Review

A Review of the Application of Biofloccualnts in Wastewater Treatment

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Abstract

The rate of increase in industrialization and daily human activities has resulted in a tremendous increase in the amounts of waste and wastewater containing organic and inorganic pollutants discharged to the environment. Coagulation-flocculation technology has been widely employed in water/wastewater treatment as a convenient and reliable technique for removing colloids, particles, and cell debris. Organic and inorganic flocculants have also been used in fermentation industries, and in waste and water treatment due to its potential to flocculate efficiently at a minute dosage. However, their use has been restricted as a result of their low efficiency, associated health risks, and non-biodegradability. As a result, the health implication of chemical flocculants has necessitated environmentally friendly biodegradable bioflocculants in wastewater treatment in terms of dye, colour, solids, and turbidity removal. Industrial applications of some microorganisms implicated in bioflocculation have been established. However, the application of actinobacteria strains that have been isolated, screened, and confirmed for bioflocculation have yet to be validated. Hence, this paper reiterates that *Actinobacteria* have been implicated in flocculation and elaborates on the need for isolation and screening of novel *Actinomycetes* with better removal and cost efficiency in order to enhance their large- and medium-scale production and establish their industrial application.

Keywords: bioflocculants, coagulation-flocculation, actinobacteria, wastewater treatment, application

Introduction

Flocculation is an essential parameter employed for the removal of suspended solids in domestic and industrial wastewater treatment. Flocculation is accomplished with the help of flocculants, which are either natural or synthetic substances that facilitate the aggregation of particles to form flocs. Flocculation has attracted wide attention as a means of separation technique in portable, domestic, and industrial wastewater treatment plants. Flocculants have been employed in the recovery of suspended solutes from solution [1]. The use of flocculants in raw water treatment, surface treatment, industry, petroleum refinery effluent, and the paper industry has been well documented [2].

The coagulation-flocculation approach is widely used in water and wastewater treatments. Also, flocculation has been a known factor in the removal or separation of colloids and suspended particles of natural organic matter, and metal ions. Additional applications of flocculating agents

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include the elimination of toxic metals, anions, color, and odour in wastewater treatment. The use of chemically synthesized flocculating agents such as aluminum sulfate, polyaluminum chloride, and ferric chloride, and polymers like polyacrylamide derivatives have been frequently employed in wastewater treatment and fermentation industries as a result of their cost effectiveness and strong flocculating efficiency. However, the application of inorganic and organic flocculating agents has been drastically reduced due to the fact that large quantities are required to achieve effective flocculation. The commonly used organic flocculants are polyacrylamide, polyacrylic acid, poly (diallyl dimethyl ammonium chloride), and polyamine [3]. Flocculation efficiency is usually measured or quantified based on turbidity removal, chemical oxygen demand, and colour removal.

Bioflocculation is the process whereby stable aggregates are formed by extracellular polymers produced by living cells. Recently, the use of microbial flocculants has become a promising substitute for chemical flocculants because of their safety and biodegradability efficiency, which have put them in high demand [4].

Bioflocculants and metal ions have played an equally tremendous role in forming and settling sludge in anaerobic and aerobic treatment systems [5]. In addition, they can serve as an alternative to centrifugation and filtration for harvesting microbial cells from broth in the food and fermentation industries [6]. Since bioflocculants are generally biodegradable in nature, their use in wastewater treatment, downstream processing, and fermentation processes have increased [7]. The choice of adopting biotechnological methods for the production of bioflocculants solely depends on the possibility of using different microorganisms to synthesize extracellular substances with different compositions. In recent years, it has been documented that many microorganisms such as actinomycetes, fungi, bacteria, and algae have the ability to produce environmentally safe extracellular polymers that act as bioflocculants [8]. On the contrary, according to literature, researchers have proved the use of some chemically synthetic flocculant substances to be detrimental to aquatic life and the environment. For instance, acrylamide monomer, which is non-degradable, has been implicated in causing cancer and Alzheimer's disease [9]. Hence, there is a need to substitute or replace the exploration of synthetic flocculants with bioflocculants produced by microorganisms.

