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Integration of Inorganic Micronutrients and Natural Starch Based Cationic Flocculant in Primary Treated Sewage Effluent (PTSE) Treatment

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In this study, a natural starch-based cationic flocculant (SBCF) was first evaluated using a granular activated carbon fluidized-bed bioreactor (GAC-FBBR) to treat a high strength synthetic domestic wastewater (primary treated sewage effluent) containing refractory organic matters. The positive effect of SBCF on microorganisms and organic removal was obviously observed. When the optimal dose of SBCF (22 mg/L) combined with three major inorganic micronutrients (CaCl_2 , MgSO_4 , and FeCl_3) at different concentrations, the best modified dosages of 0.5 mg/L of FeCl_3 , 5 mg/L of MgSO_4 and 2 mg/L CaCl_2 could significantly improve the microbial activity and organic removal simultaneously.

Keywords microbial activity; natural flocculant; refractory organic pollutants; trace nutrients; wastewater treatment

INTRODUCTION

The flocculants used in the flocculation process for the water and wastewater treatment, can be classified into three groups:

1. inorganic flocculants
2. organic synthetic polymer flocculants and
3. naturally occurring bio-polymer flocculants (1).

Inorganic flocculants are the most common flocculants used in water and wastewater treatment. Polymeric flocculants, synthetic as well as natural, because of their natural inertness to pH changes, low dosage, and easy handling, have become very popular in wastewater treatment (2). Although inorganic and organic synthetic polymer flocculants have been widely used due to their high flocculating efficiency and low cost, some of them are strong carcinogens and neurotoxic to humans. In addition,

nonbiodegradable property presents another major drawback of polymeric flocculants, which will lead to “secondary pollution” of the environment (3).

The natural based flocculants (NBFs) are environmentally friendly and biodegradable, and they also present good flocculating ability. They can minimize the environmental and health risks and have attracted more attention in water and wastewater treatment. The advantages of NBFs are

1. virtually toxic free;
2. biodegradable in the environment;
3. the raw products are often locally available, whereas industrialized flocculants may not be, and
4. the sludge from flocculation may be reused (4,5).

The common NBFs can be processed from various sources of starches, such as potato, corn, cassava, arrowroot, and yams. These starch-based flocculants (SBFs) can be nonionic, cationic, or anionic depending on the forms of processing and the substitutions. Since the 1980s, some SBFs have been applied in water and wastewater treatment. The studies carried out by Campos et al. (6) proved that the addition of starch flocculants could enhance flocculation and sedimentation. Using 0.5 mg/L activated arrowroot starch, the alum dosage could be reduced by 20% and achieve better settled and filtered water qualities when compared with the use of alum alone. In addition, Denes and Marton employed a starch-based anionic flocculant (Greenfloc® 213A) in water treatment. After flocculation of Greenfloc® 213A, approximately 87.5% of turbidity and 43.2% of COD were removed from raw water (7).

Biodegradability of the flocculant is one of the most important environmental aspects of the environmental behavior as it will cause less ecological problems in the long term than a persistent one while providing a carbon source for the microbial activities. Xie et al. (3) indicated that the bacteria are capable of utilizing natural polysaccharide as a

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carbon source. In other words, the flocculant can be biodegraded by itself under suitable conditions such as temperature, UV, moisture level, oxygen, nutrients, etc. Chang et al. (8) also evaluated the biodegradability of a copolymer of acrylamide and acryloyloxyethyltrimethylammonium chloride (AM/AETAC) by measuring biochemical oxygen demand (BOD) under aerobic conditions and performing a batch bioassay method (serum bottle test) for the anaerobic environment. The results conclusively showed the AM/AETAC polymer was subject to partial hydrolysis and degradation. Singh et al. (2) investigated the biodegradability of polymers (grafted polysaccharides) by monitoring the viscosity decay, which showed that they were very efficient, shear stable, and biodegradable flocculants. They also exhibited turbulent drag reducing characteristics.

