AQUA
Water Infrastructure, Ecosystems and Society

IWA PUBLISHING

 AQUA — Water Infrastructure, Ecosystems and Society Vol 71 No 8, 926 doi: 10.2166/aqua.2022.152

Enhanced removal of dissolved S^{2-} from a river by the pump-and-treat *ex-situ* process with enriched consortium

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ABSTRACT

S^{2-} is one of the common pollutants in heavily polluted rivers. A pump-and-treat *ex-situ* process with enriched consortium (PEPEC) was used to remove S^{2-} in this study. The kinetic model of S^{2-} removal was developed, and the inflow ratio of the PEPEC was analyzed according to the results of the kinetic models. The results showed that the S^{2-} removal ratio could reach 97.5% ± 0.5%, when the inflow ratio was controlled at 2% for the PEPEC operation. Meanwhile, the removal efficiency and operation performance were assessed for both the simulating *ex-situ* and *in-situ* bench-scale tests. Compared with the *in-situ* processes, the PEPEC showed a stable operation performance during 120 h of bio-treatment, and the concentrations of S^{2-}, COD, NH_3-N and TP in the effluent reached approximately 0.5, 20, 0.5 and 0.5 mg/L, respectively. The time consumption (8 h for one batch) and consortium dosage (3 g for the whole operation) in the PEPEC were significantly less than those in the *in-situ* processes. The PEPEC presented some potential advantages for the bio-treatment of a heavily polluted river.

Key words: enriched consortium, dissolved S^{2-}, inflow ratio, initial biomass concentration, pump-and-treat *ex-situ* process

HIGHLIGHTS

- This article developed a pump-and-treat ex-situ process with enriched consortium (PEPEC) to solve black-odor water problem.
- This article built a kinetic model and found the optimal in flow ratio of PEPEC.
- This article applied bench-scale tests to compare PEPEC and *in-situ* methods and found the advantage of PEPEC.

INTRODUCTION

Due to the development of rapid urbanization, many rivers have been heavily polluted (Wang *et al.* 2021). Research has shown that S^{2-} is one of the common pollutants (Song *et al.* 2017). S^{2-} can form other sulfur-containing pollutants such as hydrogen sulfide and metal sulfides. Hydrogen sulfide is a biologically important molecule with complex physiological functions, which can cause the well-known inhibitory effect on cellular respiration and potential inhalation hazard. Some insoluble metal sulfides like FeS could be suspended in a heavily polluted river, which induces water to be black (Wang *et al.* 2014). Many *in-situ* techniques, such as coagulation aeration and biological methods, have been utilized to remove S^{2-} from the water body (Yin *et al.* 2019). However, these techniques have some disadvantages such as being time-consuming, the high cost, the large space occupation or high microbe dosage (Cao *et al.* 2020).

In the past decades, *ex-situ* microbial techniques have rarely been used to treat a heavily polluted river considering the relatively fast stream flow and high flow rate of a river. In fact, *in-situ* remediation for river often means significant projects and a large amount of investment. However, there is partial retention or even stagnant water in some small sections of a heavily polluted river, which provides the application possibility of the *ex-situ* process. A pump-and-treat approach with inoculated microbes in a mini-bioreactor is regarded as a promising and developing *ex-situ* process. This has been successfully used to remediate groundwater and soil (Boal *et al.* 2015). Biomass concentration is an important factor that affects the treatment efficiency of a bioreactor. Enrichment consortium with a high biomass concentration can shorten the operation time and improve the treatment capacity of a bioreactor, and this technique has been used in the fermentation process

(Liu & Zhu 2018). Also, enriched consortia has positive effects on substrate degradation, and higher methane yields were observed in the reactors compared with control reactors set up with standard inoculum (Ozbayram *et al.* 2017, 2018). An enhanced stability of continuous biomethanation processes has been achieved because enriched consortia could alleviate ammonia toxicity in a continuous stirred tank reactor operating under ammonia-induced, inhibited-steady-state (Fotidis *et al.* 2017).

Therefore, a pump-and-treat *ex-situ* process with enriched consortium (PEPEC), namely a simulating *ex-situ* process, was developed to remove dissolved S^{2-} from a heavily polluted river. Effects of initial biomass and S^{2-} concentration on the removal efficiency of dissolved S^{2-} were firstly investigated, and S^{2-} removal kinetics were then analyzed. Based on the obtained results of kinetic models, the inflow ratio parameter of the PEPEC was investigated. Finally, a comparison of the bio-treatment performance between the PEPEC and the simulating *in-situ* process was carried out at the lab-scale.

MATERIALS AND METHODS

Water sample

The original water samples were individually collected at eight sampling sites from Dongsha River that was a heavily polluted urban river located in Beijing, China. The representative water sample was created by the mixture of the eight samples in order to obtain the average values to eliminate the difference among the samples. The characteristics of the water sample were analyzed, and the concentrations of S^{2-}, COD, NH_3-N and TP were 20.7 ± 1.2 mg/L, 104.5 ± 8.0 mg/L, 5.3 ± 0.4 mg/L and 3.0 ± 0.1 mg/L, respectively.