Also, an outstanding economic factor is that many developing countries can hardly afford the high costs of purchasing imported chemicals for water and wastewater treatment. This has therefore necessitated the exploration of new microorganisms that could produce flocculants with high flocculating activity and optimizing their fermentation processes in order to improve productivity. Bioflocculants have been used to treat waste and industrial water as both pure and mixed cultures. Microbial flocculants have also been implicated in the adsorption of heavy metals [10]. Also, the bioflocculant produced by Serratia ficaria was able to remove COD and turbidity at efficiencies of 64.1-80.7% and 91.8-93.7%, respectively, in agricultural wastewater [11]. In addition, bioflocculant MMF1 was reported by [12] in removing COD in indigotin printing and dyeing wastewater.

On the other hand, turbidity and COD removal from swine wastewater were achieved at flocculating efficiencies of 91% and 42%, respectively, when treated with bioflocculant (xn11 + xn7) [13]. The biofloculant produced by the mixed culture of *Halobacillus sp.* mvuyo and *Oceanobacillus sp.* pinky exhibited a significant efficiency when tested on waste and river waters [14]. Their findings confirmed that the consortium of the mixed bioflocculant eliminates COD and turbidity in

Application	Microorganism	Remarks	Reference
Biomass recovery and cell removal	Paenibacillus polymyxa	Removed Scenedesmus sp.	[1]
	Solibacillus silvestris	Removed Nannochloropsis oceanica	[17]
	Klebsiella pneumoniae	Removed Acanthamoeba cysts	[18]
Water and wastewater treatment	Oceanobacillus and Halobacillus	Treated brewery, dairy wastewater, and river water	[14]
	Azotobacter indicus	Treated dairy, wool, starch, and sugar industries wastewater	[1]
	Cobetia sp. and Bacillus sp.	Treated river water, dairy, and brewery wastewater	[19]
Decolorization	Rhodococcus erythropolis	Remove disperse dye solutions	[20]
	Serratia ficaria	Decolourized pulp effluent	[10]
	Chryseomonas luteola	Decolourized dye wastewater	[21]
Mining and other applications	Rhodopseudomonas sphaeroides	Flocculated coal slurry	[22]
	Bacillus subtilis	Synthesis of Ag nanoparticles (60 nm)	[23]
	Halomonas Maura	Synthesis of mauran/chitosan nanopareticles (30-200 nm)	[24]

Table 1. Applications of Some Bioflocculants Producing Microorganisms.

brewery wastewater, dairy wastewater, and river water at efficiencies greater than 90% when compared with polyacrylamide and aluminum chloride, which are used as conventional flocculants. [14]. Based on literature, fungi, bacteria, and algae have been greatly implicated in flocculation and in treatment of waste and industrial waters [15, 16]. However, actinomycetes also have been employed in flocculation, but their production on a large scale medium and application in treating waste and industrial waters has yet to be established.

In this paper we reviewed *Actinobacteria*, which has been implicated in flocculation, and elaborate on the need for isolation and screening of novel *Actinomycetes* with better removal and cost efficiency that will exhibit the potential of treating both waste and industrial waters. Some of the reported microorganisms that have been implicated in flocculation and their industrial applications are summarized in Table 1.

Literature has affirmed that actinobacteria have been isolated, screened, and implicated in flocculation. However, their industrial applications have yet to be established [25, 26, 27, 28, 29, 30, 31]. The genera of Actinobacteria that has been implicated in flocculation are: the genus *Arthrobacter* [25, 26], the genus *Actinobacterium* [27], the genus *Brachybacterium* [28], the genus *Streptomyces* [29, 30], and the genus *Nocardiopsis* [31]. Their Culture conditions and flocculating efficiencies are summarized in Table 2.

Culture conditions have played an immense role in bioflocculant production. Researchers have confirmed that carbon and nitrogen source requirement varies with different isolates [32, 7]. Considering the table above, nitrogen and carbon sources required by each actinobacteria differ from each strain, and this parameter is one of the determining factors in bioflocculant production. Furthermore, researchers have confirmed that bioflocculant-producing bacteria thrive in a medium that contains organic carbon. For instance, [25] reported that *Arthrobacter* sp uses lactose, glucose, and urea as the best carbon and nitrogen substrates for optimum flocculating efficiency. Based on the table above, each actinobacterium has attained its optimum flocculating efficiency with different carbon and nitrogen substrates. Initial pH is also one of the major parameters that influence bioflocculant production and flocculating activity. Salehizadeh and Shojaosadati [7] reported that initial pH of medium depicts the electric charge of cells and oxidation reduction capability, which may hinder or support the adsorption of nutrients and enzymatic reactions in different organisms. Wang et al. [33] has reported that cations enhanced or facilitated initial adsorption of flocculants in suspended colloids/particles by a means of reducing or minimizing the negative charge on polymers and colloids.