As NBFs can provide the carbon source for biodegradation, the additives are helpful for biomass growth and enhance the microbial activities in the biological process for wastewater treatment. However, besides the carbon source, the trace nutrients such as magnesium, calcium, iron, etc. are also very necessary and useful for the metabolism of microorganisms. The trace nutrients limitation could deteriorate the organic removal and affect the biofilm growth when running at a high organic loading rate (9). Gbolagade (10) reported that magnesium and calcium were the best macronutrients for the biomass production. In addition, Gobler and Sanudo-Wilhelmy (11) found that iron also enhanced some of the bacterial growth at suitable concentrations.

Thus, the main aims of this study are:

1. to evaluate the effect of trace nutrients on the biodegradability of a natural starch based cationic flocculant (SBCF) by viscosity decay,
2. to investigate the microbial activity in a granular activated carbon fluidized bed bioreactor (GAC-FBBR) with SBCF addition, and
3. to find out the optimal concentrations of inorganic micronutrients (Fe, Ca, and Mg) when combining with SBCF as a new flocculant.

MATERIALS AND METHODS

Synthetic Wastewater

The experiments were conducted using synthetic wastewater to avoid any fluctuation in the feed concentration and provide a continuous source of biodegradable organic pollutants together with refractory organics such as humic acid, tannic acid, lignin, polysaccharide, and other high molecular carbohydrates (Table 1). The synthetic wastewater originally contained some trace nutrients, which was used to simulate primary treated sewage effluent (PTSE) (just after the primary treatment process). The

TABLE 1
Composition of synthetic PTSE

Compound	Concentration (mg/L)
Glucose	230
$(\text{NH}_4)_2\text{SO}_4$	71
KH_2PO_4	13.2
Peptone	2.7
Humic acid	4.2
Tannic acid	4.2
(Sodium) lignin sulfonate	2.4
Sodium lauryl sulphate	0.94
Acacia gum powder	4.7
Arabic acid (polysaccharide)	5
Trace nutrient	
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	5.07
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	0.368
$\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$	0.275
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.44
FeCl_3	1.45
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.391
$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	0.42
$\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$	1.26
Yeast extract	20

average concentration of the dissolved organic carbon (DOC), chemical oxygen demand (COD), ammonium-nitrogen ($\text{NH}_4\text{-N}$), and orthophosphate ($\text{PO}_4\text{-P}$) contained in PTSE were 120, 300, 15, and 3.3 mg/L, respectively (COD:N:P = 100:5:1).

Natural Starch Based Cationic Flocculant (SBCF) and GAC Used

SBCF provided by HYDRA 2002 Research, Development and Consulting Ltd., Hungary was selected as a representative SBF in this study. The major components of this flocculant are cationic starch ether and water. It is completely soluble in water with a density of 1050 kg/m^3 . The coal based granular activated carbon (GAC) (ACTI-CARB GS1300, Activated Carbon Technologies Pty Ltd, Australia) was used in this study. The surface area, iodine number, maximum ash, and moisture contents are $>1100 \text{ BET m}^2/\text{g}$, $>1100 \text{ mg/g}$, 10% and 3%, respectively. The GAC was rinsed with distilled water to remove fines and dried at 105°C in the oven prior to the experiments.

Biodegradation Study of SBCF

1% SBCF solution (dissolved in distilled water) was used for this study and the biodegradability of SBCF was evaluated by monitoring the viscosity decay using a falling ball type viscosity meter (Gilmont[®] Instruments) with the range of 0.2–10 mPa.s. The viscosity measurement was

conducted at room temperature of 25°C. To evaluate the effect of nutrients on the SBCF biodegradation, $(\text{NH}_4)_2\text{SO}_4$ as nitrogen, KH_2PO_4 as phosphorus, and trace nutrients (MgSO_4 , CaCl_2 and FeCl_3) were added into SBCF solution. The pH of solution was maintained at 7.