Enriched consortium

Three single strains including *Citrobacter* sp., *Ochrobactrum* sp. and *Stenotrophomonas* sp. were all isolated from Dongsha River and were characterized by 16S rDNA sequence. Their sequences were all uploaded to the National Center for Biotechnology Information, and the GenBank accession numbers were MH181794, MH181795 and MH181796, respectively. Microbial consortium was composed of these three strains, and their mass proportions were set at 1:1:1. The initial cell density of each strain was 8.96×10^5 cells/mL (Colony-Forming Units, cfu). In our previous study, microbial consortium has been proved to be capable of efficient S^{2-}-oxidizing, and more than 60% of dissolved S^{2-} in some urban black-odor water bodies located in Beijing of China were removed (Xu *et al.* 2019). It was found that S_0, $S_2O_3^{2-}$, SO_3^{2-} and SO_4^{2-} were all formed in the whole process of S^{2-} oxidization (Zhang *et al.* 2022). Enriched consortium was obtained using a liquid medium that consisted of 20 g/L of glucose, 10 g/L of peptone and 10 g/L of yeast extract. The pH of liquid medium was adjusted to 7.0. The incubation was carried out at 25 °C and 120 rpm for 48 h. The enriched consortium harvested with centrifugation was used immediately for the following experiment.

Batch experiment

Effects of initial biomass and S^{2-} concentration on the bio-treatment efficiency of dissolved S^{2-} were investigated using a bioreactor with 3 L of working volume. According to the reported results of microbial tolerance for S^{2-} (Zhuang *et al.* 2017), three S^{2-} levels (30, 50 and 100 mg/L) were chosen as the initial concentrations in batch experiment, respectively. $Na_2S{\cdot}9H_2O$ was used to increase the initial S^{2-} concentration of each sample. The samples were inoculated at different biomass concentrations, and then were treated at 25 °C in the batch bioreactors. In addition, a group without enriched consortium served as the control. The control was used to obtain the background variation, which was subtracted from that of any other batch experiment.

Continuous experiment

The effect of inflow ratio on the removal efficiency of dissolved S^{2-} was evaluated in a continuous bioreactor (as shown in Figure 1(a)). The water sample from Dongsha River was directly used as the influent of the continuous experiment. Each bioreactor was inoculated by transferring 1.0 g/L biomass concentration of enriched consortium, and then was operated at 25 °C. After the rapid start-up, each bioreactor was continuously run at various inflow ratios. Each experiment was carried out in triplicate.

Simulating the *ex-situ* process and the *in-situ* process

The simulating *ex-situ* process, namely the PEPEC, was run using the same bioreactor as the continuous experiment. The constructed bioreactor was a cube with an effective volume of 3 L. The influent was pumped from the bottom of the

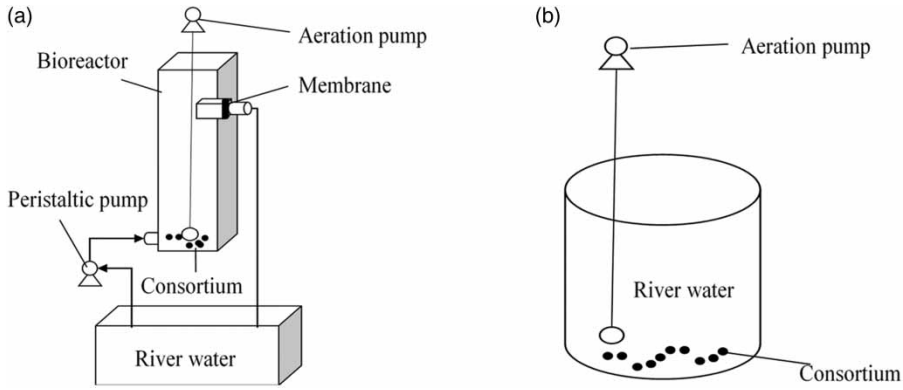

Figure 1 | The schematic diagram of the PEPEC (a) and *in-situ* process (b).

bioreactor into the bioreactor with a flow rate of 0.06 L/min by a peristaltic pump. The effluent was overflowed from the top of the bioreactor. A 2 μm filter membrane was attached to the outlet to retain microorganisms in the bioreactor. For the PEPEC, the inoculum containing 1.0 g/L of biomass concentration was transferred into each bioreactor, and the inflow ratio was set at 2%. The simulating *in-situ* experiment was carried out using a bench-scale cylindrical bioreactor (as shown in Figure 1(b)). The working volume of cylindrical bioreactor was 30 L. In the simulating *in-situ* process, two levels of biomass concentrations (0.01 and 1.0 g/L) were chosen to compare the removal efficiency of S^{2-} by enriched consortium. The water sample from Dongsha River was both used as the influent in the *ex-situ* process and the *in-situ* process. All experiments were conducted at 25 °C for 120 h. Each experiment was performed in triplicate.

Analytical methods

The concentrations of S^{2-}, COD, NH_3-N and TP were analyzed according to Chinese Water and Wastewater Monitoring and Analysis Methods (MEP 2002b). Both COD and NH_3-N were determined using a HACH multi-parameter detector (DR2800, USA); while TP and S^{2-} were measured using a UV–Vis spectrophotometer (UV-1800, Japan). The biomass concentration was determined according to the previously described method (Wang *et al.* 2014). 30 mL of mixed suspension was sampled during the reaction, and then was centrifuged at 8,000 rpm for 10 min. The supernatant was discarded. The obtained sediment was washed five times with distilled water. After centrifugation, washed sediment was weighted. The biomass concentration was calculated as the wet weight of washed sediment divided by the volume of mixed suspension. At the end of the reaction process, the three single strains were all recovered and sterilized before disposal in this study.

