Applications of Biological Flocculants (BFs) for Coagulation Treatment in Water Purification: Turbidity Elimination

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In this study, coagulation performance by using biological flocculants (BFs; produced from strain *Bacillus* sp. F6) for turbidity removal was investigated. The results demonstrated that BFs were able to eliminate turbidity from kaolin clay solution over a wide dosage range (γ_{DR} = 6–20 mg L^{-1}). The removal efficiency with BFs reached 86 % on average, lower than 95 % with $Al_2(SO_4)_3$ (γ_{Al} = 6.5 mg L^{-1}) and 96 % with $Fe_2(SO_4)_3$ (γ_{Fe} = 10 mg L^{-1}), respectively. For bioflocculants, bridging flocculation other than charge neutralization should be responsible for turbidity removal. The combined applications of BFs with $Al_2(SO_4)_3$ ($\zeta_{Al/BF}$ = 0.33) and $Fe_2(SO_4)_3$ ($\zeta_{Fe/BF}$ = 0.05) increased overall turbidity removal up to 97 %. It was also shown that combination of BFs and $Fe_2(SO_4)_3$ was effective for removing turbidity from raw water. This study provides a proof-in-concept demonstration of BFs for water purification, which can in part reduce operational costs in coagulation treatment, as well, effectively reduce the concentration of residual metallic elements (e.g. aluminum) in coagulated solution.

Key words:
Bioflocculants (BFs), coagulation, turbidity removal, charge neutralization, bridging flocculation

Introduction

Natural water body used for a water source contains suspended particles (d_p > 1.0 μm) and colloidal substances (d_s = 1.0 nm – 1.0 μm). In water treatment, suspended particles can be easily settled or filtered via physical processes for their relatively large size and density; while colloidal impurities in natural water are negatively charged and repel each other, hence, resulting in a physicochemical stability for a long period of time.

Coagulation or flocculation has been widely adopted as one of the most effective methods to remove colloidal particles in water or wastewater treatment. By adding highly charged cations, colloidal particles are destabilized, thereby, forming larger aggregates and flocs that can be effectively separated by subsequent sedimentation, flotation or filtration units. It is obvious that coagulant plays an extremely significant role in coagulation treatment.

The most frequent used chemical agents for coagulation are known as inorganic aluminum salts such as aluminum chloride ($AlCl_3$) or aluminum-based polymeric materials (i.e. polyaluminum chloride (PACl)), and their performances in eliminating turbidity and dissolved organic carbon (DOC) have been extensively investigated.[1,2] Although these coagulants have the ability to efficiently remove turbidity as well as DOC from raw water, it should be realized that the problems in correlation to the use of aluminum still remain and these problems have been taken into consideration for several years.[3,4] That is, the utilization of aluminum as the coagulant in water treatment may lead to a higher level of aluminum in the treated effluent than in raw water. In addition, residual aluminum in excessive sludge produced during coagulation tends to accumulate in the environment. Parkinson *et al.* established a link between aluminum in drinking water and human neurological disorders such as dialysis encephalopathy.[5] Excess aluminum in dialysate fluid was shown harmful to dialysis patients.[6] Moreover, the possibility of links between aluminum and neuropathological diseases including presenile dementia and Alzheimer's disease has been frequently reported.[7–11] Recently, the European Commission regulated that the maximum contaminant level of aluminum in drinking water cannot exceed γ_{CL}= 200 μg L^{-1}.[12] Consequently, in order to reduce the nosogenetic risks posed by the use of aluminum coagulants, more environment-friendly and safe coagulants for pollutant removal in water treatment remain necessary to be developed.