Cations have aided flocculation in actinobacteria by neutralizing and stabilizing the negative charge of some functional groups, thereby resulting in bridge formation between colloids [7]. The addition of cations to suspension or particles stimulates the rate of floc formation, thereby resulting in sedimentation of particles.

Isolation of Actinobacteria on Cultivation Media

Actinomycetes have been isolated from both soil sediment and water samples using a serial dilution spread plate technique. These organisms thrive well on mostly selective media such as starch casein agar, glucose yeast malt extract agar, actinomycetes isolation media, soil extract agar, glycerol-asparagine agar, colloidal chitin, and M3 agar [34]. Antibiotics such as cyclohexamide, nalidixic acid, nystatin, rifampicin, and fluconazole are usually employed to supplement the media in order to inhibit the growth of other bacteria and fungi.

Identification of Actinomycetes

The concept of characterizing actinomycetes to genus level has been validated by [35] and Goodfellow [36] using a combination of cultural, morphological, and chemical properties. Conversely, characterization of these bacteria to specie level is more cumbersome. The availability of gene sequencing has revolutionized the taxonomy of the aerobic actinomycetes and has become an important yardstick for the identification and characterization of

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Isolate	Culture conditions	Component	F/A	References		
Arthrobacter sp Raats	<i>robacter</i> sp Raats Lactose, urea, glucose, Mg ²⁺ , and pH 7.0 Glycopro		84%	[25, 26]		
Actinobacterium sp Mayor	Actinobacterium sp Mayor Sodium carbonate, Ammonium, sulfate, urea, yeast extract, Ca ²⁺ , and pH 8.0 Polysaccharide		91%	[27]		
Brachybacterium sp	<i>Brachybacterium</i> sp Maltose, urea MgCl ₂ , and pH 7.2 Glycosaminoglycar		91.17%	[28]		
Streptomyces sp	Glucose, ammonium sulfate, MgCl ₂ , and pH 6.8	Proteoglycan	89%	[29]		
Nocardiopsis aegyptia sp. nov	Glucose, peptone, CaCl ₂ , and pH 7.0	Polysaccharide	89%	[30]		
Streptomyces grieseus	Yeast extract and pH 7.0	NA	NA	[31]		

Table 2. Culture Conditions and Flocculating Efficiencies of Actinobacteria Spp Implicated in Flocculation.

F/A: Flocculating activity

NA: Not applicable

clinical isolates. The 16S rRNA gene sequencing has been employed to identify diversity of actinomycetes, which corroborates the method of chemical and morphological taxonomy.

Actinobacteria

Actinomycetes are gram-positive, aerobic, and mycelial prokaryote organisms with a high guanine: cytosine ratio [37]. They are unicellular organisms and their means of reproduction is by special spores, fission, or conidia. Most are free-living saprophytic bacteria found widely distributed in soil and water [38]. They resemble fungi morphologically and bacteria physiologically [39].

They are related to true bacteria in terms of classification, but they are generally considered as higher filamentous bacteria [40]. Actinomycetes from the genus *Streptomyces* account for two thirds of antibiotic production while the genus *Micromonospora* takes second place. Vobis [41] reported that *Micromonospora* produced a wide range of broad-spectrum antibiotic substances. The genera *Micromonospora* and *Streptomyces* are widely and evenly distributed in aquatic ecosystems and are largely more abundant than other groups of actinomycetes [42].

The genera Actinomadura, Actinoplanes, Actinosynnema, Dactylosporangium, Kibdelosporangium, Kitasatosporia, Microbisporia, Micromonospotra, Nocardia, Saccharopolyspora, Streptoalloteichus, Streptosporangium, and Streptoverticillium have also being implicated in antibiotic production [43].

Literature has revealed that ecology of this bacterium is not well documented. Actinomycetes are found in many aquatic environments. They have been isolated from marine and freshwater bodies [44, 45,]. The most common actinomycetes isolated from freshwater environments include *Actinoplanes, Micromonospora, Rhodococcus, Streptomyces*, and *Thermoactinomyces* [46].