RESULTS AND DISCUSSION

Removal efficiency of dissolved S^{2-} as a result of initial biomass concentration

Biomass concentration is one of the most important factors that can greatly influence pollutant removal efficiency. Figure 2 shows the effect of initial biomass concentration on the S^{2-} removal efficiency at different initial S^{2-} concentrations. In each case, the S^{2-} concentration decreased rapidly, and then leveled off. The final S^{2-} concentrations for all groups were similar and the values were all below 0.5 mg/L. However, the time necessary for dissolved S^{2-} removal was obviously different. A positive impact of initial biomass concentration on the spending time was observed in all. When increasing the biomass concentration from 0.01 to 10 g/L, the time spent for S^{2-} removal decreased from 100 to 23 h, and it was significantly shortened by 77% (Figure 2(a)). In contrast to initial biomass concentration, initial S^{2-} concentration had a negative influence on the spending time for S^{2-} removal, as shown in Figure 2. At 30 mg/L of initial S^{2-} concentration, more than 80% of dissolved S^{2-} was removed in the first 16 h of bio-treatment, whereas for 50 and 100 mg/L, the required time was prolonged to 23 and 30 h, respectively.

Figure 3 shows the variation of average removal rate of dissolved S^{2-} with biomass concentrations at different initial S^{2-} concentrations. The average removal rate of S^{2-} dramatically increased when biomass concentration was raised to 10 g/L, and then remained relatively stable at the higher biomass concentration. 1.2 mg/(L h) of high removal rate was achieved at 10 g/L of biomass concentration.

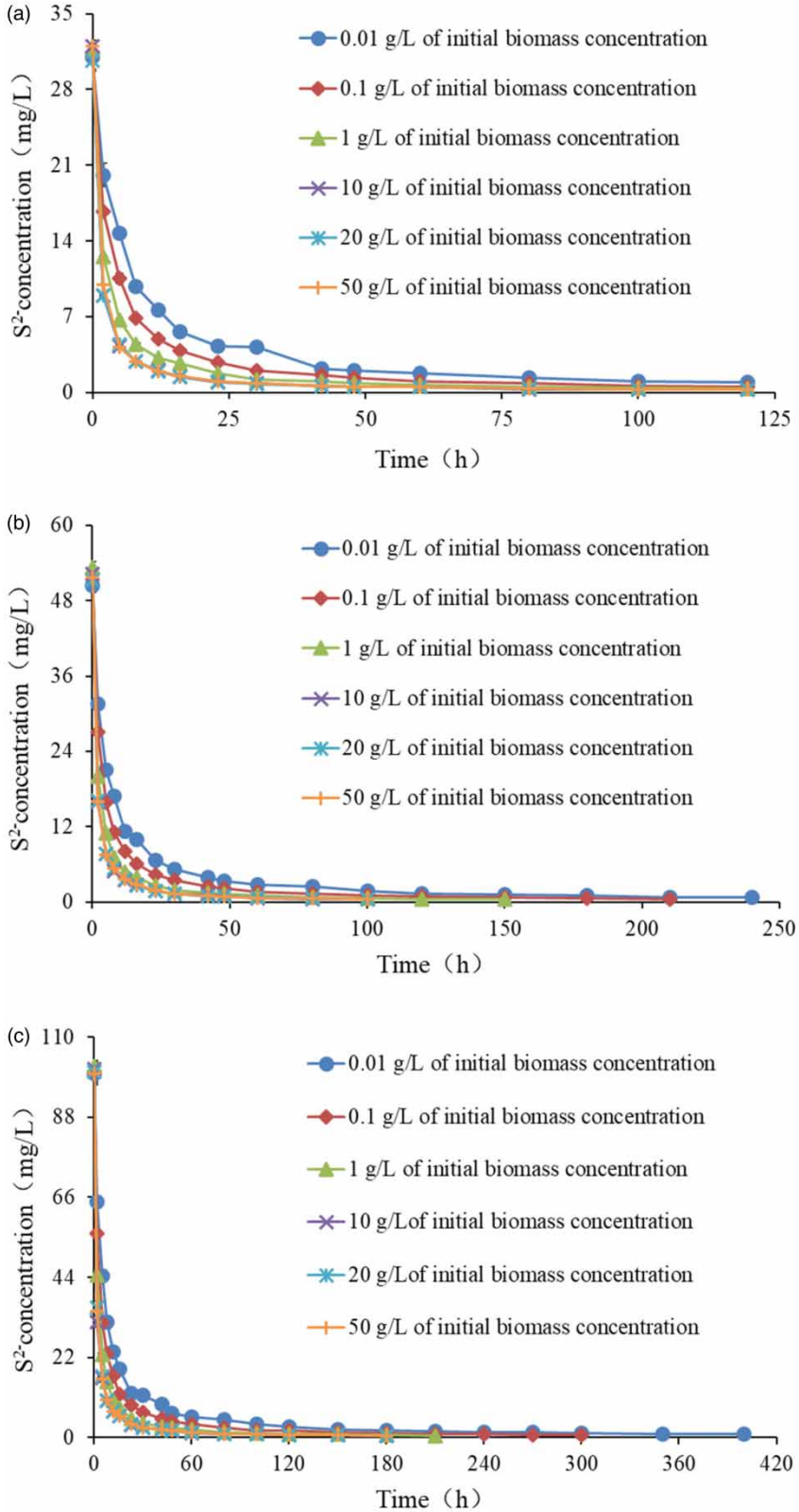

Figure 2 | Effects of the initial biomass concentration on the S^{2-} removal efficiency at different initial S^{2-} concentrations: (a) 30 g/L of initial S^{2-} concentration; (b) 50 g/L of initial S^{2-} concentration; (c) 100 g/L of initial S^{2-} concentration (Xu *et al.* 2019).