During recent years, microbially produced flocculants have been drawing increasing attention by virtue of their satisfied flocculating effectiveness and safe biodegradable nature. Many classifications of microorganisms such as bacteria, fungi, yeast and algae, which are capable of producing extracellular

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bio-polymeric flocculants, have been isolated from various sources (e.g. activated sludge and soils) and some of them have been identified.[13] Several research works have provided a demonstration of microbial flocculants in not only removing suspended solids, chemical oxygen demand (COD),[14] humic acids[15] and heavy metals[16] from waste streams, but in separation of oil from oil-water emulsions[17] and fine coal processes.[18] However, there have been no previous reports of coagulation using bioflocculants in drinking water treatment. In fact, in the view of safety and health problems, bioflocculants are possibly considered a good selection for water purification.

This paper aims to examine the performance and efficiency of coagulation in turbidity removal by using bioflocculants (BFs) on the basis of synthetic solution and raw water in the Songhua River. The scope of this study comprises three phases, as follows: (i) to compare coagulation for turbidity elimination between $Al_2(SO_4)_3$, $Fe_2(SO_4)_3$ agents and BFs individually; (ii) to investigate removal of turbidity from synthetic solution with BFs in combination with inorganic aluminum salts and iron salts, respectively; (iii) to take a case study on turbidity removal from raw water by using BFs combined with Fe salts.

Materials and methods

Coagulation materials

Bioflocculants examined in this study were produced by the bacterial strain F6. Such strain was isolated from aerobic activated sludge in aeration tank of a wastewater treatment plant, Daqing, PR China. The flocculants-producing strain F6 has been characterized as *Bacillus* sp.[19] By using the gel chromatogram technology, the molecular mass of the flocculants was measured in the range between $2.0 \cdot 10^6$ and $2.5 \cdot 10^6$ million gram per mole. The main components of the flocculants have been identified as polysaccharide. As shown in Fig. 1, SEM image of the BFs reveals a colonial morphology as a relatively uniform structure with mucilaginous substances. Prior to usage, the liquid-state bioflocculants were produced into solid-state fine powder;[19] then 1 g BF powders were dissolved in 1 L deionized water. The BF solution with a mass concentration of 1000 mg L^{-1} was then obtained. In some cases, 1000 mg L^{-1} of original BF solution was diluted into different liquid coagulant with various concentrations.

To compare coagulation performance between chemical flocculants and BFs, and to examine the coagulation efficiency with bioflocculants combined with chemical flocculants, inorganic aluminum sulfate ($Al_2(SO_4)_3$) and ferric sulfate ($Fe_2(SO_4)_3$) were used as representation of chemical flocculants in this study. The stock solution was prepared by dissolving 2.0 g of $Al_2(SO_4)_3 \cdot 18H_2O$ and 2.0 g of $Fe_2(SO_4)_3$ agents (analytical grade; Nanning, China) in 1 L of deionized water, respectively. Different concentrations of the agents were obtained via dilution procedures with deionized water. In some tests, active adjustment of coagulation pH was done by HCl (1 mol L^{-1}) or $Ca(OH)_2$ (1 mol L^{-1}) solution. The dosage for $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ is expressed as the unit of γ_{Al} = mg L^{-1} and γ_{Fe} = mg L^{-1}, respectively.

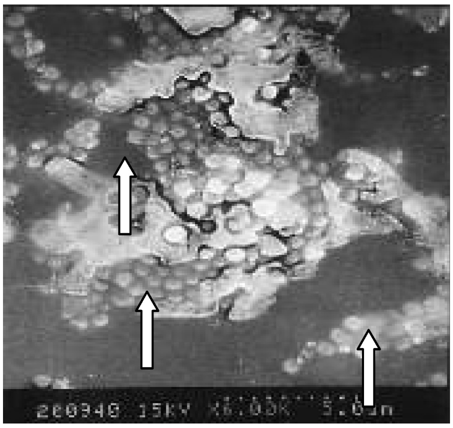


Fig. 1 – *SEM images taken of BFs produced by Bacillus sp. F6 (×6000). Arrowheads indicate the biologically produced flocculants.*

Synthetic solution and raw water

Synthetic solution in this study was prepared by dispersing a given amount of stock suspension of kaolin clay in deionized water under high-rate mixing conditions. Concentrations of 0.5 mol L^{-1} $NaHCO_3$ and 0.5 mol L^{-1} $NaNO_3$ (analytical grade) were used to adjust alkalinity and conductivity of the working solution. After a settlement of suspension overnight, a mixed solution with initial turbidity of 80 NTU and pH of 7.5 was achieved. Prepared synthetic solution was used to (i) take a comparison of BFs and chemical flocculants for coagulation; and (ii) accomplish feasible study on turbidity removal by adding Al or Fe salts to BFs, respectively.