The Genus Streptomyces

This is the largest group of Actinobacteria and it belongs to the family Streptomycetaceae. [47]. Streptomycetaceae is a family of Actinobacteria, which constitute the monotypic suborder Streptomycineae. Streptomyces are grampositive, aerobic, spore-forming microorganism found mostly in soil sediment and marine environments. Goodfellow and Williams [48] reported that they possess highly branched substrate and aerial mycelium, spores on aerial mycelium, and occasionally on substrate medium. Currently, 600 species and 38 subspecies of Streptomyces bacteria have been described. The most unique attribute of Streptomyces is the ability to produce secondary metabolites and bioactive compounds that are important in human and veterinary medicine, and in agriculture [49]. The genus Streptomyces has served as a basic source of many antibiotics, namely streptomycin, and this was the first antibiotic produced against tuberculosis. They have been implicated in the production of natural-based

antibiotics, which includes cypemycin, grisemycin, bottro mycins, and chloramphenicol [50].

The genus Arthrobacter

The genus Arthrobacter belongs to the Micrococcaceae family. Based on the quinone system and peptidoglycan structure, the genus Arthrobacter can be subdivided into two groups. Arthrobacter is a bacteria usually found in soil, and some have been recovered from clinical specimens. They are non-sporulating, gram-positive, and obligate aerobes that show respiratory metabolism with the exception of Arthrobacter globiformis and Arthrobacter nicotianae, which are attributed with anaerobic metabolism [51]. Based on their unique metabolic diversity, Arthrobacter species have been explored in industrial applications in the treatment of contaminated wastewater [52]. Arthrobacter nitroguajacolicus has been reported as a strain with the ability to transform acrylonitrile into acrylic acid [53]. Arthrobacter crystallopoietes has been implicated in the reduction of hexavalent chromium in contaminated soil, indicating its biotechnological role in bioremediation [54].

The genus Nocardiopsis

The genus Nocardiopsis are aerobic, spore-forming actinomycetes that are known for the production of branched, vegetative mycelium and aerial hyphae. Species of the genus Nocardiopsis may be characterized based on the color of their mature aerial and substrate mycelia, their ability to break down different compounds, and their ability to utilize different carbon sources [55]. The genus Nocardiopsis has been currently divided into seven distinct species: Nocardiopsis alborubidus, Nocardiopsis albus, Nocardiopsis antarcticus, Nocardiopsis dassonvillei, Nocardiopsis halophila, Nocardiopsis listen, and Nocardiopsis lucentensis. Nocardiopsis albus includes two subspecies, Nocardiopsis albus subsp. albus and Nocardiopsis albus subsp. prasina. Nocardiopsis sp. has been reported to be prolific in the production of secondary metabolites [55, 56] and recommended to contribute immensely to the chemical defense mechanisms of their host against predators with biologically active compounds and biofouling [57].

The genus Brachybacterium

The genus *Brachybacterium* is a Gram-positive, coccoid-to-ovoid shaped, non-motile bacteria. This genus was first proposed by [58] to belong to the family *Dermabacteraceae*, located in the Actinobacteria class. The genus *Brachybacterium* encompasses 10 recognized species, namely: *Brachybacterium alimentarium*, *B. conglomeratum*, *B. faecium*, *B. fresconis*, *B. muris*, *B.nesterenkovii*, *B. paraconglomeratum*, *B. rhamnosum*, *B. sacelli*, and *B. tyrofermentans*. These bacteria contain MK-7 as the major component of manaquinone and the polar lipid profile is composed of diphosphatidylglycerol,

phosphatidylglycerol, and unidentified phospholipid and glycolipids [59]. *Brachybacterium* has been proved to be highly effective in removing manganese from solutions by converting it into insoluble manganese oxides. This bacterium did not only oxidize the manganese, but the resulting oxides themselves also absorbed the metal from the culture solution – thus making *Brachybacterium* sp. a potentially useful strain in bioremediation and cleaning up pollution. Also, this genus has been characterized as a cellulose-decomposing microorganism because of its unique attribute in converting a large amount of photosynthetically produced cellulosic materials into industrial substrates [60].

Coagulation-Flocculation Technology in Wastewater Treatment

This technology is usually employed in potable water and wastewater treatment to overcome the forces stabilizing the suspended particles, thereby facilitating the collision of particles and the formation of floc. Coagulation is a process whereby particle destabilization and charge neutralization occurs as a result of the addition of a positively charged ion of metal salt. On the other hand, flocculation refers to collision that occurs as a result of agitation that allows the particles to agglomerate into larger flocs. Land erosion, dissolution of minerals, and decaying of vegetation from domestic and industrial waste discharges have been responsible for suspended materials present in water and wastewater. Such materials may comprise dissolved organic and inorganic matters, and biological organisms such as bacteria, algae, or viruses. These materials must be removed or eliminated as they cause turbidity in water plus various health risks to people.