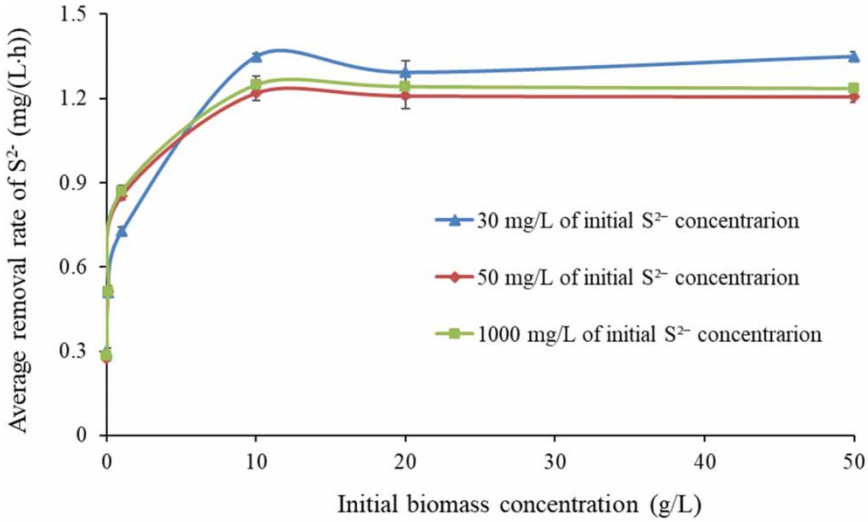

Figure 3 | Variation of the average removal rate of dissolved S^{2-} at various initial S^{2-} concentrations.

The appropriate amount of inoculation helped the microorganisms to adapt the new environment and might sustain high metabolic activity, which accelerated the removal of dissolved S^{2-} in the water sample. A further increase of biomass concentration did not contribute to shortening the time due to the competition of S^{2-} among microorganisms, which indicated a 'saturation' state of biomass concentration (Pradhan & Rai 2000). This might explain why the removal rate of S^{2-} did not increase further when biomass concentration was increased from 10 to 50 g/L. In addition, it was found that the S^{2-} removal rates were close at various initial S^{2-} concentrations, suggesting that the initial S^{2-} concentration had little influence on the average removal rate in this study.

This finding suggests that other common dissolved pollutants including COD, NH_3-N and TP could also be removed by the inoculated microorganisms. Table 1 shows the effect of initial biomass concentration on the removal ratios of COD, NH_3-N and TP after 23 h of bio-treatment under the condition of 30 mg/L initial S^{2-} concentration. The removal ratio of COD decreased by 48.9% in the case that biomass concentration increased from 1 to 10 g/L. Compared with other conditions, 50 g/L of initial biomass concentration could not support efficient removal of NH_3-N and TP, and the removal ratios were only 32.1 and 50.5%, respectively.

The results shown in Table 1 suggested the optimal range of initial biomass concentration was 0.01–1 g/L. However, the time required for S^{2-} removal was dramatically shortened with the increase of initial biomass concentration from 0.01 to 10 g/L (Figure 2). Considering both the removal ratio and the required time, the optimal initial biomass concentration was 1 g/L. Under this condition, the removal ratios of S^{2-}, COD, NH_3-N and TP were obviously enhanced, and they reached 98.5, 81.5, 94.8 and 85.1%, respectively, after 23 h of bio-treatment with the enriched consortium.

Table 1 | Effect of initial biomass concentration on the removal ratios of S^{2-}, COD, NH_3-N and TP at 30 mg/L of initial S^{2-} concentration

	Removal ratio after 23 h of bio-treatment (%)		
Initial biomass concentration (g/L)	**COD**	**NH_3-N**	**TP**
0.01	79.5 ± 1.3	93.9 ± 0.4	80.3 ± 0.4
0.1	80.9 ± 1.0	93.9 ± 0.7	85.2 ± 0.7
1	81.5 ± 2.3	94.8 ± 0.5	85.1 ± 0.2
10	41.6 ± 1.2	93.9 ± 0.6	84.0 ± 0.4
20	29.5 ± 1.7	94.0 ± 0.4	85.6 ± 0.1
50	24.4 ± 1.9	32.1 ± 2.3	50.5 ± 1.0

Removal kinetics of dissolved S^{2-}

The S^{2-} removal kinetic model was studied based on the Eckenfelder model (Adams *et al.* 1975). An ideal condition that the substrate was fully mixed at any time and would be removed completely was assumed in this study. The S^{2-} removal could be described as in Equation (1):

$$t = \frac{S_i - S_e}{K_m X S_e} \tag{1}$$

where t (h) represents the removal time of dissolved S^{2-}, S_i (mg/L) is S^{2-} concentration in the influent, S_e (mg/L) is S^{2-} concentration in the effluent, X (g/L) is initial biomass concentration and K_m (L/(g·h)) refers to specific substrate removal rate coefficient. The kinetics model of S^{2-} removal could also be expressed by the following first-order kinetic Equation (2):

$$\frac{S_i - S_e}{Xt} = K_m S_e \tag{2}$$

According to Equation (2), K_m depended on X, indicating that the S^{2-} removal kinetic model is special at various X values. On the basis of the obtained experimental data, the special kinetic model and each corresponding K_m were calculated, and the results were summarized in Table 2.

