Environment-Friendly Approach toward the Treatment of Raw Agricultural Wastewater and River Water via Flocculation Using Chitosan and Bean Straw Flour as Bioflocculants

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ABSTRACT: Currently, there is a growing concern regarding water remediation from agricultural and domestic wastewaters. Among water treatment methods, flocculation is a widely used approach. In this study, the bioflocculation of wastewaters from Sinaloa (Mexico) was examined using two bioflocculants: chitosan and bean straw flour (BSF). The jar-test results showed that chitosan exhibited high effectiveness in pollutant removal from different sampling zones (agricultural wastewater and river water). Additionally, this bioflocculant reduced remarkably the concentration of Mn and Fe. On the other hand, BSF showed high effectiveness in pollutant removal for a specific type of wastewater, being highly competitive as compared to chitosan. Besides, BSF led to 40% of Mn removal from highly contaminated river water samples. For both biomaterials, bioflocculation was driven by charge neutralization and sweep flocculation mechanisms. For a given agricultural wastewater sample, both bioflocculants performed better than the commercial poly(aluminum chloride) for pH regulation and Fe removal.

INTRODUCTION

Agricultural wastewater is one of the main problems that affect biodiversity in different regions worldwide. Agricultural effluents are originated by the excess of water employed in irrigation systems. These raw waters contain residual fertilizers and pesticides which are hazardous pollutants that eventually contaminate natural water sources such as rivers used for drinking water production. Besides, some of these pollutants are present for many years (e.g. organochlorine pesticides). Consequently, residual agrochemicals change the chemical environment of the water bodies, causing a disruption of the ecological balance and representing a potential risk for human health. As an example, Sinaloa (Mexico) is a state with high agricultural and fishing activities, where the coastal pollution plays a significant role triggering a decrease in the shrimp production. In the Culiacan Valley of Sinaloa, the presence of organochlorine and organophosphorus pesticides and polychlorobiphenyls has been determined in the sediments of the agricultural drainage system. Also in the same region, Culiacan and Presidio Rivers have registered fluctuations in their chemical environment owing to the release of urban sewage and agricultural wastes. Additionally, organochlorine pesticides have been identified in the gonad, brain, and blood of mice from two agricultural areas of Sinaloa, which suggested that the variety of organochlorine pesticides in direct or indirect contact with nontarget organisms could deteriorate the health of animals and humans due to toxic effects and accumulation. Therefore, the treatment of effluents associated to agricultural activity is clearly required.

As it is well known, water remediation is one of the main issues around the planet given that the worldwide demand of clean water and the environmental balance must be carefully sustained. At the industry scale, flocculation is a major process allowing the removal of suspended solids from wastewaters, water clarification, and also decontamination of wastewaters. Flocculation is mainly induced by polymeric organic-based flocculants such as polyacrylamide-based products from petroleum-based resources. However, the use of synthetic flocculants causes serious environmental and health problems, as well as it generates controversy related to the production of large volumes of toxic sludge and the dispersion of acrylamide oligomers, being a health hazard due to its carcinogenic and neurotoxic consequences. For these reasons, alternative natural materials referred as bioflocculants have been developed for wastewater treatment. This eco-friendly approach involves biodegradability and sustainability with the exploitation of biodegradability and sustainability with the exploitation of...
byproducts obtained from human activities such as fishing, agriculture, industry, and others. Examples of bioflocculants include alginate, cellulosic materials, starch, chitosan, xanthan gum, Moringa oleifera, okra, guar gum and Cassia tora gum, and tannins, which represent economically viable and safe substitutes of synthetic flocculants.\textsuperscript{14,15} The biopolymer chitosan, obtained from the second most abundant natural polysaccharide called chitin (Figure 1), deserves particular attention in water and wastewater treatment. This amino-polysaccharide is nontoxic, biocompatible, and biodegradable and presents outstanding performances in bioflocculation- and sorption-oriented processes. Chitosan is also interesting in direct flocculation because it combines the two functions of coagulation and flocculation in industrial wastewater treatment. It can neutralize the negative charges and also bridge the aggregate of destabilized particles.\textsuperscript{16} In the end of the 2000s, Crini’s group proposed direct bioflocculation using low-cost chitosan as a novel eco-friendly approach to treat wastewater from pulp and paper plant to treat dye molecules.\textsuperscript{16} Bioflocculant chitosan acted both by polymer bridging and charge neutralization and sorption, enhancing the formation of larger flocs. Chitosan has also drawn particular attention for the removal of permethrin, an organochlorine pesticide. The results showed that the performance was higher with the increase of adsorbent dosage up to a specific value; then further adsorbent concentrations did not affect significantly the removal percentage.\textsuperscript{17,18} In another work, the removal of commercial pyrethroid and dithiocarbamate pesticides from model wastewaters has been assessed, where results (removal efficiency around 90\%) suggested similar interactions between the biopolymer and different target molecules. For that, the charge neutralization was found to be the dominant mechanism during the flocculation process.\textsuperscript{19} Chitosan can be also used for the removal of metals and particles such as colloids and dissolved organic matter. With raw samples of river water and agricultural wastewater, a high efficiency of chitosan in turbidity removal (TR) has been reached in batch trials. In this case, an optimal dosage of chitosan was required for decreasing turbidity in both water types, whereas the flocculation mechanism was dependent on the water composition, being in agreement with adsorption charge neutralization for river water and sweep flocculation for wastewater.\textsuperscript{20}

