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# Pilot-scale experiments on brewery wastewater treatment and sludge reduction based on food chain predation

Tianlong Zheng[a], Peng Li[a], Qunhui Wang[a,*], Xuesong Li[b], Hengyu Ai[c], Kemin Gan[a], Ariunbileg Sharavsambuu[a]

[a]*Department of Environmental Engineering, University of Science and Technology Beijing, 30 Xueyuan Road, Haidian District, Beijing 100083, China, Tel./Fax: +86 010 62332778; emails: skytal_03@sina.com (T. Zheng), ronnie2323@sina.com (P. Li), wangqh59@sina.com (Q. Wang), tian-persona@163.com (K. Gan), shariunbileg@yahoo.com (A. Sharavsambuu)*
[b]*School of Water Resources and Environment, China University of Geosciences (Beijing), 29 Xueyuan Road, Haidian District, Beijing 100083, China, Tel./Fax: +86 010 82376239; email: seader2004@163.com*
[c]*Department of Environmental Engineering, Harbin University of Science and Technology, 52 Xuefu Road, Nan Gang District, Harbin 150080, China, Tel./Fax: +86 0451 86392714; email: aihengyu@hit.edu.cn*

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## ABSTRACT

In a pilot-scale experiment, hydrolyzation-food chain reactor (H-FCR) system was used to treat brewery wastewater. The performance of the system, including COD, $BOD_5$, $NH_3$-N, SS removal, and sludge reduction, were investigated. The mechanism of sludge reduction and the characteristics of biological community during multilevel contact oxidation process were also explored. The food chain reactor (i.e. four-level contact oxidation reactor with a volume ratio of 4:3:3:2) was found to be contributory to the provision of a suitable environment for the formation of the food chain of bacteria–protozoa–metazoan–larger metazoan. The ratio of metazoan density to protozoa density and metazoan density to bacteria density increased gradually with COD concentration reduction along the flow direction. This change strengthened the role of predation in reducing sludge production from the source. The sludge production decreased to 8.15% kg suspended sludge/(kg COD removed). The formation of the food chain also provided ecological basis for the stable and efficient removal of contaminants. When the mass concentrations of COD, $BOD_5$, $NH_3$-N, and SS in influent water were 1585 ± 168, 711 ± 146, 43.1 ± 12.5, and 206.7 ± 53 mg/L, respectively; and when the hydraulic retention time of the H-FCR system was 11.7 h (5.2 h for the hydrolyzation segment and 6.5 h for the four-level contact oxidation segment), the average removal rate of the above four indicators were approximately 94.9, 97.9, 87.6, and 93.6%, respectively. Water quality of effluent conformed to discharge standard of pollutants for beer industry (GB 19821-2005). Therefore, the H-FCR system is an effective method for the treatment of wastewater from food industry, including breweries, because of its high efficiency, low consumption, and little excess sludge.

*Keywords:* Hydrolyzation; Multilevel contact oxidation; Sludge reduction; Food chain reactor

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*Corresponding author.

## 1. Introduction

As beer production increased as a result of the rapid development of the brewery industry, large amounts of brewery wastewater has been widely discharged around the world. China has become one of the top five brewery wastewater making countries, which reaches as high as 400 million cubic meters annually. Brewery wastewater contains a large amount of protein, fat, fiber, carbohydrates, yeast, hops residue, and so on [1]. If not properly handled, these materials will cause serious water pollution [2,3]. Therefore, the reduction of organic matter in wastewater is the primary task involved in the conservation of water quality. During the early 1980s, brewery wastewater was treated using aerobic techniques both at home and abroad [4]. However, the high energy consumption and operation costs of these techniques eventually became an economic burden for industrial wastewater treatment plants. Given the need for a new water treatment technology, that is energy saving, the anaerobic hydrolysis process [5] for brewery wastewater treatment was introduced. After the introduction of the new technique, approximately 30–50% of energy was saved and the effluent met emission standards easily and reliably.

Today, the main techniques for brewery wastewater treatment are the anaerobic-SBR method [6], UASB-activated sludge process [7,8], MBR [9,10], and hydrolytic-aerobic techniques [11–14]. Sludge production is about 60% of the COD removal amount for conventional activated sludge technology and about 30% for conventional biofilm method [15]. However, the expenses [16,17] that sludge disposal entails have also become an economic burden for producers. Therefore, sludge reduction for the prevention of secondary pollution has become a focal point of studies on wastewater treatment in recent years.