It was found that the experimental values at each initial biomass concentration correlated well with the theoretic values, which indicated that this first-order kinetic model could describe the removal of dissolved S^{2-} with the enriched consortium. K_m in a bio-treatment system is mainly affected by biomass concentration, substrate concentration and inhibition tendency (Mathur *et al.* 2008). The K_m values decreased with an increase of biomass concentration, as shown in Table 2. The maximum of K_m was 28.261 L/(g·h); this value was obtained at 0.01 g/L of the lowest biomass concentration. When biomass concentration increased to 10 g/L, the value of K_m significantly decreased to 0.1135 L/(g·h); while with a further increase of biomass concentration to 50 g/L, K_m only decreased to 0.0255 (L/(g·h)). K_m can indicate the affinity of microorganisms or enzymes for substrates (Mathur *et al.* 2008). Consequently, the higher the K_m value, the lower the affinity is; meanwhile, the substrate is consumed more and more slowly with the decrease of affinity (Shen *et al.* 2014). Therefore, there was a negative relationship between K_m and the average removal rate of dissolved S^{2-}. A low biomass concentration disfavored the rapid adaption of enriched microorganisms to the new environment, and the affinity for substrate was weak, resulting in the low consumption rate of S^{2-}. When the initial biomass concentration gradually increased to 10 g/L, the activity of inoculated microorganisms and the affinity were both enhanced significantly. The dissolved S^{2-} as the substrate became the key limiting factor for the growth of microorganisms, resulting in the intensified competition for S^{2-} among inoculated microorganisms. Therefore,

Table 2 | Summaries of the special kinetic model and the corresponding each K_m at various biomass concentrations (Xu *et al.* 2019)

X (g/L)	K_m (L/(g·h))	R^2	Kinetic models	Eq.
0.01	28.261	0.9955	$t = \frac{S_i - S_e}{28.261 X S_e}$	(3)
0.1	4.4567	0.9978	$t = \frac{S_i - S_e}{4.4567 X S_e}$	(4)
1	0.7868	0.9990	$t = \frac{S_i - S_e}{0.7868 X S_e}$	(5)
10	0.1135	0.9971	$t = \frac{S_i - S_e}{0.1135 X S_e}$	(6)
20	0.0667	0.9959	$t = \frac{S_i - S_e}{0.0667 X S_e}$	(7)
50	0.0255	0.9985	$t = \frac{S_i - S_e}{0.0255 X S_e}$	(8)
$0 < X \leq 10$	0.7345	0.9996	$t = \frac{S_i - S_e}{0.7345 X^{0.206} S_e}$	(9)
$10 \leq X \leq 50$	1.017	0.9928	$t = \frac{S_i - S_e}{1.017 X^{0.066} S_e}$	(10)

the average removal rate of dissolved S^{2-} obviously increased due to the large consumption of S^{2-} during a short period. With a further increase of biomass concentration, although K_m decreased, the provided S^{2-} was not enough for maintaining the growth and metabolism of inoculated consortium, and substrate consumption was shifted to endogenous respiration. This might explain the reason why the average removal rate of S^{2-} did not increase any more when the initial biomass concentration was further raised up to 50 g/L (Figure 3).

Operation stability of the PEPEC as a result of the inflow ratio

Inflow ratio is an important process parameter, which may significantly influence effluent quality and treatment capacity of a process (Pradhan & Rai 2000). In order to combine with Equation (1), the inflow ratio was calculated from the effective volume of the reactor divided by the influent volume per min in this study:

$$i = \frac{Q}{V} \tag{11}$$

where Q (L) is the influent volume per min, V (L) is the effective volume of the reactor and i (%) refers to the inflow ratio. Therefore, Equation (1) could be replaced by the following equation:

$$t = \frac{iS_i + (1-i)S_e - S_e}{K_m X S_e} = \frac{i(S_i - S_e)}{K_m X S_e \times 60} \tag{12}$$

A low inflow ratio can avoid the great fluctuation of water quality due to the effective treatment of pollutant at a long hydraulic retention time (HRT) (Ren *et al.* 2016). However, a lower value does not necessarily indicate a better result. According to the previous reported results (Yu *et al.* 2014), an appropriate inflow ratio should meet two requirements: (1) the time spent on treating the pollutant from the original concentration to the target concentration should not exceed the HRT and (2) the inflow ratio should be high enough in order to achieve the maximal treatment capacity.