In the case study section, in order to examine the performance of turbidity removal from raw water (Songhua River) with BFs, raw water were sampled from the Songhua River, PR China and the basic characterizations of water quality were referred in Table 1.

Table 1 – *Physicochemical characterization of raw water samples from Songhua River*

Parameters	Values
pH (20 °C)	7.8
alkalinity (γ_{CaCO_3}/mg L^{-1})	322
turbidity/NTU	121
COD_{Mn} /mg L^{-1}	51.7

Jar tests and analyses

Jar tests were carried out by using a 1 L six-place paddle stirrer (type SC2000-6, China). The operational procedures included rapid mixing (200 rpm) for 1 min, slow mixing (30 rpm) for 15 min and settlement for 30 min in turn. After the rapid mixing, a small quantity was sampled for zeta potential measurement (3000Hsa Zetasizer, UK). At the end of settlement, supernatants were collected for the measurements of residual turbidity (RT) using a turbidimeter (555IR, WTW Company, Germany), final aqueous pH using a pH meter (Delta 320A, China) as well as residual metals (Fe or Al) using an atomic absorption method (ISP-OES, 5300DV, Perkin Elmer Company, US). In some tests, images of flocs produced with different coagulants were recorded at the end of the slow mixing phase when the aggregates started to settle by using an on-line infrared camera.

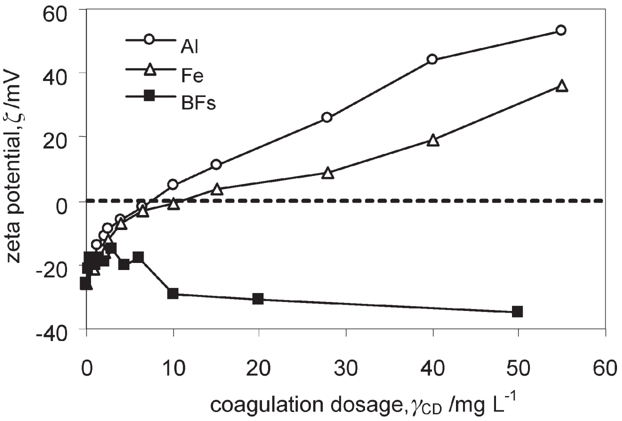


Fig. 2 – *Zeta potential during coagulation as function of coagulation dosage (pH 7.5)*

