

Microalgae Harvesting

Subjects: **Plant Sciences**

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This presents the extractions, characterisations, and applications of natural coagulant in microalgae harvesting. The promising future of microalgae as a next-generation energy source is reviewed and the significant drawbacks of conventional microalgae harvesting. The performances of natural coagulant in microalgae harvesting are studied and outperformed alum. The aim of this work is to elucidate the key aspects for extraction of natural coagulants (plant, microbial and animal) and discussed with justifications. This information could contribute to future exploration of novel natural coagulants by providing description of optimised extraction steps for a number of natural coagulants. Besides, the characterisations of natural coagulants have garnered a great deal of attention, and the strategies to enhance the flocculating activity based on their characteristics are discussed. Several important characterisations have been tabulated in this review such as physical aspects, including surface morphology and surface charges; chemical aspects, including molecular weight, functional group and elemental properties; and thermal stability parameters including thermogravimetry analysis and differential scanning calorimetry.

natural coagulant

microalgae harvesting

flocculant

green material

plant material

characterisation

Chlorella vulgaris

Chlamydomonas sp.

1. Strategy to Enhance Performance of Natural Coagulants in Microalgae Harvesting

After the extraction processes, the final end product is the natural coagulant (plant, animal or microbes). Prior to application in coagulation and flocculation, the characterisation of natural coagulant is vital. Modification of the characteristic of natural coagulant could help in improving its performance in terms of flocculating activity in microalgae harvesting. Table 1 shows the physical, chemical and thermal characteristics of various natural coagulants. Additionally, the performance of various natural coagulants in different application is tabulated in Table 1. Subsequently, the interpretation of these characteristic in related to flocculating activity and their roles in enhancing the performance of natural coagulant in microalgae harvesting are discussed.

Table 1. Characterisation of natural coagulants and its performances.

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
Banana peel (<i>Musa acuminata</i>)	-N/A-	-N/A-	-N/A-	C=O, O-H, N-H	-N/A-	-N/A-	-N/A-	0.4 g·L ⁻¹ dosage, 67% removal of chemical oxygen demand (COD) from municipal wastewater	[1]
Banana pith	-N/A-	-N/A-	-N/A-	O-H, C-H, C-OO, C-H, COOH	O (44%), C (32%), H (4.2%), N (1.5%), S (0.86%)	-N/A-	-N/A-	0.1 kg·m ⁻³ dosage, pH 4, 99% removal of COD from river water	[2]
<i>Brachystegia eurycoma</i> extract	Compact structure with dispersed but continuous crack-like openings, absence of irregular surfaces, randomly formed aggregates and/or loosely bound cluster	-N/A-	-N/A-	O-H, N-H, O=H, C-N, C≡C, C=C-H and H-C-H	-N/A-	334.44 °C to 361.73 °C	-1.708 mV	5 g·L ⁻¹ dosage, pH 8, 97% removal of COD from paint wastewater	[3]
<i>Brassica spp.</i> seed protein	Pollen grain surface	-6.8 mV	6.5 kDa	-N/A-	-N/A-	95 °C	-N/A-	-N/A-	[4][5]
Cassava peel starch	Polygonal and spherical starch granules, rough surface	-4.37 mV	1.057 × 10 ⁵ kDa	O-H, C-H	Ca, K and Na	-N/A-	-N/A-	7.5 mg·L ⁻¹ dosage, pH 7, 93% removal of total suspended solid (TSS) from dam water 50 mg·L ⁻¹	[6]