In addition, HRT was defined as the quotient of the effective volume of the reactor and the influent volume per hour. As such, the relationship between inflow ratio and HRT could be shown as the following equation:

$$\mathrm{HRT} = \frac{1}{i \times 60} \tag{13}$$

Therefore, the inflow ratio could be written as follows:

$$t = \frac{i(S_i - S_e)}{K_m X S_e} \leq \frac{1}{i \times 60} \tag{14}$$

According to the above results in this study, 1 g/L of initial biomass concentration was optimal compared with other conditions. Thus, the K_m and X was considered the best option in Equation (14), and this is shown in Equation (15):

$$t = \frac{i(S_i - S_e)}{0.7868 \times 1 \times S_e} \leq \frac{1}{i \times 60} \tag{15}$$

The theoretic inflow ratio was derived from Equation (15), and its maximal value was 2.5%. The experimental value of inflow ratio was then confirmed within the range of 1.0–3.0%. In order to evaluate the effect of inflow ratio on the operation stability of the PEPEC, the simulating experiment was carried out to investigate the variation of S^{2-} removal efficiency at various inflow ratios, as shown in Figure 4.

Figure 4(a) shows that the concentrations of dissolved S^{2-} in the effluent were different at various inflow ratios during the operation process. When the inflow ratio was below 2.0%, the effluent S^{2-} concentration ranged from 0.3 to 0.4 mg/L. At 2.0% of the inflow ratio, S^{2-} concentration was slightly raised to approximately 0.5 mg/L. However, when the inflow ratio was over 2.0%, the effluent S^{2-} concentrations dramatically increased. This increase was particularly obvious at 3.0% of the inflow ratio, where it exceeded 1.0 mg/L and showed an upward trend. It was concluded that the effluent S^{2-} concentration increased with the increase of the inflow ratio. This was because the required time for S^{2-} removal at 1.0–2.0% of

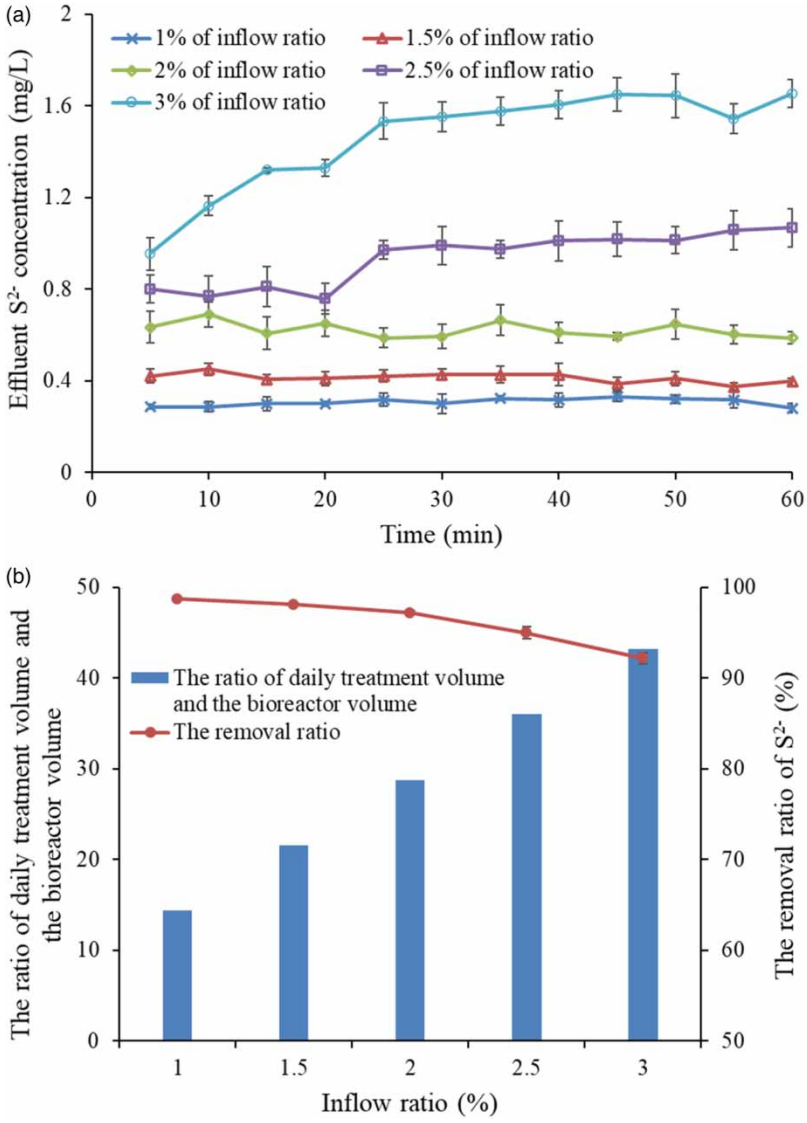

Figure 4 | Effects of inflow ratio on the operation performance of the PEPEC. (a) Variation of S^{2-} concentration in the effluent. (b) Variation of S^{2-} removal ratio and the ratio of daily treatment volume and the bioreactor volume.

inflow ratio was sufficient so that the S^{2-} could be removed at a low level. At 3.0%, the spending time (0.72 h) surpassed its corresponding HRT (0.56 h), resulting in the accumulation of untreated S^{2-} in the bioreactor and increase of the S^{2-} concentration in the effluent.