Effect of SBCF on Microbial Activity

The effect of SBCF on microbial activity was investigated by comparing GAC inoculation (75 mL of GAC) in three fluidized bed bioreactors (FBBRS) with 100% recirculation for 15 days. A volume of 10 L/day PTSE was fed into each FBBR at a feeding rate of 180 mL/min. Three GAC-FBBRs were operated simultaneously at an actual depth of 240 mm with bed expansion of 60 mm. One of the GAC-FBBRs was a parallel control system without SBCF addition, while 11 mg/L SBCF (containing 5 mg/L TC) and 22 mg/L SBCF (containing 10 mg/L TC) were added in the other two GAC-FBBRs every day, respectively. The biomass attached on the GAC, the oxygen uptake rate (OUR) and DOC were monitored.

Optimizing the Concentrations of Inorganic Micronutrients

Besides the organic carbon, trace nutrients such as calcium, magnesium, and iron are also very important for biomass growth. Thus, SBCF was combined with the three major trace nutrients in different concentrations, and jar tests were conducted using GAC as attached growth media (25 g/L GAC) for 20 days. The mixtures of SBCF and inorganic nutrients as bioflocculant were added to 1 L beakers to treat PTSE. A certain amount of GAC was taken out periodically for analyses. The microbial activities were investigated in terms of biomass growth, and OUR.

Analysis

DOC was measured using the Analytikjena Multi N/C 2000. The analysis of biomass (monitored as mixed liquor volatile suspended solids, MLVSS) was according to Standard Methods (12). For measuring MLVSS, two samples were taken each time and the average value was then calculated. YSI 5300 Biological Oxygen Monitor was used to measure the oxygen uptake rate (OUR) due to its usefulness in measuring samples including respiration, oxidative activity, and cellular metabolism studies.

RESULTS AND DISCUSSION

Biodegradability of SBCF

There are many factors which affect the biodegradability of SBCF such as temperature, moisture level, oxygen, UV, etc (2). To eliminate the influence of these factors, the temperature, moisture, oxygen, and UV were controlled in the common room conditions. Since all of the solutions were prepared using distilled water, the decay

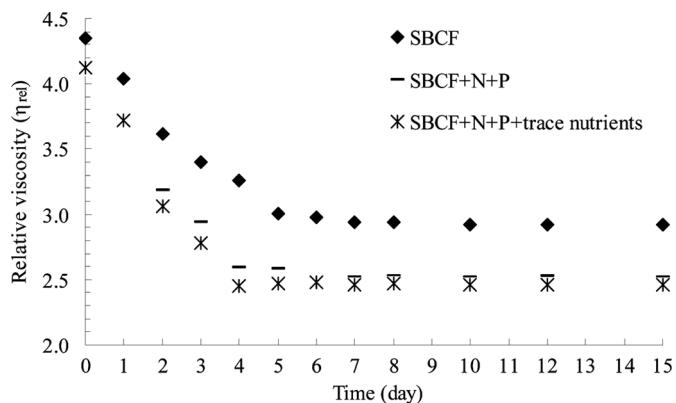


FIG. 1. Relative viscosity versus time to study the biodegradation of SBCF with and without nutrients addition (Temperature = 25°C).

of relative viscosity η_{rel} (ratio of the testing solution's viscosity to pure distilled water's viscosity) was used to examine the biodegradability of SBCF and the results were shown in Fig. 1. As can be seen, the biodegradation has been observed for all the SBCF solutions with and without the addition of nutrients. The relative viscosity of SBCF solution decreased from 4.35 to 2.98 during the first 6 days and reached a stable stage afterwards. However, faster biodegradation was found in the cases of SBCF solution with the N and P addition (η_{rel} decrease of 1.63), and SBCF solution with N, P, and trace nutrients (η_{rel} decrease of 1.66). It indicated that as a starch-based flocculant, SBCF is biodegradable and the addition of nutrients could enhance the SBCF biodegradation.