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
								dosage, pH 7, 100% removal of <i>E. coli</i> from dam water	
Cactus leaves	Presence of cracks and cavities	-N/A-	-N/A-	O-H, C=O, COOH	Na, K, Ca, Mg	-N/A-	-N/A-	10 mg·L ⁻¹ dosage, 90% removal of kaolin	[7][8]
Cassava Peel (periderm and cortex)	Non-porous and heterogeneous characteristics, smooth and globular in shape	-N/A-	-N/A-	O-H, CH, CH ₂ , C=O, C-O, COOH	K ₂ O (5.5%), CaO (4.2%), Fe ₂ O ₃ (1.5%), SO ₃ and SiO ₂ (0.87%), Al ₂ O ₃ (0.74%), C (0.10%),	-N/A-	-N/A-	-N/A-	[9]
<i>Cassia obtusifolia</i> seed gum	Fibrous networks with rough surface and porosity	-N/A-	-N/A-	O-H, C-H, C=O,	-N/A-	289 °C	-N/A-	2.47 g·L ⁻¹ dosage, 82% removal of TSS, settling time of 35.16 min	[10]
<i>Ceratonia siliqua</i> seed gums	Rough cuticle on the adaxial and the abaxial surface, stomatal pores	-N/A-	5–8 kDa	O-H	-N/A-	-N/A-	-N/A-	-N/A-	[4][11]
Chitin	Microporous, fish scale shaped nanofibrous surface	+18 mV	-N/A-	N-H, O-H, C-H, C=O	-N/A-	-N/A-	-N/A-	0.3 g·L ⁻¹ dosage, pH 6, 68% removal of turbidity from surface water	[12][13][14]
Chitosan extracted from lobster shell	Rough surface, irregular block,	-N/A-	-N/A-	R-NH ₂ , O-H	Ca, K, Na, Mg and Fe	-N/A-	-N/A-	-N/A-	[15][16]

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
<i>(Thenus unimaculatus)</i>	crystalline with cluster and porosity structure								
Citrus <i>Limettioides</i> peels	Porous structure	-N/A-	-N/A-	CH, CH ₂ , CH ₃ , C=O, COOH, M(RCOO) _n ,	O, Na, Ca	-N/A-	-N/A-	-N/A-	[9][17]
<i>C. obtusifolia</i> seed gum	Rough, fibrous, porous and bulky	+6.41 mV	-N/A-	O-H, C-H, CH ₃ , CH ₂	-N/A-	280–300 °C	-N/A-	19 × 10 ⁻³ mol gum, 6 × 10 ⁻² mol of NaOH, 87% removal of TSS and 85% removal of COD from palm oil mill effluent (POME) at 50 °C	[18][19]
<i>Cocos nucifera</i> seed protein	Porous structure, clustered, aggregated shapes	-N/A-	5.6 kDa	O-H, N-H	-N/A-	-N/A-	-N/A-	10 g·L ⁻¹ dosage, 96% removal of As(III) in 8 h, 80 rpm and 50 °C	[4][20]
<i>Cucumis melo</i> peels	-N/A-	-N/A-	54 kDa	O-H, N-H, CH, CH ₂ , CH ₃ , C=O, R-COOH, M(RCOO) _n , C-O or -C-N	-N/A-	-N/A-	-N/A-	0.5 g·L ⁻¹ dosage, pH 7, 91% removal of Mn(II) 0.5 g·L ⁻¹ dosage, pH 6.5, 91% removal of Pb(II)	[9][21][22]
<i>Cyamopsis tetragonoloba</i> seed gums	Nanoparticles	-6.66 mV	50–800 kDa	O-H	-N/A-	-N/A-	-N/A-	-N/A-	[4][23]
<i>Dolichos lablab</i> seed gums	Aggregated free, rough	-N/A-	-N/A-	N-H, O-H, C-H, C-C, -COOH	C, O	-N/A-	-N/A-	0.6 mL·L ⁻¹ dosage, pH 11, 99%	[4][24]

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
								removal of turbidity	
Garden cress (<i>Lepidium Sativum</i>)	Flake-shaped structures with non-uniform distribution and emerged as interconnected channels, porous and heterogenous characteristics	-16 mV	-N/A-	O-H, C-H, C=O, OCH ₃	-N/A-	-N/A-	-N/A-	15 mg·L ⁻¹ dosage, pH 5, 99% removal of turbidity from river water	[25]
Grafted 2-methacryloyloxyethyl trimethyl ammonium chloride lentil extract	More compact and less porous compared to lentil extract	+15.08 mV	-N/A-	-N/A-	C (62%), O (36%), Cl (2.0%)	-N/A-	-N/A-	5.09 mL·g ⁻¹ dosage, pH 10, 99% removal of turbidity in surface water and industrial wastewater	[26]
<i>H. esculentus</i>	Compact, cross linkage of molecules	-N/A-	100 kDa	O-H, C-H, C=O	-N/A-	180 °C	36.12 mV	-N/A-	[27][28]
Kenaf crude extract (KCE)	-N/A-	-8.3 mV	-N/A-	-N/A-	-N/A-	-N/A-	-N/A-	100 mg·L ⁻¹ dosage, 85% removal of kaolin, 40 mg·L ⁻¹ , 83% removal of turbidity from river water	[29]
<i>Klebsiella pneumoniae</i>	-N/A-	-N/A-	-N/A-	COO ⁻ , O-H, N-H	C, N, O	-N/A-	-N/A-	pH 7, 40% removal of Cd	[8][30]
<i>Lens culinaris</i>	Rough surface with pores and obvious surface abrasions	-3.58 mV	-N/A-	O-H, C-H, COOH, C=O, C-O	C (60%), O (40%), K (0.39%)	-N/A-	-N/A-	26.3 mg·L ⁻¹ dosage, 99% removal of kaolin, 3 min settling time	[31]