Recently, the hydrolyzation–multilevel contact oxidation process for sludge reduction, especially based on food chain predation [18–22], has attracted increasing attention. Food chain reactor (FCR) could provide a suitable environment for the formation of the food chain of bacteria–protozoa–metazoan–larger metazoan and the formation of the food chain into activated sludge to reduce the sludge production that offers interesting perspectives. Wang et al. [21] used a three-level contact oxidation reactor to treat kitchen wastewater. Their research realized biophase separation and obtained high organic matter removal as well as low sludge production (0.13–0.22 kg sludge/kg COD). Ratsak and Verkuijlen [22] studied the reduction of excess sludge in wastewater treatment plants by predatory activity of *aquatic oligochaetes* and also existed the similar phenomenon in the activated sludge which consisted of inorganic and organic substances, bacteria, protozoa, and metazoan. Li et al. [20] used a hydrolyzation-food chain reactor (H-FCR) to treat dairy wastewater; the excess sludge production was reduced to as low as 7.7%. Despite the availability of several studies, the mechanisms of sludge reduction by hydrolyzation–multilevel contact oxidation process had never been reported during the brewery wastewater treatment.

In the current study, a pilot-scale H-FCR, also called a hydrolyzation–four-level contact oxidation reactor, was used to treat brewery wastewater. First, the hydraulic retention time (HRT) of the hydrolyzation segment and the FCR was determined based on the change of biodegradability and the removal efficiency of COD and ammonia. Then, the removal of contaminants in the wastewater, such as COD, $BOD_5$, $NH_3$-N, and SS as well as reduction sludge, were investigated continuously for the entire duration of the study. The mechanism of sludge reduction, including its relationship with COD removal in the FCR segment, was also investigated through the distribution and the predation relation of the biological community.

## 2. Material and methods

### 2.1. Source and characteristics of wastewater

The experimental wastewater was obtained from a brewery plant. The biodegradability of the wastewater (shown as $BOD_5$/COD) was about 0.45. Table 1 shows the main properties of water quality.

Table 1
Parameters of raw brewery wastewater

| Parameter | Range of values | Average value ± SD of 27 samples |
|---|---|---|
| COD (mg/L) | 1,020–1,880 | 1,585 ± 168 |
| $BOD_5$ (mg/L) | 475–874 | 711 ± 146 |
| $NH_3$-N (mg/L) | 34.5–63.6 | 43.1 ± 12.5 |
| Total Nitrogen (mg/L) | 46.3–90.2 | 67.4 ± 15.3 |
| Total phosphates (mg/L) | 7.6–13.4 | 10.2 ± 1.6 |
| Suspended sludge (mg/L) | 150–300 | 206.7 ± 53 |
| Temperature (℃) | 14–27 | 22.2 ± 4.5 |
| pH | 7.0–8.5 | 7.5 ± 0.5 |

Note: SD is abbreviation of standard deviation.

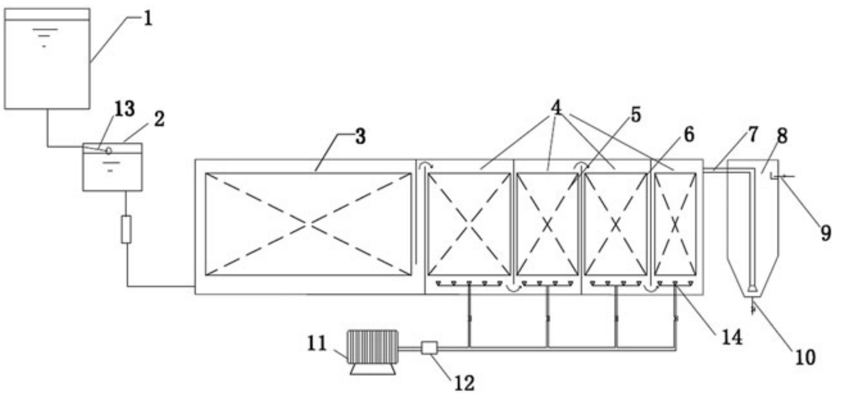

Fig. 1. Set-up of H-FCR system. 1: high-positioned flume, 2: volume-constant flume, 3: hydrolyzation tank, 4: multilevel oxidation tank, 5: fillers, 6: baffle, 7: water pipe for entry to sedimentation tank, 8: sedimentation tank, 9: outlet pipe, 10: discharge pipe, 11: aeration device, 12: rotameter, 13: ride, 14: air blast head.

### 2.2. Experimental apparatus and experimental methods

The experimental apparatus was an H-FCR (Fig. 1; provided by Shanghai Best Environmental Technology Corporation, Shanghai, China), which consists of two parts: hydrolyzation segment and FCR segment. The aerobic section of the FCR segment was divided into four contact oxidation reactors along the treatment process. Their efficient volumes were 0.12, 0.09, 0.09, and 0.06 $m^3$, respectively. Also carriers (called spiral biological carriers) which placed into multilevel contact oxidation tanks, was used to enhance the performance of H-FCR system. The water was 0.97 m deep. The volume ratio of the hydrolyzation segment and the multilevel contact oxidation segment was 0.8:1. Brewery wastewater was treated in the hydrolyzation segment and four-level contact oxidation segment. Then, the wastewater was flowed into a sedimentation tank where sludge and water were separated. The excess sludge was discharged regularly and recycled in the hydrolyzation segment. An aeration device was installed at the bottom of the multilevel contact oxidation segment. The distribution ratio of DO concentration of the four-level contact oxidation reactor was 4:3:2:2, and the DO concentration of contact oxidation tank I, contact oxidation tank II, contact oxidation tank III, and contact oxidation tank IV were 4.0–6.0, 3.0–4.0, 2.0–3.0, and 2.0–3.0 mg/L, respectively.