Results and discussion

Comparison of coagulation performance with BFs and chemical coagulants

The comparative coagulation performance between $Al_2(SO_4)_3$, $Fe_2(SO_4)_3$ and pure BFs from *Bacillus* sp. F6 for turbidity removal was examined. As shown in Fig. 2, variation of zeta potentials for coagulating with $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ was observed quite similar, i.e. as coagulant dosage was increased, zeta potentials remained negative and increased to a value close to isoelectric point, where turbidity removal efficiency reached the optimal value of 95 % for $Al_2(SO_4)_3$ (γ_{Al} = 6.5 mg L^{-1}) and 96 % for $Fe_2(SO_4)_3$ (γ_{Fe} = 10 mg L^{-1}), respectively (Fig. 3). This appears to be consistent with the fact that the dominant mechanism for turbidity removal at low dosage is charge neutralization.[20] Thereafter, further increasing dosage resulted in an increase of zeta potentials higher than zero and a corresponding decrease of turbidity removal efficiency, which indicated the occurrence of particle restabilization. Fig. 4 shows that pH of solution coagulated by $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ is largely dependent on coagulant dosage and there is only a narrow dosage region to maintain optimal pH for turbidity removal. On the contrary, the coagulation behavior using BFs at the same water conditions was found quite different when $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ were used. By using BFs, turbidity removal efficiency sharply increased to 84 % (γ = 6 mg L^{-1}), then remained within a wide range of dosage from γ = 6 to 20 mg L^{-1} (Fig. 3). Zeta potentials were always below zero ($\xi < -16$ mV) during coagulation, even for the case of optimal dosage (Fig. 2). Unlike $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$, an increase of BFs dosage could not lead to pH decrease (Fig. 4). Although the mechanism of bio-flocculation is not understood well, the results obtained here suggest that coagulation with BFs should be a process that is independent of charge neutralization. It is known that colloidal particles present negative charges at pH range of 6–8. Likewise, BFs carry negative charges over

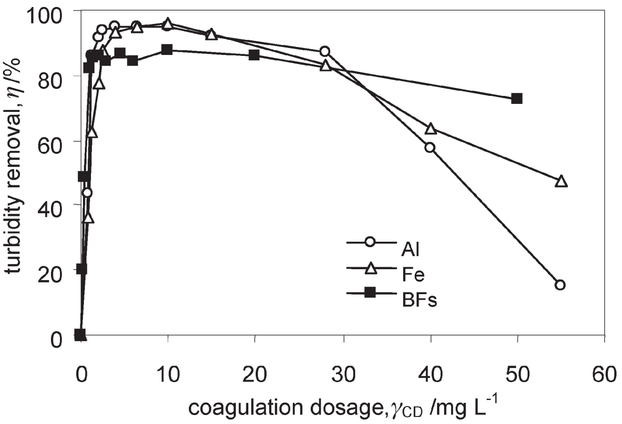


Fig. 3 – *Removal rate of turbidity from synthetic solution as function of coagulant dosage (pH 7.5)*

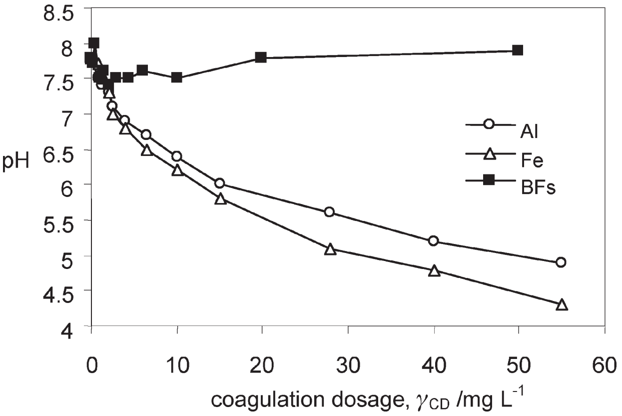
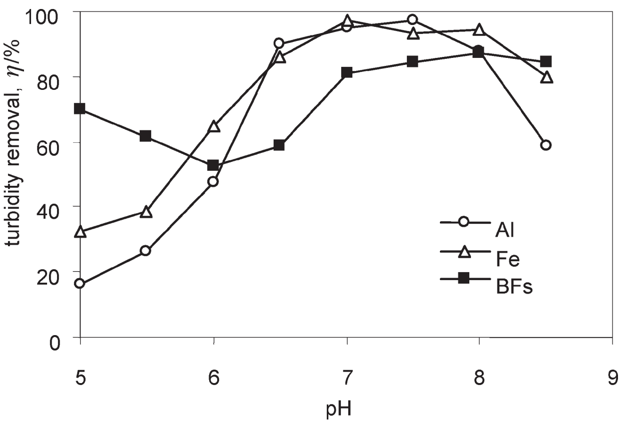