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
Lentil extract	Highly porous surface, scattered pieces of compounds attached	-5.91 mV	-N/A-	O-H, C-H, C=O, N-H, C-O-C	C (59%), O (39%)	280 °C	-N/A-	-N/A-	[26]
Maerua decumbent	-N/A-	-N/A-	-N/A-	O-H, C-H, N-H, C=O, C-O, C-N	C (39%), O (42%), H (3.8%), N (1.2%), S (0.31%)	-N/A-	-N/A-	1 kg·m ⁻³ dosage, pH 5.56, settling time 52.31 min, 99% removal of turbidity from paint industry wastewater 0.8 kg·m ⁻³ dosage, pH 5.11, settling time 53.53 min, 79% removal of COD from paint industry wastewater	[32]
Malva nut gum	A branch-like surface structure	-58.7 mV	2.3 × 10 ⁵ kDa	-N/A-	-N/A-	-N/A-	-N/A-	0.06 mg·L ⁻¹ dosage, pH 3.01, 97% removal of kaolin	[33]
Mango peels	Well-pronounced heterogeneous cavities that are well distributed	-N/A-	-N/A-	O-H, N-H, CH, CH ₂ , CH ₃ , C=O, C-O or -C-N	C, H, N, S	-N/A-	-N/A-	-N/A-	[9][34]
Moringa oleifera [47]	Group-like, composed of many small particles	+6 mV	6.5 kDa	O-H, C-H, C=O, N-H, C-OH, S=O	-N/A-	-N/A-	-N/A-	50 mg·L ⁻¹ dosage, 94% removal of kaolin	[8][29][35][36]
Nirmali seeds [48]	highly porous with reticulated structure	-N/A-	12 kDa	COOH, O-H	-N/A-	-N/A-	-N/A-	1.5 mg·L ⁻¹ dosage, 96% removal of	[8][37]

the previous study by Obiora-Okafo and Onukwuli [43] proved that a compact net structure coagulant showed higher flocculating activity as compared with a branched structure. Furthermore, changes to the surface morphology of coagulants after coagulation and flocculation show proof of interaction between the coagulants and suspended particles. In view of surface morphology as a strategy to enhance the flocculating activity, modification on physical structures such as grafting could be done to create a high density of pores and ultimately more favourable to coagulation. With these, the mass harvesting of microalgae in the industrial scale is applicable.

On the other hand, surface charge, or zeta potential, is one of the factors that will affect the flocculating activity. Theoretically, zeta potential is the measure of the electrical charge of particles that are suspended in liquid [49]. Practically, the higher the negative surface charge of natural coagulant, the greater it's flocculating activity against positive suspended particles and vice versa for the positive surface charge of natural coagulant against negatively suspended particles. Thus, the study of surface charge shows a preliminary estimation of flocculating activity of natural coagulant. Besides, the nature of surface charge (positive or negative) indicates the potential treated group