The whole trial, which lasted for six months, included start-up phase (one month), the optimization of process parameters (nearly two months), stable operation phase (two months), and supplement research (one month). The hydrolyzation segment started up on January, when the room temperature was 14–16℃; and during the stable operation period, the temperature was maintained at a range of 14–27℃. The total HRT of the H-FCR system was 11.7 h (5.2 h for the hydrolyzation segment and 6.5 h for the four-level contact oxidation segment).

### 2.3. Analysis methods

The indicators of source water were measured according to the water and wastewater monitoring and analysis method [23]. Microscopic examinations of different parts were regularly carried out using a microscope (Nikon, YS100, Nikon Corporation Instrument Company, Tokyo, Japan). Activated sludge flocs, filamentous bacteria, protozoa, and metazoan were characterized according to Shen and Zhang [24] using Biolog GN microplates [25] (Biolog Inc., Hayward, CA).

The confidence limits of experimental data ($\mu$) were calculated by formula [1];

$$\mu = \bar{x} \pm t \times \frac{s}{\sqrt{n}} \tag{1}$$

where, $\bar{x}$ is the mean value, $s$ is the standard deviation, and $n$ is the number of replicate experiments. The $t$ value depends on freedom degree and confidence degree, and it was evaluated from a $t$ distribution table. A confidence degree of 95% was used in this study. All the experimental results represent the mean of at least three times.

Excess sludge yield [21] was calculated as follows:

$$\frac{\{\text{MLSS discharged} - (\text{suspended solid influent} - \text{suspended solid effluent})\}}{\{\text{COD influent} - \text{COD effluent}\}}$$

## 3. Experimental results

### 3.1. *Effect of HRT on the removal of COD and ammonia during H-FCR system*

Table 2 shows that the HRT of H-FCR system consists of two parts: hydrolyzation segment and multilevel contact oxidation segment. When the mass concentration of COD in influent water was 1,585 ± 168 mg/L, the HRT of H-FCR system was set to 8.1, 9.9, 11.7, 15.3, and 18.9 h. Then, the COD removal efficiency under various influent concentrations was investigated. The results are shown in Fig. 2.

Fig. 2 shows that COD removal rate decreases sharply with the increase in influent concentration when the HRT are 8.1 and 9.9 h. When HRT is 11.7 h, the COD removal efficiency decreases slightly and remains above 94.5% as influent concentration increases. When HRT increased continuously, COD removal rate increased slightly but not significantly.

Table 2
HRT conditions of H-FCR system during investigation of pollutants removal

| No. | Total HRT of H-FCR system (h) | Hydrolyzation segment (h) | Multilevel contact oxidation segment (h) |
|---|---|---|---|
| 1 | 8.1 | 3.6 | 4.5 |
| 2 | 9.9 | 4.4 | 5.5 |
| 3 | 11.7 | 5.2 | 6.5 |
| 4 | 15.3 | 6.8 | 8.5 |
| 5 | 18.9 | 8.4 | 10.5 |

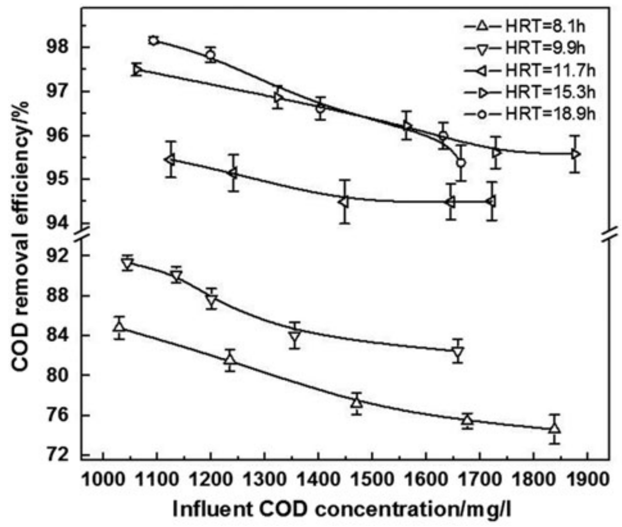

Fig. 2. COD removal efficiency varies with influent concentration under different HRTs for the H-FCR system.