Fig. 4 – *Variation of finished pH as function of coagulation dosage (initial pH 7.8)*

the same pH region (Fig. 2). Hence, charge neutralization between particles and BFs impossibly operates. Instead, some other mechanisms such as bridging flocculation should be responsible for the aggregation. That is bridging occurs when biopolymer extends to the solution for a distance larger than the distance over which the inter-particle repulsion can act, and the BFs will tend to absorb onto particles to form large aggregates.[13]

For the coagulation with active pH adjustment, the optimal pH for turbidity removal with $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ was similar (7.0–7.5), while for the BFs from *Bacillus* sp. F6, the optimal pH was determined as 8.0 (Fig. 5). pH that impacts the flocculating activity of the BFs was reported to vary widely, ranging from 3.0 to 8.0,[21–24] and mechanisms of how pH affects the bio-flocculation are not clear. Additionally, it can be seen in Table 2 that increasing $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ dosage could cause an obvious accumulation of residual Al and Fe concentration in treated samples. Previous study reported that there was a visible chance that it may cause increased Al in treated water when using $Al_2(SO_4)_3 \cdot 14H_2O$ as coagulant, particularly, for the case of low-efficiency coagulation (i.e. dosage higher than γ_{DR} = 20 mg L^{-1}). In comparison, residual Al or Fe was apparently undetectable throughout a wide range of dosage for coagulating with BFs.



F i g . 5 – *Effects of pH on removal rate of turbidity from synthetic solution using different coagulants*

T a b l e 2 – *Residual metallic elements in treated solution for different dosage*

Dosage (γ_D/mg L^{-1})[a]	Residual Al or Fe in coagulated solution		
	$Al_2(SO_4)_3$ (γ_{Al}/mg L^{-1})	$Fe_2(SO_4)_3$ (γ_{Fe}/mg L^{-1})	BFs
8	0.2	–	–
10	2.1	3.2	–
20	4.1	5.0	–

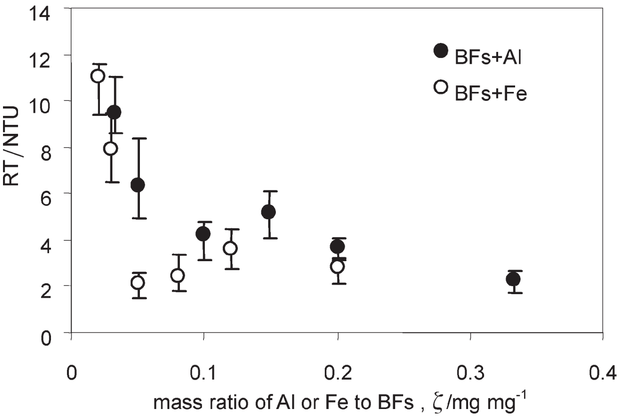
[a]Dosage of $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ is expressed as γ_{Al}/mg L^{-1} and γ_{Fe}/mg L^{-1}, respectively.

Although BFs has been demonstrated to be, in part, more advantageous than $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ for coagulation, it should be recognized that turbidity removal efficiency with BFs is at a level of 85 % on average, a value lower than 96 % that with $Al_2(SO_4)_3$ or $Fe_2(SO_4)_3$.

Combination of BFs with chemical agents for enhancing turbidity removal from synthetic solution