Effect of SBCF on Microbial Activity

Fig. 2 and Table 2 showed the biomass attached on GAC and OURs of biomass in three GAC-FBBRs with and without SBCF addition. The biomass attached on GAC in FBBR with 22 mg/L SBCF addition elucidated the fastest increase (up to 5.75 g/L) within the operation

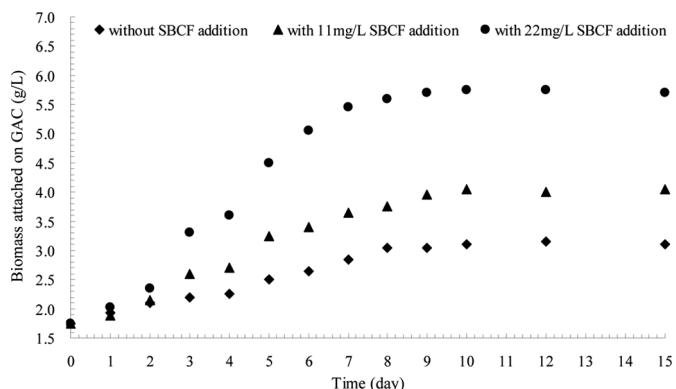


FIG. 2. Biomass growth on GAC in three GAC-FBBRs with and without SBCF addition.

TABLE 2
Comparison of OUR variation of biomass attached on GAC with and without SBCF addition

Day	OUR (mgO ₂ /L·h)		
	Without SBCF addition	With 11 mg/L SBCF addition	With 22 mg/L SBCF addition
1	10.87	6.96	11.34
5	12.49	13.38	16.63
10	8.64	12.27	15.65
15	10.66	12.07	16.44

period, while the control FBBR and FBBR with 11 mg/L SBCF addition had 3.1 g/L and 4.05 g/L biomass growth respectively. The OUR can be used as an indicator for microbial activity on GAC at different period of acclimatization, as it presents the dissolved oxygen (DO) consumption rate of biomass on the GAC. The OUR of biomass on the GAC in FBBR with 22 mg/L SBCF also had better performance and could reach equilibrium faster than others. Both of the biomass and OUR results showed that there were many microbial substances on the GAC in FBBR with the addition of 22 mg/L SBCF. Hence, with the higher dosage of SBCF, the higher biomass growth could be observed in FBBR.

The DOC removal efficiencies of three GAC-FBBRs with and without SBCF addition were illustrated in Fig. 3. DOC removal efficiencies of all the FBBRs kept increasing until the 10th day when the biomass growth on GAC started to reach steady phase. Compared with other GAC-FBBRs, the GAC-FBBR with 22 mg/L SBCF addition had much better performance, resulting in the highest DOC removal of 66% while only 49% and 55% of DOCs were removed by control GAC-FBBR and the one with 11 mg/L SBCF addition. Under the same operating

conditions, the performance of GAC-FBBR was dependent on the number of microorganisms attached onto the GAC for organic biodegradation. Thus, the DOC removal efficiency also revealed that the SBCF could provide a carbon source for the biomass growth and be very helpful for microbial activities as a biodegradable flocculant.

The Effect of Inorganic Micronutrients on SBCF Performance

Combined SBCF with Individual Inorganic Trace Nutrient

The combinations of SBCF (22 mg/L) with two individual inorganic trace nutrients (CaCl₂ and MgSO₄) were evaluated by comparing the microbial activity of biomass growth on GAC. GAC was acclimatized for 20 days through the jar tests. When the concentrations of CaCl₂ varied from 2 to 10 mg/L, 2 mg/L and 5 mg/L concentrations resulted in the higher biomass growth, which led to biomass of 2.95 mg/L, 2.9 mg/L respectively (Fig. 4). The results also indicated that CaCl₂ was helpful for biomass growth. For all the cases, significant growth of biomass was observed in the first 10 days. Similarly, the better OUR values were obtained with 2 and 5 mg/L CaCl₂ addition (1.89 and 1.69 mgO₂/L·h respectively) (Table 3). However, with 10 mg/L of CaCl₂, the OURs dropped more than 2 times (0.79 mgO₂/L·h) indicating an overdose of the nutrient. Hotchkiss (13) has also reported that the bi-valent salts were more toxic than the mono-vaalent salts. High concentration of calcium was toxic and could inhibit the growth of bacteria. Although 2 and 5 mg/L CaCl₂ addition did not present much difference on the biomass growth, the 2 mg/L CaCl₂ always exhibited the strongest micro-activity of the microorganisms attached on GAC which was correspondent to the highest DO consumption rate within 30 mins. Thus, 2 mg/L CaCl₂ was selected as the favorable concentration for a combination with SBCF.