On the other hand, agricultural byproducts involving bean plant residues could be proposed as a low-cost, renewable, and sustainable cellulosic material with potential application in water remediation. As an example, mung bean husk (Vigna mungo) has been tested as a sorbent for the removal of Cd\textsuperscript{21} from aqueous solutions.\textsuperscript{21} The authors reported that the sorption process was dependent on experimental conditions, for example, pH, and the maximum removal of Cd\textsuperscript{21} was recorded as 35.41 mg g$^{-1}$ (at pH 5). They concluded that this eco-friendly and low-cost material could be efficiently used as a sorbent for metal removal.\textsuperscript{21} Another study performed an optimization of batch and dynamic flow conditions for Sb\textsuperscript{3+} sorption using green bean husk (Vigna radiata). The sorption results also demonstrated that the metal removal was affected by process parameters such as pH, sorbate and sorbent concentration, and contact time. The maximum Sb\textsuperscript{3+} sorption capacity apparently was 20.14 mg g$^{-1}$ (at pH 4, 25 °C and 60 min).\textsuperscript{22} Another example is the sorption of ranitidine hydrochloride onto a biochar-based material. The process has been evaluated in a fixed bed column employing superheated steam-activated biochar derived from bean husk. The result showed that the highest adsorptive capacity of the sorbent was 12 mg g$^{-1}$.\textsuperscript{23} In all these examples, bioproducts have been used as a sorbent but they could be also applied as eco-friendly bioflocculants.

According to the current literature regarding direct bioflocculation, additional exploration using eco-friendly flocculants for the remediation of raw agricultural wastewater is required. In this study, the performances of chitosan and bean straw flour (BSF) (Phaseolus vulgaris L.) for the treatment of raw agricultural wastewater (from the state of Sinaloa, Mexico) and urban river water (source for drinking water production) were evaluated using a direct flocculation process. In order to carry out this task effectively, a set of flocculation jar-tests was conducted under different conditions.

### RESULTS AND DISCUSSION

Wastewater in Contact with Humans and Animals in Sinaloa. In the last decades, water remediation has attracted growing interest within the scientific community given that the water demand and the ecological balance are in risk and must be carefully sustained. Despite the use of new technologies and the expanded role of federal governments toward the environmental protection, there is still an important concern owing to the widespread problem of water pollution. As an example, Figure 2 shows some zones of the state of Sinaloa (Mexico) where wastewater samples for this study were taken. (A) Campo Cinco y Medio (CCM) (Culiacan): the arrival of dairy shed effluents and agricultural wastewater (A-1), domestic wastewater arrival (A-2), and the sampling point of the resultant mixed water (A-3); unfortunately, people of the village, surrounding towns, and ecosystems are vulnerable to this hazardous effluent with milky appearance. (B) El Salado (ES) (Elota): the sampling point involving agricultural wastewater derived of conventional maize and tomato crops (B-1) and water supply used directly for domestic purposes (B-2). In this last case, agricultural wastewater flows close to the domestic water source of Salado village (~40 m); in addition, this sewage is subsequently deposited on the Elota River, and thus their contaminants could be eventually reaching the Pacific Ocean. Therefore, sewage should be treated in order to remove toxic substances such as the 32 compounds detected at concentrations between 0.03 and 1294 ng g$^{-1}$ dry weight in agricultural drainage systems of Culiacan Valley. From analyses, the chemicals registered were as follows: organochlorine pesticides, organophosphorous pesticides, and polychlorobiphenyls, as well as permethrin, triadimefon, and...
fipronil, from which 5 pesticides were found to be above the permissible concentration.9