Ammonia-nitrogen removal efficiency also was investigated with various influent concentrations during different HRTs of H-FCR system. Fig. 3 shows that the ammonia-nitrogen removal rate decreases significantly with the increase in influent concentration when the HRT are 8.1 and 9.9 h. Similar to the variation of COD, when the HRT is 11.7 h, the ammonia-nitrogen removal efficiency also reduces slightly and keeps above 87.0% as influent concentration increases. Afterwards, ammonia-nitrogen removal rate had no apparent increase with the increase of HRT.

Considering economic factors and the effect of COD and ammonia–nitrogen removal, the optimal HRT of the H-FCR system was 11.7 h (5.2 h for the hydrolyzation segment and 6.5 h for the four-level contact oxidation segment).

### 3.2. *Overall performance of H-FCR system during the stable operation period*

The removal of contaminates was investigated during the optimum condition of H-FCR system and the results are shown in Fig. 4.

Fig. 4 shows that COD, $BOD_5$, $NH_3$-N, and SS concentration of effluent were below 80, 20, 15, and 70 mg/L, respectively. The average removal efficiencies of COD, $BOD_5$, $NH_3$-N, and SS were 94.9, 97.9, 87.6, and 93.6%, respectively. The water quality of effluent meets discharge standard of pollutants for beer industry (GB 19821-2005).

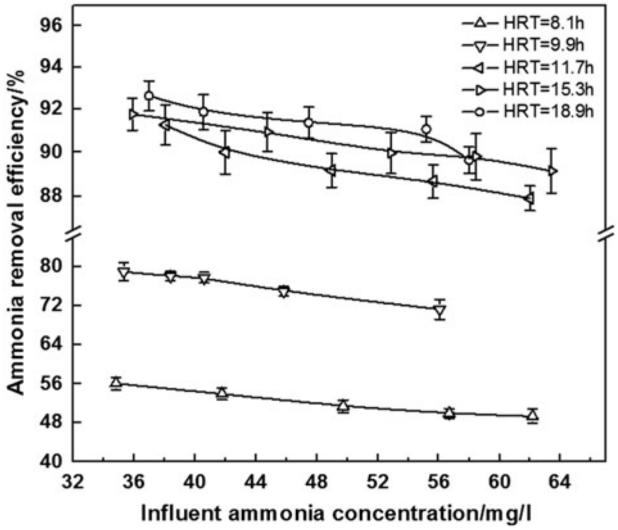

Fig. 3. Ammonia removal efficiency varies with influent concentration under different HRTs for the H-FCR system.