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
								turbidity from surface water	
okra	Porous and rough	-8.3 mV	-N/A-	-N/A- [8]	Mg (7.2%), Al (4.1%), Si (3.7%), P (11.8%), S (8.2%), Cl (7.7%), K (22.0%), Ca (7.5%), O (27.8%)	-N/A-	-N/A-	3 g·L ⁻¹ dosage, 85% removal of fluoride from hydrofluoric acid synthetic wastewater 20 mg·L ⁻¹ dosage, 94% removal of kaolin, 40 mg·L ⁻¹ dosage, 98% removal of turbidity from river water	[35]
Prosopis spp. seed [52]	Homogenous in size and shape with a flake-like morphology	-N/A-	62 kDa	-N/A-	-N/A-	Ca, Mg, Fe, Zn	-N/A-	[15]	[4][29][38]
Sabdariffa crude extract (SCE)	-N/A-	-6.4 mV	-N/A-	-N/A-	-N/A-	-N/A-	-N/A-	60 mg·L ⁻¹ dosage, 88% removal of kaolin, 40 mg·L ⁻¹ dosage, 96% removal of turbidity from river water	[29]
Sago	Smooth and solid surface with no pores	-N/A-	-N/A-	N-H, O-H, C=O	-N/A-	-N/A-	-N/A-	0.1 g·L ⁻¹ dosage, pH 7, 69% removal of turbidity from surface water	[12]
Tannin	-N/A-	-13.6 mV	1250 kDa	O-H, R-NH ² , C=O, COOH [53]	-N/A-	200 °C	-N/A-	14 mg·L ⁻¹ dosage, 75% removal from kaolin	[4][8][39]

1.2. Chemical Characteristics

The flocculating activity of natural coagulants also depends on the specific chemical properties of the polymer. One of the key polymer characteristics includes various functional groups. The particular functional groups to be evaluated are COO⁻ and OH⁻ as their existence usually contributes to the flocculating activity of natural coagulant. Besides, the increase in positively charged functional groups allows more interactions with the negatively charged suspended particles, and thus improve the binding capabilities of natural coagulants [52]. Modification on functional groups of natural coagulants is also proposed and evinced by researchers in the past studies to increase the flocculating activity. For example, functionalising of cationic starch and TANFLOC, in which, the starch and tannins added with quaternary ammonium groups to increase the flocculating activity and serve as the low-cost as well as more effective alternatives for flocculation process Selective Flocculation Enhanced Magnetic Separation of Ultrafine Disseminated Magnetite OresSelective Flocculation Enhanced Magnetic Separation of Ultrafine

Natural Coagulant	Surface Morphology	Surface Charge	Molecular Weight	Functional Group	Elemental Property	Thermogravimetry Analysis	Differential Scanning Calorimetry	Performance	Reference
								11 mg·L ⁻¹ dosage, pH 5 to 7, 97% removal of <i>Chlorella vulgaris</i>	
<i>Tamarindus indica</i> seed gums	No fissures, cracks or interruptions	-N	700–880 kDa	3 4 -N/A-	-N/A-	97.67 °C	128.40 J/g	15 ppm dosage, 94% removal of turbidity from river water	[4][40][41]
<i>Telfairia occidentalis</i> seed	Coarse fibrous substance largely composed of cellulose and lignin, presence of pores (micro-, macro- and mesopores, compact net structure	-N/A-	-N/A-	O-H, N-H, C=H	-N/A-	-N/A-	-N/A-	247.40 mg·L ⁻¹ dosage, pH 2, 99% removal of dye in 34.32 mg·L ⁻¹ concentration with 540 settling time	[42][43]
<i>T. foenum graecum</i> seed gums	-N/A-	-N/A-	32.3 kDa	O-H, C-H, C=O, N-H, C-OH, C-O-C	C,O	295 °C to 430 °C	-N/A-	-N/A-	[28][4][44]
Vegetable tannin	-N/A-	-N/A-	-N/A-	-N/A-	-N/A-	430 °C	-N/A-	pH 7, removal of color and turbidity from dairy wastewater	[45]
<i>Vigna unguiculata</i> seed proteins	Fairly uniform, hexagonal structure, spiked or rugged surface, rough surface, coarse fibrous	-N/A-	6 kDa	O-H, N-H, C=O, C=C-H, C=CH, C-H	-N/A-	-N/A-	-N/A-	256.09 mg·L ⁻¹ dosage, pH 2, 99% removal of dye of 16.7 mg·L ⁻¹ with 540 min settling time	[4][42][43][46]

of okra mucilage during the harvesting process [56]. The presence of galacturonic acid in mucilage will act as an active coagulating agent and provide a bridge for particles adsorption. Further, the partial deprotonation of carboxylic functional group of mucilage in aqueous solution has given rise to the chemisorption between charged particle with COO⁻ and OH⁻ [8]. Therefore, it will aid in flocculating activity. To conclude, the selection of natural coagulant for microalgae harvesting should be focused on mucilage as its primary concern.