Zeta Potential of Dispersions Containing Chitosan or BSF. It is known that molecular and electrostatic interactions among coagulant—floculant and pollutants depend on solution pH owing to the variations in particle charge. Hence, the electrophoretic phenomenon in aqueous systems containing chitosan or BSF was studied before Jar-test experiments. For that, measurements of zeta potential (ζ) were performed at different pH values by adjusting with NaOH (0.1 M) and HCl (0.1 M). As it can be seen in Figure 3, the high stability of chitosan solutions having pH < 6 was demonstrated with ζ data higher than 20 mV, which is obviously related to the protonation of the amino group (pKₐ near to 6.4)19 yielding positively charged polymeric chains. The isoelectric point (ζ = 0) of chitosan chains was between pH 7 and 8, and the ζ profile was similar to that previously reported in the literature.24 On the other hand, BSF particles were negatively charged at pH between 5 and 9, whereas that ζ close to zero was obtained at pH 4. The ζ profile of BSF was found to be in accordance with that observed by researchers exploring cellulose dispersions;25 evidently, cellulose is the main component of cell walls in bean plants. From ζ measurements for chitosan at pH within 6.5—8.5 and BSF at pH < 6, it may be initially supposed that flocculation is mainly triggered by nonelectrostatic interactions such as hydrogen bond, hydrophobic—hydrophilic balance, and van der Waals forces; at these pH values, both floculants have ζ close to zero. However, the flocculation can be influenced by different factors related to the chemical nature of the floculants and the field conditions of the samples.

Pollutants Removal from Wastewater Samples. Chitin (source of chitosan) and BSF can be obtained as byproducts derived from industrial operations in Sinaloa, and eventually these materials could be used for water remediation in this state and other regions. Thus, flocculation trials were performed for water samples having the following average values of some parameters: CCM: turbidity ≈ 160 NTU, pH ≈ 6.33, total dissolved solids (TDS) ≈ 1175 mg L⁻¹; La Escalera (LE): turbidity ≈ 50 NTU, pH ≈ 7.48, TDS ≈ 292 mg L⁻¹; and ES: turbidity ≈ 22 NTU, pH ≈ 7.63, TDS ≈ 1015 mg L⁻¹. Evidently, high values of some parameters are a consequence of alarming levels of different contaminants. The first test consisted of analyzing the turbidity evolution at different floculant dosages (Figure 4). The effectiveness in removing turbidity from CCM samples was highly dependent on the floculant dosage. At 10 mg L⁻¹ of the floculant, chitosan and BSF decreased turbidity effectively; then the residual turbidity was in a close range using 30 mg L⁻¹ of BSF (10 NTU) and 5 mg L⁻¹ of chitosan (6 NTU). With LE and ES samples, chitosan also led to very low turbidity levels (near to 5 NTU) after the treatment, and the optimal dose of the floculant was different depending on the initial turbidity and sampling zone. Apparently, the higher the initial turbidity, the lower the chitosan dosage that is required. For CCM and LE profiles, the residual turbidity raised slightly with increasing chitosan concentration after a given dosage; namely, an adequate number of polymer chains are required to be attached to the particle’s surface, resulting in an efficient flocculation process. On the other side, BSF reduced the turbidity of LE samples by around 50%, while the change in the appearance of ES samples

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Figure 2. Sampling zones based on wastewater: (A) CCM [(A-1) arrival of dairy shed effluents and agricultural wastewater; (A-2) domestic wastewater arrival; and (A-3) sampling point of the resultant mixed water]. (B) ES [(B-1) sampling point involving agricultural wastewater and (B-2) supply system of water used directly for domestic purposes]. Photos were taken and edited by the authors.

Figure 3. Zeta potential (ζ) profile at different pH values for dispersions containing chitosan or BSF.