Meanwhile, the removal ratio of S^{2-} exhibited a negative relationship to the inflow ratio (Figure 4(b)). The removal ratio of S^{2-} reached more than 97% at 1.0–2.0% of inflow ratio, while it declined at 2.5 and 3.0%. This suggested that a better removal efficiency of S^{2-} would be attributed to the lower inflow ratio. In addition, the higher the inflow ratio selected, the higher the processing capacity obtained. The ratio of daily treatment volume and the bioreactor volume was used to express the processing capacity in this study. As shown in Figure 4(b), the ratio of daily treatment volume and the bioreactor volume was improved with an increase of the inflow ratio. For example, for the 3 L bioreactor, the daily treatment amount could reach 86.4 L/d at 2.0% of inflow ratio, while only 43.2 L/d was obtained at 1.0% of inflow ratio. Although S^{2-} concentration in the effluent was below 1.0 mg/L which reached Grade V of the Surface Water Quality Norm of China (MEP 2002a) at 2.5% of inflow ratio, a higher quality of effluent and processing capacity could be achieved at 2.0% of inflow ratio. Therefore, in order to obtain a better treatment performance, the inflow ratio would be controlled at 2.0% for the PEPEC.

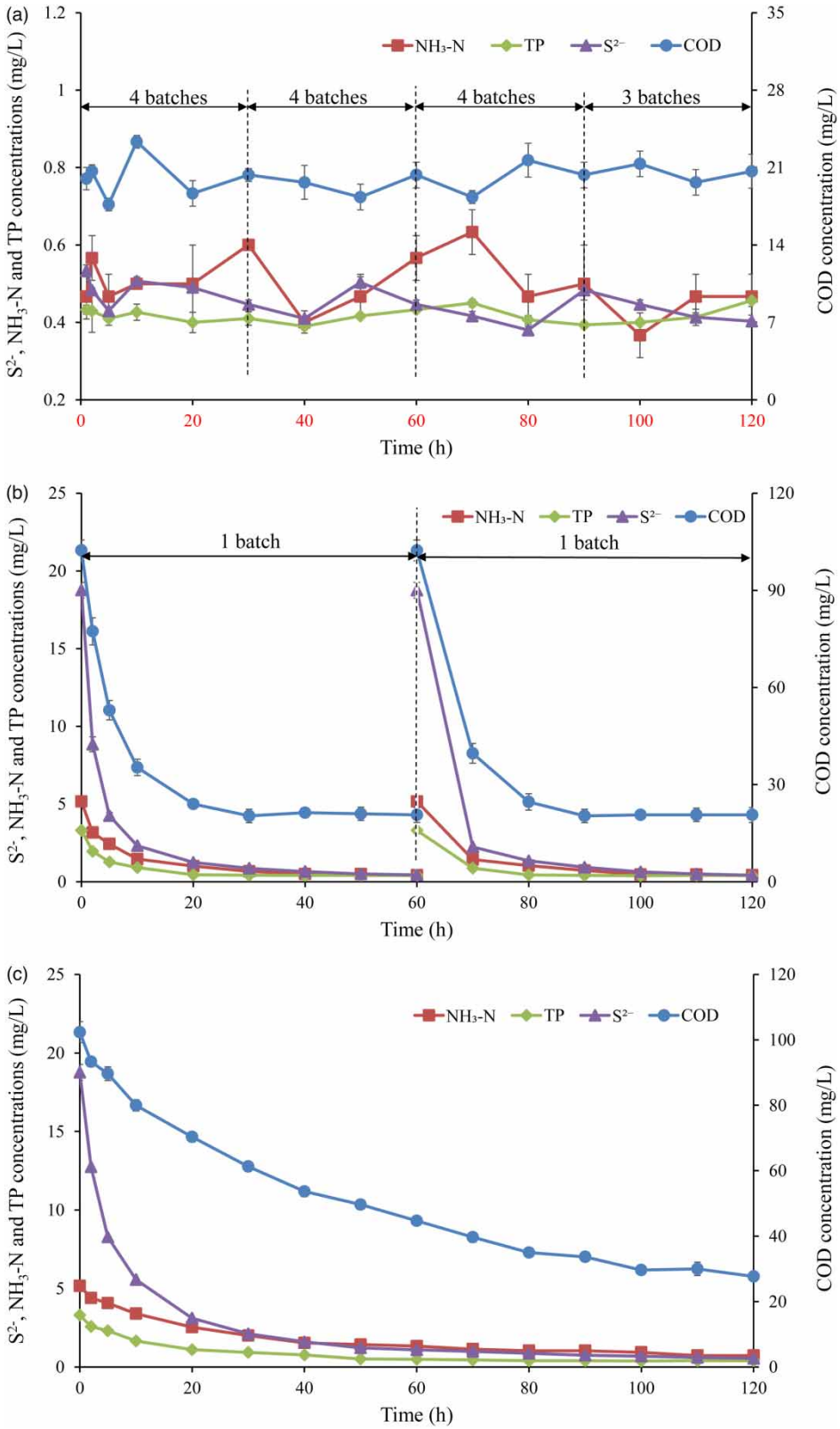

Figure 5 | Operation performances of three simulating processes. (a) The PEPEC with 1 g/L of initial biomass concentration; (b) the *in-situ* process with 1 g/L of initial biomass concentration; (c) the *in-situ* process with 0.01 g/L of initial biomass concentration.