The evaluation of combined SBCF and different concentrations of MgSO₄ for GAC acclimatization was also

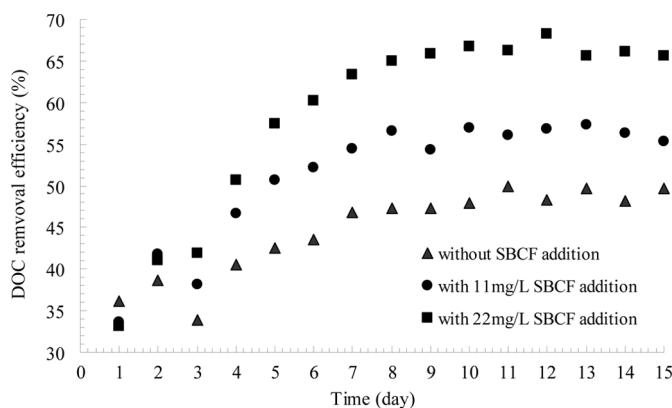


FIG. 3. DOC removal efficiencies of three GAC-FBBRs with and without SBCF addition (average initial DOC = 120 mg/L).

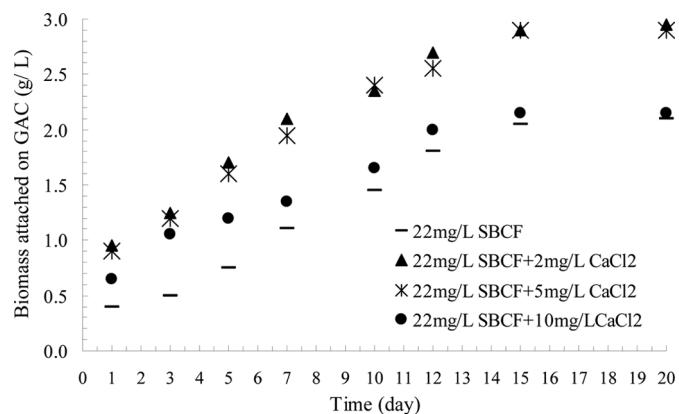


FIG. 4. Biomass attached on GAC for 22 mg/L of SBCF combined with different concentrations of CaCl₂.

TABLE 3
Comparison of OUR values for SBCF combined with different concentrations of CaCl_2

Day	OUR (mg $\text{O}_2/\text{L} \cdot \text{h}$)			
	22 mg/L SBCF	22 mg/L SBCF + 2 mg/L CaCl_2	22 mg/L SBCF + 5 mg/L CaCl_2	22 mg/L SBCF + 10 mg/L CaCl_2
5	0.54	0.64	0.88	0.59
7	0.76	1.15	1.13	0.96
10	0.71	1.40	1.21	0.88
12	0.95	1.98	1.40	0.99
15	0.88	1.89	1.69	0.79

TABLE 4
Comparison of OUR variation for SBCF combined with different concentrations of MgSO_4

Day	OUR (mg $\text{O}_2/\text{L} \cdot \text{h}$)			
	22 mg/L SBCF	22 mg/L SBCF + 2 mg/L MgSO_4	22 mg/L SBCF + 5 mg/L MgSO_4	22 mg/L SBCF + 10 mg/L MgSO_4
5	0.54	0.78	1.28	1.64
7	0.76	0.96	1.28	2.04
10	0.71	1.28	1.50	2.10
12	0.95	1.22	1.81	1.69
15	0.88	0.95	1.89	1.83

performed (Table 4 and Fig. 5). The combined flocculant using 22 mg/L SBCF and 2 mg/L MgSO_4 gained the lowest biomass growth (2.55 g/L), whereas both of the biomass attached on the GAC for addition of 22 mg/L SBCF combined with 5 mg/L and 10 mg/L MgSO_4 achieved a steady phase and was around 3.1 g/L after 15 days operation. OURs indicated the stronger microbial activity when employed MgSO_4 concentrations of 5 and 10 mg/L (1.89 and 1.83 mg $\text{O}_2/\text{L} \cdot \text{h}$ respectively). Thus, 5

and 10 mg/L were selected for conducting the next-step experiment.