Figure 4. TR from wastewater samples using chitosan and BSF at different dosage. Sampling zones: (A) CCM (160 NTU), (B) LE (50 NTU), and (C) ES (22 NTU).
was barely visible using this agricultural byproduct. The surprising result for the system CCM water and BSF could be related to the collaboration of some specific pollutants and the acid environment of CCM samples. Therefore, additional experiments focused on water remediation using CCM samples with quite different turbidity levels are presented below. On the other hand, it is relevant to indicate that flocculation tests using corn straw flour were performed. However, this material did not achieve good performance; thus the turbidity of samples remained unchanged. Hence, BSF contains some chemical entities that play an important role in the flocculation process.

In order to explore the flocculation mechanism, a study of the floc formation, solid redispersion (breakage) and reformation was conducted, finding that floc was re-formed after the desired flocculation time regardless of the biolocculant type. Based on TR profiles and reformation experiments using CCM and LE samples, the flocculation mechanism of adsorption charge neutralization and sweep flocculation were assumed for chitosan and BSF, respectively. For ES samples, sweep flocculation could be the predominant mechanism with both chitosan and BSF. In order to drive toward a better understanding, some specifications regarding both flocculation mechanisms are remarked. In the case of adsorption charge neutralization (colloidal particles are neutralized and attracted), pollutant removal decreases when an optimum dosage is used, and a complete reformation of flocs after breakage is observed. For sweep flocculation (particles are swept out forming an amorphous precipitate), particle removal improves with the progressive increase in the concentration of the flocculant, and weak flocs with reformation partially reversible is obtained.

The evaluation of turbidity reduction from wastewater (LE, 50 NTU) at different times was carried out by comparing the optimal dosage of bioflocculants and commercial poly-(aluminium chloride) (PAC) (Figure 5). PAC is one of the most frequent flocculants used in industry. Its popularity arises not only from its effectiveness as a flocculant but also from its ready availability and low cost. However, it has drawbacks such as water pollution by Al (under certain conditions) and production of large amounts of toxic sludge. In this work, the percentage of TR was determined based on the following calculation: TR (%) = \((T_0 - T)/T_0\)(100), where \(T_0\) and \(T\) are the initial turbidity and the remaining turbidity at a given time, respectively. Considering the maximal removal from each curve, changes of TR were registered having a similar trend and different TR with the time evolution (similar profile). At the first 20 min, all trials showed similar TR; however, a significant difference was observed at 30 min, where commercial PAC allowed a high TR in contrast to both bioflocculants. From 60 to 80 min, TR using chitosan increased gradually up to values close to that using PAC, whilst the BSF exhibited a maximal removal near to 55%.

The flocculation behavior in systems involving wastewater from CCM was further investigated because TR from these samples was remarkably well performed with both bioflocculants (chitosan and BSF). It should be kept in mind that CCM effluent was apparently the most contaminated of the sampling zones owing to the presence of waste from dairy shed and agricultural and domestic activities. Thus, high turbidity levels were registered because of the uncontrolled discharge of pollutants. The Jar-test was carried out using samples with different initial turbidity (160 and 84 NTU) depending on the daily discharge over the main drainage. In order to explore the flocculation behavior at lower turbidity, samples having turbidity of 57 NTU were prepared as dilution from the sample of 160 NTU. Both chitosan and BSF reduced the sample turbidity to values within the range 6–12 NTU, with chitosan being slightly more outstanding. As an example, images of some flocculation tests are shown in Figures 6 and 7.

Figure 5. Settling rate in agricultural wastewater (LE, 50 NTU) by comparing BSF (30 mg L\(^{-1}\)), chitosan (5 mg L\(^{-1}\)), and commercial PAC (5 mg L\(^{-1}\)).

As it can be seen, the opaque appearance of hazardous raw samples turned clear after the treatment irrespective of the initial turbidity and the bioflocculant. In all cases, an efficient separation was attained, where chitosan led to an improved sediment consolidation as compared to BSF, namely, BSF formed flocs containing higher percentage of adsorbed water. This result was expected for chitosan; nevertheless, it was conveniently surprising for the byproduct BSF used in the direct flocculation. For CCM samples, additional contaminants could be helping in the flocculation process when BSF is
anions reported as PO₄³⁻, tried to reduce turbidity exploring wastewater samples from CCM is notable. Results of CCM samples. Based on the analyses performed, the lower phosphates in wastewater, treatments quality of water. This phenomenon is known as eutrophication. In order to remove phosphates from wastewater, treatments employed, while this effect was less marked for other wastewater samples (LE samples: 50 NTU; maximal removal close to 55%); additionally, the control sample (with no biofloculant) remained highly turbid even after 1 h. This means that the high performance of BSF is mainly related to the chemical composition of CCM samples. Given that handling and analyses of this type of raw wastewater represent a risk for human health, only a set of experiments was performed.