1.3. Thermal Characteristics

The thermal stability of natural coagulants is also a crucial parameter to be studied in enhancing the flocculating activity. Indeed, an optimum temperature will increase the flocculating activity. However, the temperature higher than 80 °C will usually destroy the chemical composition of natural coagulants [57]. Moreover, the temperature has direct effects on floc formation, breakage and reformation. To illustrate, floc formation is slower at a lower temperature, whereas breakage of floc is greater at higher temperatures. On the other hand, thermogravimetry analysis determines the minimum temperature causing decomposition of organic components in natural coagulant

and differential scanning calorimetry allows study relating to the heat flow required to decompose the natural coagulant. In general, the thermal characteristics reveal the thermal stability of natural coagulant and it has no direct impact on microalgae harvesting because coagulation will not occur in extreme temperature.

| 2. Application of Natural Coagulant in Microalgae Harvesting

In the previous section, the extraction and characteristic of natural coagulant, as well as the strategies to enhance its flocculating activity, are reviewed. In this section, the application of natural coagulant in microalgae harvesting will be the focal point. To recall, alum always appears to be the first option in industrial applications when comes to the selection of coagulant for microalgae harvesting. The reason being, it is widely available, it promotes coagulation by neutralisation and most importantly it is ready to be dissolved with water.

However, the emerging usage of plant-based coagulant has achieved higher harvesting efficiency compared with chemical coagulant and there are reviews on their effectiveness and relevant coagulating mechanisms for the treatment of wastewater and microalgae harvesting [\[56\]](#)[\[58\]](#)[\[59\]](#). To illustrate, the plant-based coagulant could be applied on microalgae harvesting at relatively low cost [\[60\]](#). Compared to alum, the natural coagulant is deemed to be environmentally friendly because it is extracted from plants, animal or microbial and usually existed in non-toxic form [\[61\]](#). The water soluble active compound in natural coagulant will be removed after several cycle of kidney filtration, leaving less possibility of producing toxicity in the body [\[62\]](#). In view of sludge production after the harvesting process, natural coagulant does not produce suspended alum residual and indeed produces less organic residual due to its biodegradability. In contrast, alum requires chemical reaction to break down and will not decompose naturally. In a specific type of microalgae harvesting, for instance, extraction of DHA rich microalgae oil as a dietary supplement, natural coagulant appears to be the best option as it harvests a higher amount of microalgae biomass compared to alum and at the same time, it is safe for consumption. Thus, it will not pose any health concern even there is residual remained in algae biomass. The natural coagulant is proven to achieve higher flocculating activity in comparison to alum and their performance is shown in Table 1 and Table 2. In addition, by utilising the natural coagulants, it reduces the alum dependency and ultimately achieves sustainability in the microalgae-based biofuel production industry as well as various fields, including wastewater treatment and medical to name a few. Figure 1 shows the advantages of natural coagulant in microalgae harvesting.