Most of the suspended materials or particles are smaller in size and they also carry negative charges in aqueous medium. As a result, to facilitate the process of settling, the particles have to come together to form larger flocs. However, this procedure is tedious as a result of electrostatic repulsion forces that hinder the particles from coming together coupled with the negative charge on the material. Therefore, it requires a longer time for settling and this problem can be solved by destabilizing the particles with the aid of a coagulant. Destabilization can be achieved either with one or a combination of two or more of the following mechanisms after the addition of a coagulant agent [61, 62]:

- a) Compression of the electrostatic double layer.
- b) Adsorption and charge neutralization.
- c) Adsorption and inter particle bridging.
- d) Enmeshment in precipitate using an excess coagulant dose.

Having destabilized the particles that are present in the wastewater, flocculation then facilitates the aggregation or conglomeration of flocs after the addition of an appropriate flocculating agent. Finally, particles must collide and this can take place under natural circumstances, (perikinetic floc formation), whereby aggregation is achieved by thermal motion of fluid molecules. It can also be achieved by dissipating mixing energy (orthokinetic floc formation) [62].

The Flocculation Mechanism

The concept of the flocculation mechanism has been classified into charge neutralization, electrostatic patch, and polymer bridging [63]. The flocculation mechanism for different types of flocculants and some of the examples of the mechanism are illustrated in Tables 3 and 4.

Charge Neutralization

This refers to the cancellation of negative charge when positively charged colloids or ions come in contact with a negative charge of a particle. Resultantly, electrostatic repulsion between the particles disappears and it enhances the process of coagulation. This also occurs when colloid particles and flocculants are of opposite charge. For instance, in a case where colloidal particles in industrial wastewaters are negatively charged, the use of inorganic flocculants and cationic polyelectrolytes are recommended [64].

Flocculants category	Flocculants type	Flocculation mechanism
Chemical coagulants	Inorganic metal salts	Charge neutralization
Chemical flocculants	Polyelectrolytes with low MW and low CD Poly electrolytes with high MW and low CD Poly electrolytes with low MW and high CD Polyelectrolytes with high MW and high CD	Charge neutralization Bridging Electrostatic patch Electrostatic patch + bridging
Bioflocculants	Cationic chitosan Anionic cellulose, tannin and sodium alginate Anionic/neutral plant-based flocculants	Charge neutralization + bridging Bridging Bridging
Grafted flocculants	Amphoteric/cationic/anionic graft copolymers	Charge neutralization + bridging

Table 3. Flocculation Mechanism for Different Types of Flocculants.

Flocculant type	Flocculant characteristics	Flocculation medium	Flocculation mechanism	Reference
Quaternary ammonium based derivative of polyacrylamide (cationic)	High MW (16 X 10 ⁶) High CD (100%)	Colloidal dispersion of anionic polystyrene latex particles	Bridging	[63]
Cationic polyacrylamide (C-PAM) Polyethyleneimine (cationic) polyDADMAC (cationic)	High MW, Low CD Low MW, High CD Medium,MW, Medium CD	Suspension of calcium carbonate	Bridging Electrostatic patch Charge neutralization	[64]
Cationic copolymers of acrylamide/ diallyldimethyl ammonium chloride	Medium MW (3 x 10 ⁵), Low CD (10%) Medium MW (1.2 x 10 ⁵), Medium CD (40%) Medium MW (1.2 x 10 ⁵), High CD (100%)	Suspension of silica particles	Bridging Charge neutralization + bridging/ bridging	[67]
Optionic and any lowids (C DAM)	High MW, Low CD	Suspension of calcium	Bridging	[68]
Cationic polyacrylamide (C-PAM)	High MW, High CD	carbonate	Electrostatic patch	
Cationic polyacrylamide (C-PAM)	High MW (7.2 x 10 ⁶), High CD (80%) High MW (13 x 10 ⁶), Medium CD (50%)	Suspension of calcium carbonate	Electrostatic patch + bridging bridging	[69]

Table 4. Examples of Flocculation Mechanisms.

Electrostatic Patch Mechanism

This involves charged polymers or colloids binding to a particle of the opposite charge. Particles attract each other through patches of opposite charges, causing coagulation of the suspension.

Flocculation can be induced by several methods. Metals such as alum and ferric chloride are commonly used coagulants. They usually dissociate in water and the metal ions can cause flocculation through the process of charge neutralization.