Table 1 summarizes the average values for several parameters of raw and treated wastewater related to ES and CCM samples. Based on the analyses performed, the lower water quality for samples from CCM is notable. Results of turbidity reduction exploring wastewater were discussed earlier. In the case of ion removal, the concentrations of anions reported as PO₄³⁻, SO₄²⁻, and NO₃⁻ were recorded for raw water and after the treatment using chitosan and BSF in the Jar-test. The content of phosphates in water has to be carefully monitored; large quantities can stimulate the growth of aquatic micro- and macro-organisms negatively affecting the quality of water. This phenomenon is known as eutrophication. In order to remove phosphates from wastewater, treatments such as adsorption, anion exchange, and biological methods have been proposed. Unlike phosphorus, nitrogen is commonly accepted to be relevant only in marine eutrophication. Therefore, measurements of total nitrogen content of water after flocculation were omitted in the present research. However, monitoring nitrite concentrations is also essential for ecosystem preservation; then it has been demonstrated that these ions affect adversely the fish and shellfish growth, water balance, blood oxygen carrying capacity, osmoregulation, and so on. In the present contribution, variations of the PO₄³⁻ content were negligible, as reported previously by other authors. According to the pH of samples and zeta potential profiles of floculants, the surface of the floculants are charged (Chitosan positively; BSF negatively) and given that the HPO₄²⁻ (predominant form of phosphate ions at the pH of the treated samples) is also with negative charge, the adsorption of phosphates should be favored by electrostatic attraction when chitosan is used. However, the global chemical environment in these raw samples could complicate the phosphate level reduction; thus, it is a challenging task to find structure-property-function relationships toward flocculation in field conditions. For sulfates, the removal mechanism is very similar to the removal mechanism of phosphates; nonetheless, the adsorption site may present selectivity for one of them. In our research, the biofloculants employed were unable to remove phosphates, sulfates, and nitrates, indicating the absence of adequate adsorption sites for these ions. Although, a decrease in the concentration of SO₄²⁻ was detected (from 225 to 175 mg L⁻¹) using BSF for the treatment of samples from CCM. Turunen et al. also used chitosan for the removal of phosphorous from agricultural wastes with different levels of phosphorus and turbidity, but the phosphorous concentration was reduced only in high polluted agricultural wastewater. Similar to our results, these authors concluded that the biofloculants are more effective in high polluted water than in less polluted water.

On the other hand, chitosan performed well in removing Fe in both types of samples (ES and CCM). Metal ions were notably trapped by the two biofloculants, and the pH of the samples was also regulated after the flocculation (close to 7). The commercial flocculant PAC exhibited lower effectiveness in both tasks. In contrast with Chitosan and BSF, it is clear that metal removal by binding to available sites is less expected when PAC is the flocculant because of their differences as chelating agents. This result highlighted that chitosan and BSF are promising materials for wastewater treatment. A detailed discussion about interactions between metal ions and floculants is subsequently presented using river water samples. According to this study, it is recommended that farm industries should perform a treatment of their effluents before this hazardous water is released into the environment. Thus, subsequent pollution of natural resources, which still being contaminated by residual chemicals, could be avoided. To this aim, authors consider that this contribution provide a treatment strategy via direct bioflocculation that can be easily adapted to the reality of these industries.

Table 1. Quality Parameters of Raw and Treated Wastewater Related to Different Sampling Zones

| Sampling Zone | Flocculant | Turbidity (NTU) | TDS (mg L⁻¹) | pH | PO₄³⁻ (mg L⁻¹) | SO₄²⁻ (mg L⁻¹) | NO₃⁻ (mg L⁻¹) | Fe (mg L⁻¹) |
|---------------|------------|----------------|--------------|----|----------------|----------------|---------------|-------------|
| ES raw water  | chitosan   | 22             | 1015         | 7.63| 1.00           | 90             | 6             | 0.185       |
|               | BSF        | 16             | 1015         | 7.22| 1.00           | 90             | 6             | 0.182       |
| CCM raw water | chitosan   | 160            | 1175         | 6.33| 1.00           | 225            | 10            | 0.049       |
|               | BSF        | 10             | 1181         | 7.17| 1.10           | 175            | 10            | <0.010      |
|               | PAC        | 4              | 1180         | 6.52| 1.10           | 200            | 10            | 0.027       |
Turbidity and Ion Removal from River Water Samples. Trials using urban river water were carried out in order to evaluate the flocculation behavior in the absence of agricultural wastes. Figure 8 displays the plots of TR from Humaya River (29 NTU) and Tamazula River (32 NTU).