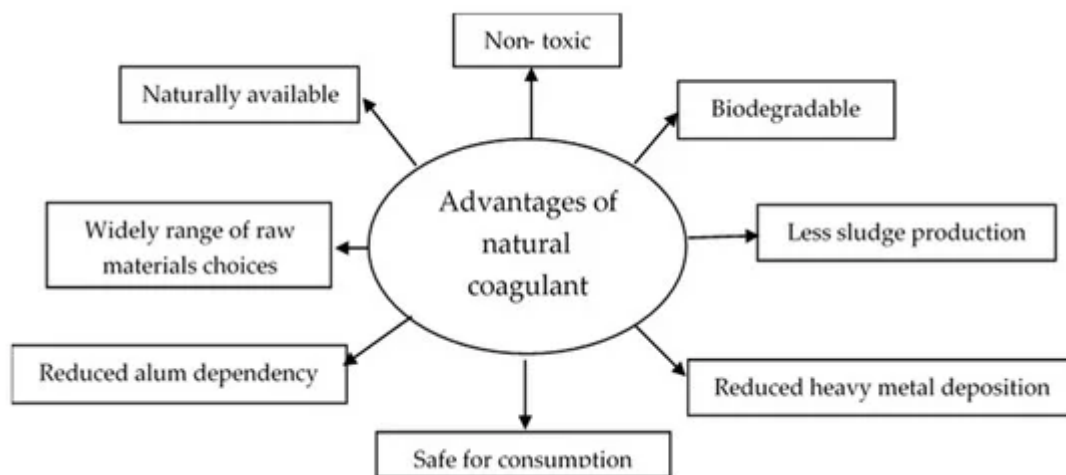


Figure 1. Advantages of utilising natural coagulant in microalgae harvesting.

Furthermore, natural coagulants have also been proven by other researchers as an effective way to harvest microalgae. It was found that the usage of bio-coagulants for harvesting microalgae could eliminate the toxicity contamination on harvested microalgae biomass [63]. The study carried out by Tran et al. [64] to harvest *Chlorella vulgaris* with alkyl-grafted chiton $\text{Fe}_3\text{O}_4\text{-SiO}_2$ showed 90% of biomass removal by merely employing $0.013\text{g}\cdot\text{L}^{-1}$ dosage. On the other hand, a plant-based coagulant, *M. oleifera*, showed a 76% of harvesting efficiency on *Chlorella* sp. biomass after 100 min with $8\text{mg}\cdot\text{L}^{-1}$ dosage and 96% of harvesting efficiency in 20 min when combining *M. oleifera* with chitosan [65]. Furthermore, 60% of microalgae removal efficiency was achieved with $12\text{mg}\cdot\text{mL}^{-1}$ of *F. indica* extract after 120 min of settling time [66]. To sum up, the utilisation of natural coagulants in microalgae harvesting is a trend of research in the past few years. Unfortunately, it was set up and investigated merely at a laboratory scale. Table 2 shows the application of natural coagulants on microalgae harvesting.

Table 2. Application of microalgae harvesting using natural coagulants.

Natural Coagulant	Operating Condition	Performance	Reference
Alkyl-grafted chiton $\text{Fe}_3\text{O}_4\text{-SiO}_2$	$0.013\text{g}\cdot\text{L}^{-1}$ dosage	90% removal of <i>Chlorella vulgaris</i>	[64]
<i>M. oleifera</i>	$8\text{mg}\cdot\text{L}^{-1}$ dosage	76% removal of <i>Chlorella vulgaris</i>	[65]
<i>M.oleifera</i> with chitosan	$8\text{mg}\cdot\text{L}^{-1}$ dosage	96% removal of <i>Chlorella vulgaris</i>	[65]
<i>F. indica</i>	$12\text{mg}\cdot\text{mL}^{-1}$ dosage	60% removal of microalgae	[66]
<i>Pleurotus ostreatus</i> strain HEI-8	pH 3, glucose content $20\text{g}\cdot\text{L}^{-1}$, fungi pelletisation time 7 days, 100 rpm	65% removal of <i>Chlorella</i> sp.	[67]
<i>Citrobacter freundii</i> (No. W4) and <i>Mucor circinelloides</i>	pH 7, glucose concentration	97% removal of <i>Chlorella pyrenoidosa</i>	[68]

Natural Coagulant	Operating Condition	Performance	Reference
	1.47g·L ⁻¹		
Tannin	11 mg·L ⁻¹ dosage, pH 5 to 7	97% removal of <i>Chlorella vulgaris</i>	[69]
Tannin	5 mg·L ⁻¹ dosage, pH 7	80% removal of <i>Oocystis</i> microalgae	[70]
<i>Eucalyptus globulus</i>	20 mg·L ⁻¹ dosage	95% removal of <i>Scenedesmus</i> sp.	[71]
Cassia gum	80 mg·L ⁻¹ dosage	93% removal of <i>Chlamydomonas</i> sp.	[72]
Cassia gum	35 mg·L ⁻¹ dosage	92% removal of <i>Chlorella</i> sp.	[72]

condition of natural coagulant. After several trials in the coagulation process, a statistical approach such as linear regression method is feasible in extracting the optimum parameters of natural coagulant in coagulation with collected data and equations.

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