Combination of SBCF Together with FeCl_3 , CaCl_2 and MgSO_4

According to the optimum concentrations of SBCF (22 mg/L), CaCl_2 (2 mg/L) and MgSO_4 (5 and 10 mg/L), FeCl_3 was varied at three different concentrations of 0.5, 1, and 2 mg/L. The combinations of different compounds were shown in Table 5. The biomass attached on GAC and OURs were measured and the results were shown in Fig. 6 and Table 6. As can be seen from the figure, the

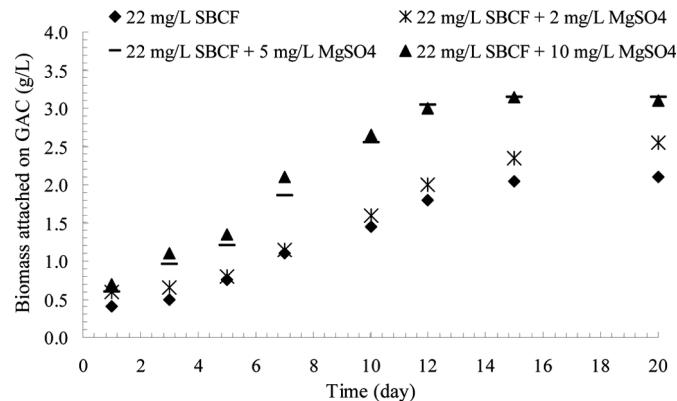


FIG. 5. Biomass attached on GAC for 22 mg/L of SBCF combined with different concentrations of MgSO_4 .

TABLE 5
The combined conditions of inorganic trace nutrients with SBCF

Flocculant ID	SBCF (mg/L)	FeCl_3 (mg/L)	MgSO_4 (mg/L)	CaCl_2 (mg/L)
A	22	0.5	5	2
B	22	1	5	2
C	22	2	5	2
D	22	0.5	10	2
E	22	1	10	2
F	22	2	10	2

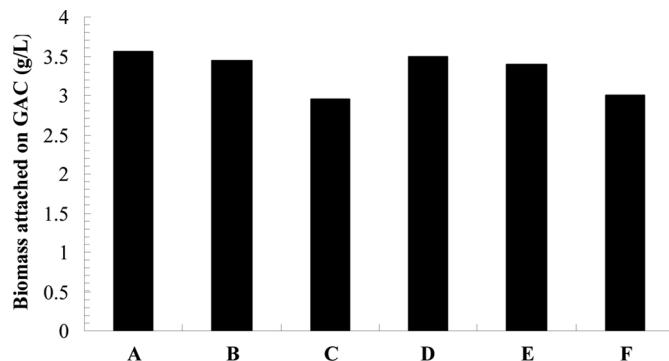


FIG. 6. Biomass attached on GAC with different combinations of flocculants on 15th day (SBCF of 22 mg/L and CaCl_2 of 2 mg/L for all conditions; MgSO_4 of 5 mg/L for A, B, C and 10 mg/L for D, E, F; FeCl_3 of 0.5 mg/L for A, D; 1 mg/L for B, E; 2 mg/L for C, F).