Table 2 shows average values for several parameters of raw and treated urban river water. Results of TR from this type of water were discussed earlier. In the case of Humaya River, the pH of raw samples (pH = 8.39) was barely within the range established by NOM-127-SSA1-1194 (pH from 6.5 to 8.5), whilst the Fe content in Tamazula River (0.238 mg L−1) was detected close to the permissible limit (0.300 mg L−1). It can be seen that after pollutants removal, the pH of both samples was regulated to a more neutral value (close to 7) irrespective of the bioflocculant type. Regarding anion removal, concentrations reported as PO₄³⁻ and SO₄²⁻ remained without significant variation after the flocculation process using chitosan and BSF; a similar behavior was previously discussed for wastewater samples. Furthermore, according to our monitoring of raw water, the concentrations of these anions were significantly lower in river water. For metal ions, the entrapment of Fe and Mn in both types of samples (Tamazula and Humaya rivers) was assessed. The good performance of chitosan for the metal ion removal is attributed to its functional groups, given that the mechanism involved during the removal of Fe and Mn with chitosan is through chelation or complex formation. Despite chelation being the predominant process for the removal of these metals ions by forming coordinate covalent bonds due to the amino groups, some hydroxyl groups may participate in coordination by the release of protons. Hence, BSF mainly removes Fe or Mn by coordination with the hydroxyl groups, given that the mechanism involved during the removal of Fe and Mn with BSF; a similar behavior was previously discussed for wastewater samples. Furthermore, according to our monitoring of raw water, the concentrations of these anions were significantly lower in river water. For metal ions, the entrapment of Fe and Mn in both types of samples (Tamazula and Humaya rivers) was assessed. The good performance of chitosan for the metal ion removal is attributed to its functional groups, given that the mechanism involved during the removal of Fe and Mn with chitosan is through chelation or complex formation. Despite chelation being the predominant process for the removal of these metals ions by forming coordinate covalent bonds due to the amino groups, some hydroxyl groups may participate in coordination by the release of protons. Hence, BSF mainly removes Fe or Mn by coordination with the hydroxyl groups. This result highlighted that chitosan is a promising material for turbidity and metal removal during the production of potable water. Specially, it is mandatory to have low concentration of hazardous metals, which are very common contaminants of water. In the case of Fe and Mn, they can be leached out from their bearing rocks to the water or be discharged to the water from industrial activity; an excess of Fe and Mn can cause some inconvenience like giving metallic taste to drinking water, and these metals also tend to affect the health of the human body because of their gradual accumulation. In the case of Mn, it was previously reported that the concentration of this ion in Humaya River is eventually higher than the levels established in Mexican laws, being a problem in potabilization plants of Culiacan (Mexico) during the production of colorless water, and the population demand of a higher quality service. However, the concentration of detected Mn was within the range of permissible limits. Hence, some trials were carried out using low turbidity river water (Tamazula River, pH close to 7.5) as a matrix for the preparation of synthetic samples with a higher content of Mn.
by adding manganese salt, obtaining the following concentration: (A) 0.120, (B) 0.170, (C) 0.280, (D) 0.374, and (E) 0.424 mg L\(^{-1}\). From Figure 10, it can be observed that the effectiveness of BSF in Mn removal was improved as the metal concentration increased; while chitosan resulted to be an efficient material that triggered a flocculation process involving a higher metal concentration removal. Chitosan resulted in river water with Mn content within the levels required by Mexican laws (<0.15 mg L\(^{-1}\)). The performance varied depending mainly on the bioflocculant nature, which could be related to the preference of this metal for complex formation (Mn\(^{2+}\) prefers six-coordinated complex structures).\(^{38}\) Hence, the interaction via the coordinate bond was apparently more suitable between metal ions and chitosan. This polysaccharide contains both amino and hydroxyl groups that can bind to metal ions, whilst BSF comprises proteins, lipids, and carbohydrates, among other components, resulting in diverse functional groups. Besides, it was previously assumed that chelation is the predominant process for metal removal, with chitosan being a better chelating agent because of its particular intrinsic properties.