Comparisons of the PEPEC and the *in-situ* process

The removal efficiency and processing characteristics of the PEPEC and the *in-situ* process for treating the original water sample were evaluated, and the results are shown in Figure 5.

The PEPEC was the first process and it was operated at 1 g/L of initial biomass concentration; while the *in-situ* process was run at 1 g/L (process II) and 0.01 g/L (process III), respectively. No matter which process was used, efficient removal of main dissolved pollutant was achieved. However, the treatment time for these three processes was significantly different. For PEPEC, the concentrations of S^{2-}, COD, NH_3-N and TP in the effluent could reach 0.5, 20, 0.5 and 0.5 mg/L after the rapid start-up, respectively; and then they remained relatively stable. As for process II and process III, the spending time (t) in treating dissolved pollutants to obtain the effluent concentration similar to that of PEPEC was approximately 60 and 120 h, respectively. Therefore, 15 batches could be finished during 120 h of operation using PEPEC if the treatment water volume was 30 L for each bath; but for process II and process III, batch number significantly declined to only 2 and 1, respectively. Compared with the *in-situ* processes, PEPEC showed stable operation performance and good treatment capacity.

Table 3 displays the comparisons of these three processes in treatment efficiency and processing performance. In the case that the target values of effluent concentrations and removal ratios of S^{2-}, COD, NH_3-N and TP were similar, each of the processes had different microbial consortium dosages and treatment time when the total water volume was 30 L.

The dosage of microbial consortium for PEPEC required during the reaction was 10 times that of process III, but the treatment time was only 6.67%. Although the same initial biomass concentration was inoculated, the dosage of microbial consortium and treatment time for PEPEC were only 10 and 13.3% of that of the process II, respectively. Moreover, the *ex-situ* process and the *in-situ* process exhibited an obvious difference in the daily treatment capacity. This capacity of PEPEC was 7.5 times and 15 times higher than those of process II and process III, respectively.

In addition, enriched consortium needed to be inoculated only once in PEPEC as a result of the microbe recovery by the bioreactor. As for process II and process III, the dosage would be raised with the increase of operation batches due to the microbial un-recovery for the *in-situ* process. This might show a potential 'guerrilla' capability with less cost of the *ex-situ* process for treating heavily polluted river water like Dongsha River. The water volume of each retention area along eight sampling sites of Dongsha River was in the range of 50–200 m^3. According to the operation performance of PEPEC, theoretically, 10 mini-bioreactors (1 m × 1 m × 1 m) distributed along the river could meet the requirement for the bio-treatment within 1 week. However, there would be still a long way to go from laboratory to scale-up application of the PEPEC. Effects of other factors such as excess sludge, temperature, river sediment, etc., would be investigated in the foregoing research work.

Table 3 | Comparisons of treatment efficiency and processing performance among three different processes

Items	The PEPEC	*In-situ* process II	*In-situ* process III
Water volume (L)	30	30	30
Enriched consortium dosage (g)	3	30	0.3
Initial biomass concentration (g/L)	1	1	0.01
Flow rate (L/min)	0.06	–	–
Treatment time per 30 L of water (h)	8	60	120
Daily treatment capacity (L/d)	90	12	6
Effluent concentration of S^{2-} (mg/L)	0.4–0.5	0.4	0.5
Removal ratio of S^{2-} (%)	97.5–98.0	97.8	97.1
Effluent concentration of COD (mg/L)	18–23	21	28
Removal ratio of COD (%)	77.2–82.2	79.8	72.0
Effluent concentration of NH_3-N (mg/L)	0.4–0.6	0.4	0.7
Removal ratio of NH_3-N (%)	88.1–92.1	91.6	84.8
Effluent concentration of TP (mg/L)	0.4–0.5	0.4	0.4
Removal ratio of TP (%)	82.7–86.2	87.9	86.1

Note: The PEPEC was performed at 1 g/L of biomass concentration and 2% of inflow ratio; the *in-situ* process II and the *in-situ* process III were performed at 1 and 0.01 g/L of biomass concentration, respectively.

CONCLUSION

The initial biomass concentration had an obvious impact on the S^{2-} removal efficiency. The initial S^{2-} concentration could only have an impact on treatment time but had little influence on the average removal rate of S^{2-}. According to the kinetic models of S^{2-} removal, the inflow ratio was optimized at 2%. The removal ratio of dissolved S^{2-} could reach 97.5% ± 0.5% when the inflow ratio was controlled at 2%. Compared with the *in-situ* processes, the PEPEC showed a stable operation performance during 120 h of bio-treatment, and the concentrations of S^{2-}, COD, NH_3-N and TP in the effluent reached approximately 0.5, 20, 0.5 and 0.5 mg/L, respectively. The PEPEC showed less consortium dosage and time cost, and it also achieved higher daily treatment capacity than the *in-situ* processes. This work demonstrated that the PEPEC presented some potential advantages for the bio-treatment of a heavily polluted river.

ACKNOWLEDGEMENTS

This work was financially supported by the Central Level, Scientific Research Institutes for Basic R&D Special Fund Business (2019YSKY008) and the Natural Science Foundation of Beijing, China (Grant No. 8182058).

DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

CONFLICT OF INTEREST

The authors declare there is no conflict.

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First received 28 February 2022; accepted in revised form 21 July 2022. Available online 8 August 2022