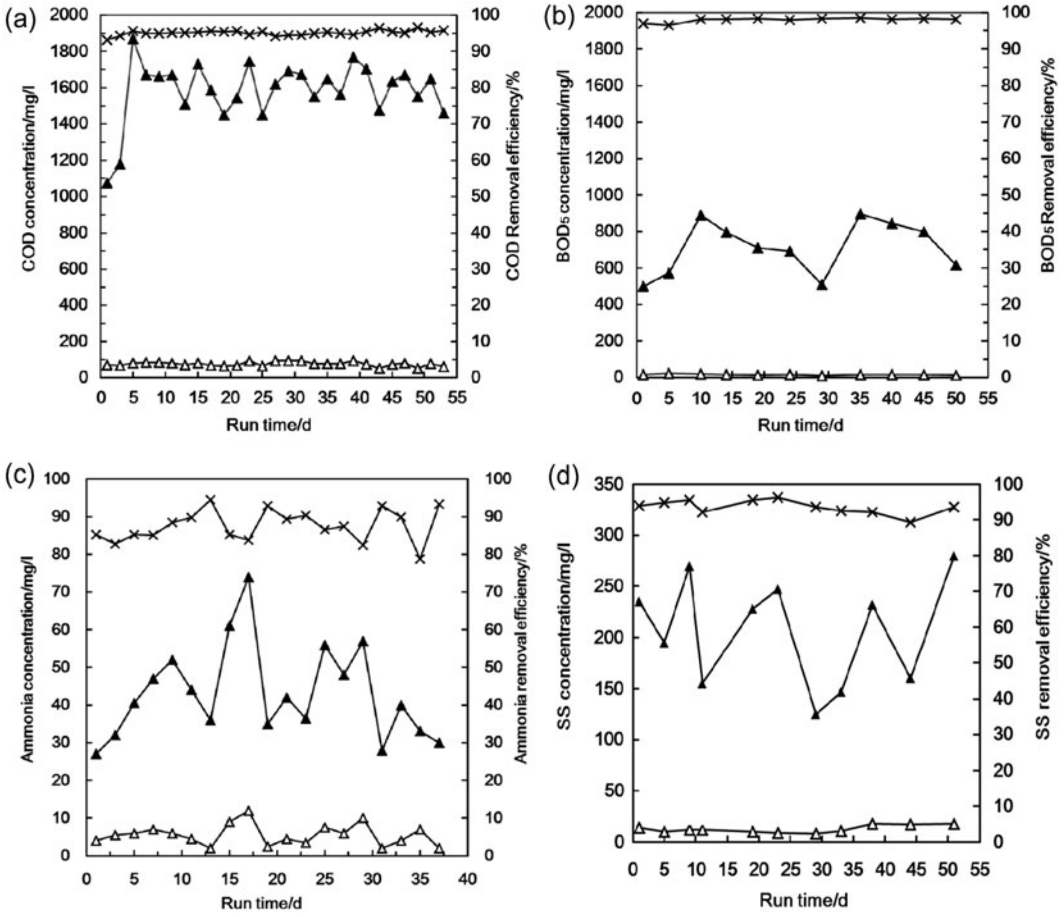

Fig. 4. Contaminant removal efficiency for the entire duration of the experiment (a) COD, (b) $BOD_5$, (c) $NH_3$-N, and (d): SS; ▲ for influent, △ for effluent, and × for removal efficiency.

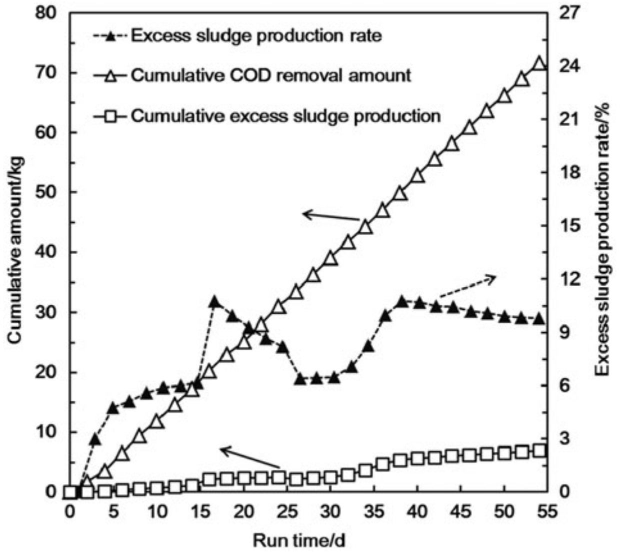

Fig. 5. Calculated and actual excess sludge production of FCR segment during the experimental period.

### 3.3. Reduction of sludge for the FCR segment

The excess sludge production of the FCR segment was continuously and steadily investigated for the entire two month duration of the experiment. The relationship between total sludge production and total COD removal amount was also analyzed. Fig. 5 shows that the ratio of excess sludge production ranges from 0.030–0.108 kg suspended sludge/(kg COD removed). The figure also shows that the average excess sludge production rate is only 8.15% kg SS/(kg COD removed), which is about 15% of that (0.3–1.2 kg TS/(kg BOD removed)) of conventional activated sludge technology [26] and 25% of that (0.01–0.23 kg TSS/(kg COD removed)) of conventional biofilm method [27]. The results indicate that the FCR segment is greatly conducive to the sludge reduction.

## 4. Discussion

As mentioned previously, the H-FCR system played well on the removal of contaminants and greatly affected sludge reduction. The following discussion is focused on the advantage of H-FCR system on the removal of the contaminants and the relationship between biological community distribution and predation during a four-level oxidation process.

### 4.1. The advantage of H-FCR system on the removal of the contaminants

During the overall performance of H-FCR system, the high removal efficiency of contaminates and little excess sludge production were achieved, which indicated that the system was an effective method for the treatment of brewery industrial wastewater. On one hand, the hydrolyzation segment owned stronger resistance of lower wastewater temperature, because the start-up phase was successfully taken at the temperature of 14–16˚C. Also, the hydrolyzation segment could easily degrade the refractory organics and tremendously increased the biodegradability from 0.45–0.68, which reduced the pressure of downstream treatment; on the other hand, four-level contact oxidation segment was good at eliminating the contaminants and reducing the sludge, because of the set-up structure (the volume ratio of four contact oxidation tanks were 4:3:3:2) and the interaction among biological community (e.g. bacteria, protozoa, metazoan, and large metazoan). The set-up structure and the interaction of biological community were detailed in the following section. In a word, the H-FCR system was a promising method during the treatment of brewery wastewater from food industry.

### 4.2. Biological community distribution of FCR segment

The FCR segment consisted of four contact oxidation reactors. During the experimental period, the biomass of each tank was relatively steady and reduced gradually. Each tank had a biological community with different dominant species, as shown in Table 3.