On the other hand, pH is an important factor in the sorption procedure because it influences the capability of the Mn\(^{2+}\) ions to dissolve, the amount of adsorption of the adsorbate and also the quantity of counter-ions on the adsorbent that contains the functional groups. Commonly, the Mn removal is reduced at lower pH due to the competition between protons (H\(^+\)) and Mn\(^{2+}\) for the adsorption site.\(^{37}\) When the pH increases the Mn removal increases reaching a maximum value at pH 7; at greater pH values, the Mn removal decreases. Thus, the matrix from Tamazula River presented appropriate pH (between 7 and 8) for Mn removal. In addition, from zeta potential profiles of chitosan (at pH between 6.5 and 8.5) and BSF (at pH < 6), it was supposed that both materials should exhibit non-electrostatic interactions with pollutants because of ζ close to zero. Furthermore, experiments using samples based on the above-mentioned water matrix and copper salt were conducted. For that, synthetic samples having concentrations of 1.35 and 2.75 mg L\(^{-1}\) of Cu were treated. BSF exhibits a low efficiency in copper entrapment, whereas chitosan allowed the removal of around 50% of this metal from both samples. Based on calculations,\(^{38}\) the effectiveness in Cu removal is affected by the ligand nature, and the formation of octahedral complexes is preferred when the metallic center is the ion Cu\(^{2+}\). According to the results with samples containing 2.75 mg L\(^{-1}\) of Cu, chitosan can help in the production of potable water with copper content within the levels required by Mexican laws (<2.0 mg L\(^{-1}\)).

### CONCLUSIONS

Chitosan and BSF represent eco-friendly materials, which could be used in wastewater treatment plants via direct flocculation. This study demonstrated that chitosan exhibited high effectiveness in the removal of different pollutants regardless the water composition (types of wastewater and river water) in field conditions. Our results also showed that the flocculation mechanism, being adsorption charge neutralization or sweep flocculation, was dependent on the initial water quality. The use of this bioflocculant resulted in turbidity levels and heavy metal concentrations within the limits established by the Mexican Environmental Regulation (NOM-127-SSA1-1194). On the other hand, the proposed byproduct BSF, renewable and sustainable material, exhibited a high effectiveness in direct flocculation using hazardous samples (CCM). For these samples, the performance of BSF was remarkably competitive with the results reached using chitosan, and both bioflocculants were notably better than the commercial PAC for pH regulation and Fe removal. Additionally for synthetic samples, BSF was able to trap a high amount of Mn (close to 40%) from river water matrix when the Mn content was 0.424 mg L\(^{-1}\). Irrespective of the water type, BSF triggered a mechanism of sweep flocculation. Based on complementary trials, flocculation using the byproduct corn straw flour was not successful. Hence, BSF contains some chemical entities that play a key role during the flocculation process. Although chitosan performed better for water remediation, the price involved using chitosan is higher than the cost with BSF. Besides, future chemical modification of BSF could enhance its flocculation performance in a variety of samples. On the other hand, agricultural wastes as bioflocculants represent a potential solution that the farmers should apply in their own wastewater in order to mitigate contamination before their wastes reach rivers and lakes. Thus, this contribution provides relevant data to be exploited in environmental technologies and industrial sustainability.

| urban river | flocculant | turbidity (NTU) | TDS (mg L\(^{-1}\)) | pH | PO\(_4^{3-}\) (mg L\(^{-1}\)) | SO\(_4^{2-}\) (mg L\(^{-1}\)) | Fe (mg L\(^{-1}\)) | Mn (mg L\(^{-1}\)) |
|-------------|------------|----------------|---------------------|-----|---------------------|---------------------|----------------|----------------|
| Tamazula    | raw water  | 32             | 129                 | 7.60 | 0.45                | 30                  | 0.238          | 0.095          |
|             | chitosan   | 3              | 130                 | 7.35 | 0.50                | 25                  | 0.052          | 0.010          |
|             | BSF (30 mg L\(^{-1}\)) | 18         | 132                 | 7.30 | 0.55                | 30                  | 0.216          | 0.079          |
| Humaya      | raw water  | 29             | 80                  | 8.39 | 0.49                | 15                  | 0.190          | 0.080          |
|             | chitosan   | 7              | 81                  | 7.32 | 0.49                | 20                  | 0.049          | 0.015          |
|             | BSF (30 mg L\(^{-1}\)) | 21         | 82                  | 7.33 | 0.40                | 15                  | 0.200          | 0.079          |