lowest dose of FeCl_3 (0.5 mg/L) illustrated the highest number of biomass yield (3.55 mg/L for Flocculant A and 3.5 mg/L for Flocculant D). On the contrary, the high dose of FeCl_3 led to decline of the biomass growth while the decreased values of OUR also confirmed the negative effect of high FeCl_3 concentration on bioactivity. For instance, the OURs of Flocculant C and F dropped significantly from 8.74 to 3.43 $\text{mgO}_2/\text{L} \cdot \text{h}$ and from 11.54 to 4.07 $\text{mgO}_2/\text{L} \cdot \text{h}$, respectively. The result was similar to the previous research about the influence of FeCl_3 concentrations on the microorganism growth (11). In addition, 5 and 10 mg/L MgSO_4 exhibited similar biomass growth on GAC and OURs when different doses of FeCl_3 were varied. Based on the economical point of view, the 5 mg/L was chosen to be the optimal MgSO_4 concentration for the combined flocculant. Figure 7 showed the DOC removal efficiency of different combined flocculants. Similar to the biomass and OURs observed, Flocculant A and D resulted in $71.8 \pm 13.4\%$ and $70.6 \pm 13.1\%$ organic removal form PTSE. Meanwhile, the worst DOC removals were obtained when applying Flocculant C and F ($63.1 \pm 8\%$ and $60.7 \pm 9\%$ respectively). Thus, compared with other combined flocculants, Flocculant A (22 mg/L SBCF + 0.5 mg/L

TABLE 6
Comparison of OUR variation with different combined flocculants

Day	OUR ($\text{mgO}_2/\text{L} \cdot \text{h}$)					
	A	B	C	D	E	F
3	1.72	1.32	1.25	1.49	1.39	0.63
5	9.62	3.58	5.26	9.75	4.29	1.01
7	11.29	4.78	4.44	11.20	8.37	5.92
10	14.03	7.84	6.56	12.08	4.58	3.65
12	15.77	10.73	6.42	15.18	10.61	6.83
15	8.74	5.42	3.43	11.54	6.12	4.07

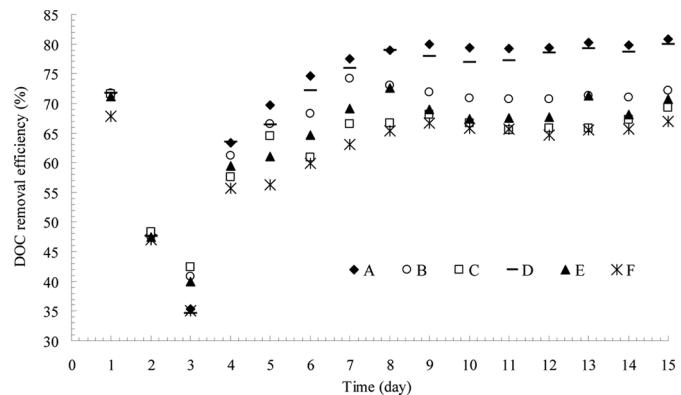


FIG. 7. DOC removal efficiency of the different combined flocculants (initial DOC = 120 mg/L; SBCF of 22 mg/L and CaCl_2 of 2 mg/L for all conditions; MgSO_4 of 5 mg/L for A, B, C and 10 mg/L for D, E, F; FeCl_3 of 0.5 mg/L for A, D; 1 mg/L for B, E; 2 mg/L for C, F).

$\text{FeCl}_3 + 5 \text{ mg/L } \text{MgSO}_4 + 2 \text{ mg/L } \text{CaCl}_2$) is considered as the most effective flocculant and will be applied for the further studies.

CONCLUSIONS

The study investigated the performance of a SBCF for treating a synthetic PTSE containing refractory organic pollutants. Several important inorganic trace nutrients (CaCl_2 , MgSO_4 , and FeCl_3) were selected to modify the SBCF in order to improve the organic removal and enhance the bio-activity of microorganisms attached on GAC. The outcomes can be summarized as follows:

- SBCF showed good biodegradability by viscosity decay and the inorganic nutrients addition could enhance SBCF biodegradation. However, a higher concentration of FeCl_3 resulted in inhibition of biomass growth.
- As a biodegradable flocculant, SBCF could provide the carbon source for the biomass growth and enhance the organic removal in GAC-FBBRs. 22 mg/L SBCF addition led to almost double the amount of biomass on GAC in FBBR than that without the SBCF addition.
- The modified flocculant containing 22 mg/L of SBCF, 0.5 mg/L of FeCl_3 , 5 mg/L of MgSO_4 and 2 mg/L CaCl_2 was evaluated through the batch tests and considered as a better flocculant used in future study.

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