During FCR segment, the whole biological community, which consisted of bacteria–protozoa–metazoan–large metazoan, formed the food chain. This result is similar to that described in previous literature [28]. Table 3 shows that the first and second tanks were allotted for the dispersion culture of bacteria, which had high concentrations of protozoa, such as ciliates, flagellates, and amoeba. A small concentration of protozoa was observed in the third and fourth tanks. For the distribution of metazoan in the four-level contact oxidation tanks, the number of rotifers in the second and third tanks was larger than that in the first tank, and aeolosoma began to increase gradually at the first tank and peaked at the fourth tank.

Table 3 also visually showed the number of different microbes in the four-level contact oxidation reactors. Protozoa could control the number of bacteria by predation [29], so did the metazoan. Nonetheless, the succession law of metazoan was different from that of protozoa. Compared with the number of protozoa, metazoan which belonged to the high trophic level in the biological community was an order of magnitude lower. Furthermore, a low microbial number was noticed when the trophic level was high.

In the previous study [30], the number of high trophic level was about 10%, and the amount of energy lost was almost 90% in the transmission of the food chain rule of terrestrial and freshwater ecosystems. This energy loss was caused by the inefficient biomass conversion during energy transfers from a low trophic level (bacteria) to a high level (protozoa and metazoan). The advantage of the H-FCR system lies on the low energy it requires to synthesize an organism, which is a process that may result in the decrease of the high trophic level [31]. Thus, the space for a high trophic level is relatively small. In the study, a reactor was designed to comprise four contact oxidation tanks with decreasing volume following biological energy conversion efficiency [32]. The volume ratio of 4:3:3:2 was similar to that of a pyramid. This pyramid mode provides a suitable environment for different micro-organisms and weakens interspecific competition by keeping the biofacies separated [27]. The micro-organisms at different trophic levels remain in great numbers, thereby contributing to the formation of the food chain of bacteria–protozoa–metazoan–large metazoan.

### 4.3. Relationship between biological community distribution and COD removal in the FCR segment

A significant correlation between the COD removal rate and the ratio of different microbes was observed during the experimental period. The first tank contributed the highest COD removal with an average percentage of 46.5%. The second, third, and fourth tanks contributed COD removal percentages of 24, 16, and 5.5%, respectively (Fig. 6). The wastewater was becoming clear (i.e. COD descending) with the decrease of nutrients, but with an increase in the ratio of metazoan density to protozoa density and metazoan density to bacteria density.

Fig. 6(a) shows that the average COD removal efficiency of the first tank (46.5% of FCR segment) makes up 31.2% of the total COD removal (94.9%). The volume of the first tank was the largest; therefore, the

Table 3
Biological community distribution of FCR segment during the experimental period

| Microbes | Amount (cell/ml) | | | |
|---|---|---|---|---|
| | First tank | Second tank | Third tank | Fourth tank |
| *Vorticella microstoma* | 2,317 ± 76 | 1,507 ± 90 | 733 ± 76 | 160 ± 20 |
| *Epistylis plicatilis* | 783 ± 76 | 1,200 ± 100 | 610 ± 85 | – |
| *Carchesium polypinum* | 6,000 ± 100 | 3,620 ± 72 | – | – |
| *Opercularia* | 2,293 ± 190 | 3,800 ± 150 | 420 ± 30 | – |
| *Aspidisca aculeata* | 1,697 ± 55 | 800 ± 20 | 203 ± 15 | 217 ± 25 |
| *Polypinum* | 4,167 ± 153 | 2,700 ± 100 | – | – |
| *Litonotus* | 395 ± 18 | 101 ± 11 | – | – |
| *Bodoedax* | 790 ± 10 | 81 ± 9 | – | – |
| *Suctoria* | 120 ± 5 | 96 ± 4 | – | – |
| *Paramecium* | 600 ± 20 | 323 ± 15 | – | – |
| *Rotifer* | 140 ± 5 | 483 ± 15 | 350 ± 20 | 180 ± 10 |
| *Nematode* | 810 ± 36 | 653 ± 45 | 420 ± 30 | 223 ± 25 |
| *Pristinaequiseta* | 607 ± 40 | 423 ± 25 | 163 ± 15 | 31 ± 4 |
| *Aeolosoma* | 30 ± 5 | 12 ± 3 | 70 ± 5 | 90 ± 10 |

Note: The amount of microbes was calculated by formula (1) which was set in Section 2.3 of manuscript.