![Figure 10. Manganese removal from synthetic samples using river water matrix (15 NTU). Flocculant concentrations: chitosan (5 mg L\(^{-1}\)) and BSF (30 mg L\(^{-1}\)). Manganese concentrations: (A) 0.120, (B) 0.170, (C) 0.280, (D) 0.374, and (E) 0.424 mg L\(^{-1}\).](image-url)
EXPERIMENTAL SECTION

Materials. Chitosan with a medium molecular weight was purchased from Sigma-Aldrich (Mexico). Chitin origin: shrimp shells; form: powder; molecular weight: 190–310 kDa; degree of deacetylation: 75–85%. Chitosan was used from aqueous solution (1% acetic acid) at 5000 mg L⁻¹. The BSF was prepared from the bean straw (P. vulgaris L.), an agricultural byproduct, which was collected directly from the farm after bean harvest in Sinaloa, Mexico. The straw was triturated for size reduction and sieved through both 250 and 425 μm pore sieves, and chemical modification using hazardous reagents was avoided. The proximate composition of the flour comprised ash (9%), protein (3%), lipids (0.2%), carbohydrate (47%), crude fiber (36%), and moisture (4.8%). PAC, commercial grade, was provided by the Water Supply and Sewerage Board of Culiacan, Mexico, and used as received. The characteristics of the PAC are yellow powder; Al₂O₃ close to 30%; density of 1.2 (20 °C); pH within the range 3.5–5.0.

Agricultural wastewater samples were taken from CCM (24°34’4.78” N, 107°26’39.36” W), LE (25°26’51.8” N 108°11’15.0” W), and ES (23°54’03.9” N, 106°49’59.2” W) in Sinaloa, Mexico. Wastes from CCM drainage are a mixture composed of dairy shed effluents, agricultural wastewater, and domestic wastewater, whilst effluents from LE and ES are derived of agricultural activity. River water samples were taken from Humaya River (24°49’3.58” N, 107°24’15.54” W), Tamazula River (24°48’57.32” N, 107°23’9.13” W), and Culiacan River (24°48’35.33” N, 107°24’25.3” W), which are urban rivers used as supply sources for water potabilization plants in Sinaloa, Mexico.

Jar-Test. All experiments were performed by using a PB 700 Standard Jar-Floc Tester with six mixers (Phipps and Bird) at 25 °C. The procedure was followed according to the literature.  Briefly, glass beakers were filled with 500 mL of the water sample, and then an appropriated volume of flocculant–coagulant was added to each beaker followed by mixing thoroughly via mechanical stirring at 100 rpm for 5 min. Subsequently, the shaking speed was slowed at 60 rpm and maintained for 30 min; afterward, the mechanical stirring was stopped, and the sediment settlement and consolidation were studied for a prescribed time of 25 min. Samples were then collected in the upper part of the beaker to measure the various analytical parameters of the effluent. The pH of the solution was not adjusted prior to the addition of the flocculant. Flocs and the aqueous phase were separated through decantation. In this test, flocculant concentrations and flocculation time were studied.

Characterization. Dynamic light scattering was used to determine the zeta potential (ζ) of dispersed materials. Measurements were carried out at 25 °C using a Zetasizer (NanoZS) from Malvern Instruments (ZEN3690) equipped with a red laser (λ = 630 nm). Turbidity was measured using a Hach 2100N turbidimeter having a stable halogen-lamp with a red laser (λ = 630 nm). Turbidity was measured using a Hach 2100N turbidimeter having a stable halogen-lamp with a red laser (λ = 630 nm). Turbidity was measured using a Hach 2100N turbidimeter having a stable halogen-lamp with a red laser (λ = 630 nm). Turbidity was measured using a Hach 2100N turbidimeter having a stable halogen-lamp with a red laser (λ = 630 nm). Turbidity was measured using a Hach 2100N turbidimeter having a stable halogen-lamp with a red laser (λ = 630 nm).

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Notes
The authors declare no competing financial interest.

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