the load is more than $4\text{ kgCOD}/(\text{m}^3 \cdot \text{d})$, the sludge yield of the system increases exponentially. Therefore, the maximum load of the system is $4\text{ kgCOD}/(\text{m}^3 \cdot \text{d})$.

(2) The effect of load on the biofilm microenvironment of the system was significant. With the increase of load, the aerobic environment inside the biofilm of reactor decreased gradually, and the anoxic environment gradually increased. When the load was $4.0\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the aeration of ORP and the end of aeration were respectively $(-74.73 \text{ to } 187.89\text{mV})$ and $(-53.21 \sim 260.74\text{mV})$. The corresponding biofilm aerobic and anoxic environment accounted for 9%, 17% and 91% and 83% respectively, forming a microenvironment of alternating aerobic / anoxic environment. With the increase of load, the sludge ATP in the system showed a regular increase in the aeration stage and a decrease in the shutdown stage. When the load is $4\text{kgCOD}/(\text{m}^3 \cdot \text{d})$, the ATP changes in the sludge at the aeration stage and the shutdown stage.

The effect of the removal efficiency of the load reactor is significant. When the load was 4kgCOD/(m³• d), the effluent COD, NH₄⁺-N, TN and SS were 41, 6.4, 17.3 and 6.9mg/L respectively, and all reached the first class B standard of the pollutant discharge standard for urban sewage treatment (GB18918-2002).

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