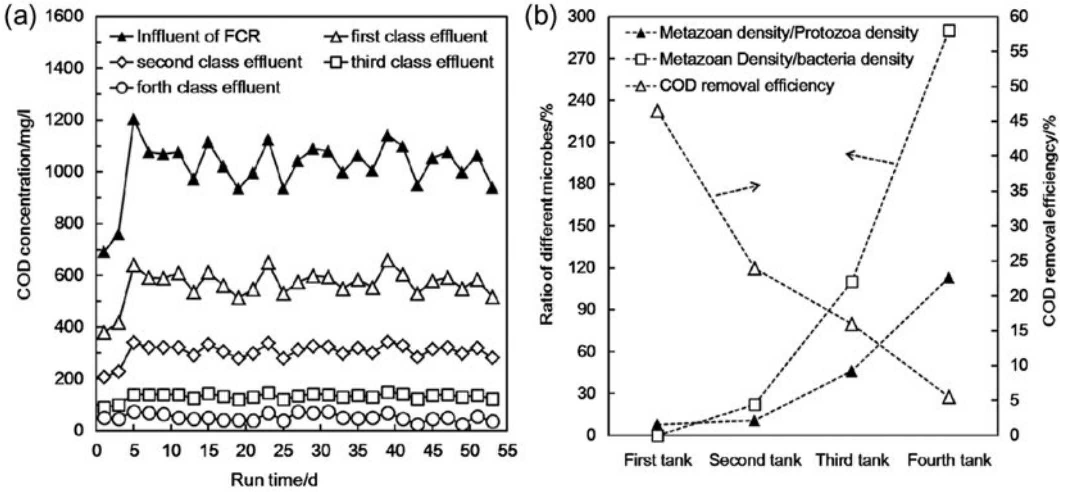

Fig. 6. COD removal efficiency and the ratio of different microbes in the FCR segment.

biomass made up of mixed liquid suspended solids was the highest (7,000–8,000 mg/L). Fig. 6(b) shows that the majority of ammonification and nitrification bacteria were found to concentrate inside the first tank, and the substrate utilization rate of bacteria was higher than that of protozoa and metazoan. Additionally, the bacteria could directly degrade organic compounds. A spot of protozoa and metazoan in the first tank had benefitted from the interaction among bacteria, protozoa, and metazoan. On one hand, the formation of bacterium zoogloea could resist the predation of protozoa and metazoan, whereas the bacterial secretions stimulated the growth of protozoan; on the other hand, the activity of protozoan and metazoan created dissolved organic matter in promoting the growth of bacteria [33]. Therefore, the existence of this interaction strengthened the degradation of pollutants by microbe in the first tank. The first tank not only improved removal efficiency of COD but also decreased the burden of later tanks.

The number of protozoa increased gradually in the four reactors. Among them, the second tank further degraded organic matters that depended mostly on predation of bacteria, protozoa, and floc oxidation. In the third and fourth reactor, the concentration of organic substrate (e.g. bacterial substrate) was so low that it became unsuitable to the protozoa within a short generation time. Therefore, the metazoan with a long generation time became the dominant species in the third tank, especially in the fourth tank.

### *4.4. The predation relation within a biological community in the FCR segment*

After 30 d, suspended sludge and biofilm were carefully collected from the reactors. The predation within the biological community could be observed clearly by microscope (1,000 × magnification).

The predation ability of metazoan was strong enough because of its trophic level among the bacteriaprotozoa–metazoan community. Actually, the bacterium could self-flocculate by secreting polysaccharide to resist prey-predation. Therefore, the zoogloea could provide well protection neither be fed nor be broken by protozoa. However, it was useless and was easily ingested by metazoan which stayed at the top of food chain. For instance, Fig. 7(a) showed that *rotifer* swallowed floc (zoogloea). As revealed in the different microphotographs, the ciliary movement of *rotifers* (metazoan) was stronger than that of *vorticella* (protozoa). Given this feature, the protozoa play a key role in the prey-competition system. *Rotifer* activity could promote biofilm updates, reduce non-active substances, and loosen biofilm. This activity could also facilitate the transfer of nutrients and oxygen to prevent internal corruption and increase biofilm activity to enhance purification efficiency. Moreover, *rotifers* can prey on free bacteria and tiny particles, remains of dead cells, and humus colloid.

Fig. 7(b) shows that *nematodes* (metazoan) prey on *filamentous bacteria* (fungi). When the concentration of bacteria is $5 \times 10^9$–$10 \times 10^9$, the female *nematodes* can ingest 5,000 bacteria every minute at 20˚C [28,34]. The *nematodes* eat $1.94 \times 10^{-6}$ g in dry weight daily. This value is equivalent to 650% of the self-weight of the *nematodes*; however, only 12% are assimilated. *Nematodes* usually begin to grow with a bacterial concentration of 100 cells per milliliter. Its growth rate is proportional to the bacterial concentrations until a plateau of 1,000 cells