It has been known that turbidity removal can be enhanced by combined use of inorganic salts with polyelectrolyte serving as coagulant aid.[25] Accordingly, in order to enhance coagulation performance, coagulation of BFs in combination with $Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$ was studied and compared. As stated above, 6–20 mg L^{-1} of BFs resulted in average of 84 % turbidity removal (RT = 11.2 NTU); whereas combination of $Fe_2(SO_4)_3$ ($\zeta_{Fe/BF}$ = 0.05) or $Al_2(SO_4)_3$ ($\zeta_{Al/BF}$ = 0.33) with BFs achieved a RT as low as 2 NTU, accounting for 12 % increase of turbidity removal (i.e. 96 % in total) as shown in Fig. 6. Owing to an efficient coagulation, residual Fe or Al was almost undetectable in treated samples. $Fe_2(SO_4)_3$ was demonstrated to be more efficient in aiding coagulation than $Al_2(SO_4)_3$, due to the fact that when the lowest residual turbidity of 2 NTU was obtained, the amount of $Fe_2(SO_4)_3$ required ($\zeta_{Fe/BF}$ = 0.05) was much lower than that of $Al_2(SO_4)_3$ ($\zeta_{Al/BF}$ = 0.33) (Fig. 6). This indicates that a substantial increase of turbidity removal is possibly achieved by means of adding only a small amount of ferric salts to BFs.

It is obvious that the enhancement of coagulation performance obtained here is a consequence of the addition of Fe^{3+} ion into BFs. The role of trivalent Fe^{3+} is to increase the initial adsorption of biopolymers on suspended particles via decreasing the negative charge on both the polymer and the



F i g . 6 – *Effects of mass ratio of inorganic metallic coagulant addition (Al or Fe) to BFs (10 mg L^{-1}) on turbidity removal (pH 8.0; error bars×1bSD based on experiments conducted in triplicate)*

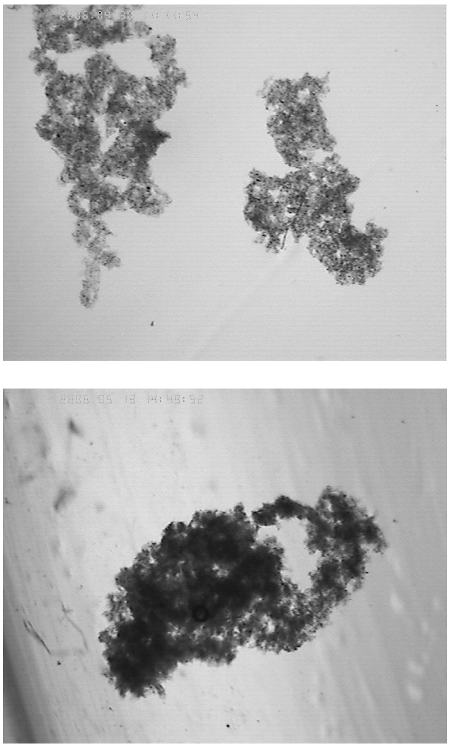


Fig. 7 – *Photographs taken of flocs formed during flocculation (left: individual 10 mg L^{-1} BFs and right: 10 mg L^{-1} BF+0.5 mg L^{-1}Fe salt)*