Bridging

This is a process whereby charged colloids come together to the surface of two different particles to form a bridge between the particles. This allows the particle to come together and is responsible for flocculation. High molecular-weight polymers with a low charge density adsorb on the surface in a way that long loops extend to the second surface. This allows polymers that are hanging to interact or attach together, thereby forming a bridge between the particles or colloids [65-67]. Polymer bridging has been reported as the mechanism for flocculation in treatment of textile wastewater with *Plantago psyllium* mucilage and *Tamarindus indica* mucilage [68, 69]. The length of polymers is a major determining factor that activates proper bridging.

Assessment of Flocculation Efficiency

The addition of flocculants into a cylinder or flask results in the formation of an interface. The upper phase known as the supernatant contains the liquid while the sediments settle below the container. The Jar test and settling test are the parameters used in assessing flocculation efficiency.

Jar Test

This technique involves the addition of a flocculant to the solution at a dose that varies from 0.025 ppm to 1 ppm. The working principle is achieved by stirring at a uniform speed. First, at a speed of 75 rpm for 2 minutes and then at a slow speed of 25 rpm for 5 minutes. Finally, a 10-minute settling time will be allowed before measuring turbidity of the supernatant using a turbidometer. In a situation where flocs are widely or evenly distributed, it is necessary to explore a higher velocity to suspended solids in order to obtain larger flocs.

Settling Test

In the case of a settling test, 100 ml graduated measuring cylinder and a stopwatch is usually employed. This involves the addition of a suspension sample and the flocculants in the same cylinder. The cylinder is shaken properly to ensure thorough mixing. Thereafter, the measuring cylinder is positioned upright and the height of the interface between water and settled sediment is measured over time. The settling test has been proven to be more accurate in terms of accuracy than the jar test.

Current Challenges and Future Direction

Environmental pollution is a global problem. Water pollution caused by industrial pollutants is a public menace, making both private and government sectors interested in mitigating this problem. Coagulationflocculation technology is an important physicochemical step in the treatment of wastewater. This has been employed to eliminate or reduce suspended colloidal particles accountable for the presence of organic matter and turbidity in wastewater, which contributes to biological oxygen demand (BOD) and chemical oxygen demand (COD) content of the water [70]. At present, flocculants are prevalent in a variety of industrial processes such as wastewater treatment, drinking water purification, and the downstream process in fermentation [71].

Even though some flocculating agents have been established in removing various pollutants from wastewater in a laboratory scale, there is still a necessity to improve their efficiency in the removal of suspended colloids, particles, and other forms of organic and inorganic pollutants before the wastewater is discharged into the environment. Although chemically synthetic flocculants are playing dominant roles in waste and water treatment, they are nonbiodegradeable and toxic to the environment. Some of these synthetic flocculating substances have threatened public health and increased environmental risks. For example, polyacrylamide, one of the most popular flocculants, includes acrylamide monomers which are verified as both neurotoxins and strong carcinogens to human beings [72]. On the other hand, aluminum salts are by far the most widely used coagulants in water and wastewater treatment. However, several disadvantages of using aluminum salts - including Alzheimer's disease and similar health problems associated with residual aluminum in treated waters – have been identified [73].

A significant major global economic factor is that many developing countries can hardly afford the high costs of imported chemicals for water and wastewater treatment. As a result, they depend on inorganic and organic synthetic flocculants that are toxic and non-degradable [74]. The potential application of bioflocculants in the treatment of different wastewaters, decoulorization of dyewastewaters, cell removal, and biomass recovery has been well investigated and established [75, 14, 19]. However, in spite of all the bioflocculants recently identified and explored, none has been practically applied in industry because of poor productivity and exorbitant production costs [76]. Hence, it is desirable that other cost-effective, biodegradable, and environmentally friendly bioflocculants with strong flocculating activity are isolated and screened to supplement - if not replace - alum, ferric salts, and synthetic polymers. Many of the bioflocculants employed in flocculation have been used as pure culture. There is an urgent need to explore the interactions between actinobacteria microorganisms in a mixed culture to enhance better flocculating efficiency in the treatment of wastewater.

To date, the application and efficiency of screened actinobacteria in flocculation have yet to be validated, especially for large-scale medium production and wastewater treatment. Thus for the sake of producing actinobacteria in a large-scale medium coupled with better and higher efficiency in flocculation, the screening of actinobacteria has become a subject of paramount and urgent research in our laboratory.

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