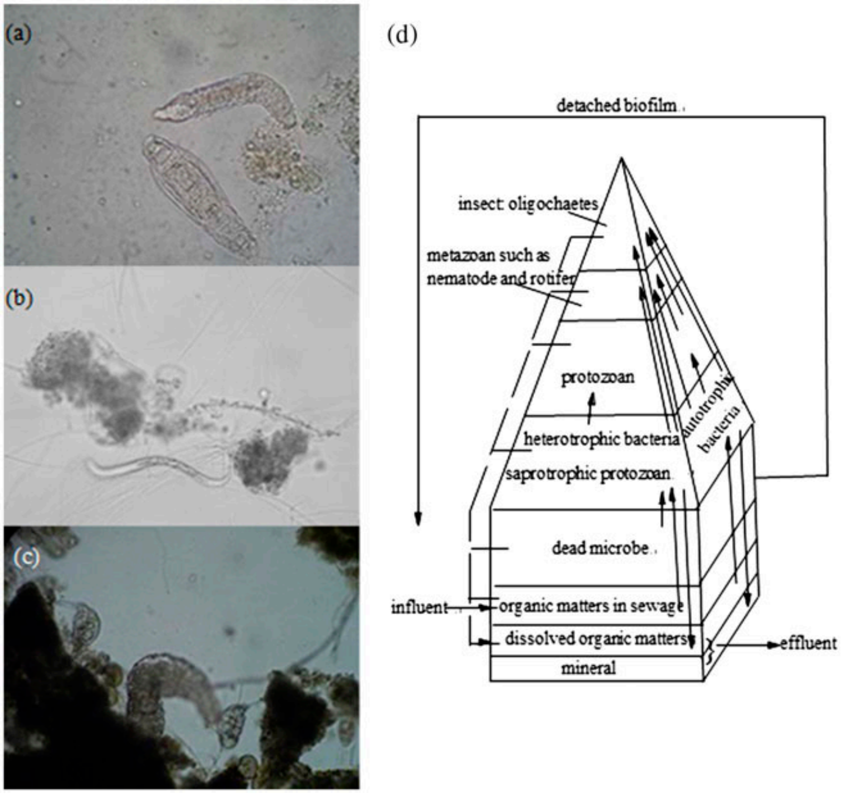

Fig. 7. Biological predatory behavior. (a) *Rotifer* sp. preying on sludge flocs; (b) *Nematoda* preying on filamentous bacteria (predation between metazoan and fungi); (c) *A. hemprichii* preying on *Trochilia minuta* (predation between metazoan and protozoan); and (d) Composition of the food chain of the FCR segment.

per milliliter is reached. Therefore, bacteria are an ideal food supply for nematodes.

An *Aeolosma hemprichii* (metazoan) is a preying *rotifer* (protozoa), as shown in Fig. 7(c). The predatory quantity of a large metazoan (e.g. *A. hemprichii*) is higher than that of a protozoan (e.g. ciliates), although the number of *A. hemprichii* is far less than that of *rotifers* and *nematodes*. Because the *A. hemprichii* is omnivorous it eats organic detritus in water, undergoes both sexual and asexual reproduction (the former is dominant), and requires an appropriate temperature of 20˚C. In addition, *A. hemprichii* is at the top of the food chain in biofilm, and it can reduce sludge production by predation [19,35]. In the process of recycling *A. hemprichii*, the system realizes the residual sludge resource up to a certain extent.

Fig. 7(d) shows that the trophic level increases gradually along the flow direction; a food chain of bacteria–protozoa–metazoan is then formed, and this result is similar to that described in previous work [22]. Given the rich organic matter by brewery wastewater, bacteria increases and reproduces massively, subsequently causing an increase in excess sludge production. Protozoa are the primary predators in the wastewater treatment system because they consume more than half of the bacteria, particularly free bacteria [36], inside the aeration tank. The predation of protozoa can significantly reduce the bacterial concentration of effluent, increase the quality of effluent, and decrease sludge production. Compared with protozoa, metazoan [28] are much larger and better at predation, even in small numbers. Metazoan is at the top of the food chain. They can reduce sludge production by preying upon protozoa, bacteria, and suspended solids.

## 5. Conclusion

(1) The H-FCR system demonstrated enhanced effects under the following conditions: temperature: 22.2 ± 4.5˚C, HRT: 5.2 h for the hydrolyzation segment and 6.5 h for the four-level contact oxidation segment, and pH: 7.5 ± 0.5. The mass concentrations of COD, $BOD_5$, $NH_3$-N, and SS in influent water were 1,585 ± 168 mg/L, 711 ± 146 mg/L, 43.1 ± 12.5 mg/L, and 206.7 ± 53 mg/L, respectively. The concentrations of effluent were below 80, 20, 15, and 70 mg/L, respectively. The water quality of the effluent meets discharge standard of pollutants for beer industry (GB 19821-2005).

(2) The four-level contact oxidation tank of the FCR segment (volume ratio of 4:3:3:2) was utilized to provide a suitable environment for the formation of the food chain of bacteria–protozoa–metazoan–large metazoan.

(3) The ratio of metazoan and protozoa increased gradually with COD concentration reduction along the flow direction. The result proves the role of predation in reducing sludge production from the source. In the study, sludge production rate decreased to 8.15% kg SS/(kg COD removed).

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