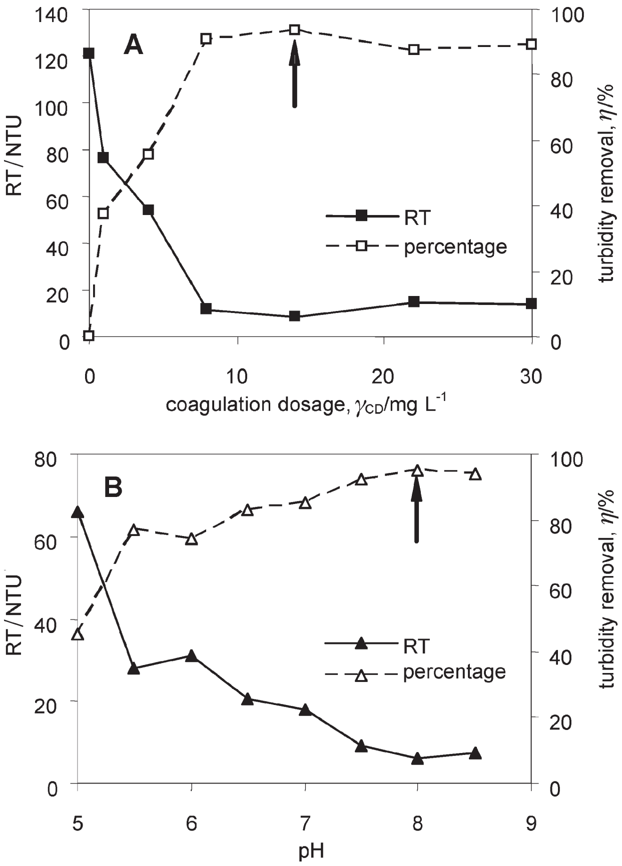


Fig. 8 – *Determination of optimum coagulant dosage (A) and pH (B) for removal rate of turbidity from raw water using Fe-base BFs ($\zeta_{Fe/BFs}$ = 0.05). The arrowheads indicate the optimal points.*

particles,[26,27] consequently, stimulating flocculating activity by neutralizing and stabilizing the residual negative charge of functional groups and by forming bridges between particles. On the other hand, it is also likely that Fe^{3+} and its hydrolysis products serve as crystal core to support the adsorption of biopolymer onto its surface so that more strong and dense flocs can be formed by bridging flocculation (Fig. 7 right). In contrast, the flocs formed based on single BFs are seen to lack crystal core, resulting in the aggregates with porous and permeable structure (Fig. 7 left). The effect of cations inducement on flocculating activity of BFs in coagulation treatment is not coincided,[21,28] depending on several factors such as bacterial strains, original mediums to be treated, type of cations as well as operating conditions.

Case Study: Coagulation of raw water applying BFs for turbidity removal

A case study was subsequently performed to examine turbidity removal from raw water (121 NTU; Songhua River) by the use of combination of $Fe_2(SO_4)_3$ and BFs ($\zeta_{Fe/BF}$ = 0.05). Experimental results showed that the turbidity could be decreased from initial 121 NTU to 8.3 NTU (removed by η = 93.2 %) at optimal coagulant dosage of γ_{BF} = 14.6 mg L^{-1} for BFs and γ_{Fe} = 0.9 mg L^{-1} for $Fe_2(SO_4)_3$ (Fig. 8A), based on which optimal pH for turbidity elimination (η = 94.6 %) was about 8.0 that appears similar to the results gained based on synthetic solution (Fig. 8B). It was noted that pH 7.8 of raw water was close to optimal pH 8.0, implying that maximum turbidity removal could possibly be achieved without active pH adjustment. Despite the same flocculants used, turbidity was being removed from raw water at a lower efficiency than that from synthetic solution, probably because of the negative effect of complex components present in raw water on coagulation with BFs.

Conclusions

Effective elimination of turbidity from kaolin clay solution and raw water could be achieved by using BFs from strain *Bacillus* sp. F6. Bridging flocculation other than charge neutralization appears to be the most likely mechanism governing the particle aggregation. The addition of a small amount of $Fe_2(SO_4)_3$ ($\zeta_{Fe/BF}$ = 0.05) or $Al_2(SO_4)_3$ ($\zeta_{Al/BF}$ = 0.33) as crystal core materials substantially increased turbidity removal efficiency up to η = 96 % or η = 95 %, primarily due to the combined function of charge neutralization and bridging, which promotes the formation of large and dense flocs. When BFs are applied, there is no observation of residual Fe or Al accumulation in treated solution throughout the whole experiments. Bioflocculant is shown here more advantageous in coagulation treatment for water purification for its health-related safety and biodegradable natures.

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List of abbreviations and symbols

BFs – bioflocculants
COD – chemical oxygen demand
RT – residual turbidity, NTU
γ – mass concentration, mg L^{-1}
ξ – zeta potential, mv
η – turbidity removal efficiency, %
d_p – diameter of particles, μm
d_s – diameter of colloidal substances, nm, μm
PACl – polyaluminum chloride
$\zeta_{Al/BF}$ – mass ratio of aluminum salts to bioflocculants
$\zeta_{Fe/BF}$ – mass ratio of ferric salts to bioflocculants

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