

Investigation on the removal of contaminants from washing machine discharge using *Strychnos potatorum* (clearing nut) – A potential purifying agent

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ABSTRACT

The objective of the present study is to develop eco-friendly purifying agents from natural materials, *Strychnos potatorum* (SP) – a coagulant. The seeds of SP were used for wastewater treatment (washing machine effluent). Physico-chemical parameters like turbidity, total dissolved solids (TDS), electrical conductivity (EC), pH, calcium (Ca^{2+}), magnesium (Mg^{2+}) and phosphate (PO_4^{3-}) were tested and the data obtained were compared with BIS standards. In the current work, physical and chemical parameters have been examined to determine the quality of water for washing machine discharge, before and after treatment (coagulation) using SP seeds. The testing procedure was carried out using a Jar test of coagulant mass 1000 mg/L with rapid mixing at 150 rpm for 5 min, slow mixing at 30 rpm for 25 min and a settling period of 1 h. Coagulation results were reported for the use of SP seeds, where the effectiveness of removing turbidity and PO_4^{3-} was determined to be 81.54 and 41.45%. The presence of various functional groups in SP seed was assessed by fourier transform infrared (FT-IR) spectroscopy. Qualitative identification of phyto-chemical constituents of seed extract for this species was also carried out. The experimental findings strongly highlighted the ability of SP seeds to remove turbidity and PO_4^{3-} from wastewater.

1. Introduction

Excessive use, wastage, pollution and mismanagement of available water resources may lead to serious crises in future and cause many problems in health and livelihood. The world water development report predicts that the demand for water will increase by 55% in the year 2050 [1]. Due to water scarcity, two main methods can be considered: water storage and recycling. It is only possible to treat the wastewater at the waste production site, therefore, it is wise to take steps to recycle the water. Removing pollutants from wastewater and allowing it to re-enter local water systems is the process of water recycling. The term “wastewater treatment process” refers to a number of processes crucial for clearing up turbid water [2]. Industrial and domestic waste are the two categories of wastewater. On the one hand, industries create industrial waste as an undesirable by-product [3,4]. Domestic waste, on the other hand, is generated by everyday household tasks like using the toilet, washing machine or sink [5]. Three-quarters of home wastewater is

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washing machine trash. In underdeveloped nations, treating wastewater affordably has been shown to be the main challenge.

The various treatment techniques, such as adsorption [6–8], membrane technology [9], ion exchange process [10], electrochemical techniques [11] etc., are encouraged to remove pollutants. Each individual has advantages and practical constraints. Although many technologies have been implemented as solutions to provide adequate safe water, coagulation is currently considered the most affordable and suitable method to treat wastewater [12–14]. Natural coagulants derived from plants can generate economic benefits. Systematic screening of plant species is routine in many laboratories for the purpose of detecting new bioactive compounds. The active compounds derived such as proteins, polyphenols, polysaccharides from natural coagulants have a substantial impact on the efficiency of turbidity reduction.

The plant materials like *Opuntia* spp. [15], *Musa acuminata* [16], *Carica papaya* [16], *Azadirachta indica* [16], *Phaseolus vulgaris* [17], *Pisum sativum* [18], *Moringa oleifera* [19,20], *Strychnos potatorum* [18] as natural coagulants have been used to remove pollutants from wastewater. Among them, *Strychnos potatorum* species are valuable traditional medicines found in India, Sri Lanka and Burma [21]. The seeds of SP were reportedly employed to clear muddy lake water around 4000 years ago. Seeds are globose in shape and used for treatment of bronchitis, burning sensation, conjunctivitis, chronic diarrhoea, dipsia, dysentery, diabetes, eye troubles, gonorrhoea, leucorrhoea, hepatopathy, gastropathy, nephropathy, scleritis, renal and vesical calculi [22]. In accordance with examining the literature, the removal of dye in industries using SP seeds has been studied [23,24]. Mathota Arachchige et al. (2022) investigated research on Polyacrylamide-SP seed derived activated carbon composite for removal of hardness in drinking water [25]. The removal of contaminants from wastewater using this species is listed in Table 1. Even though research is carried out on this species, the widespread use of SP in domestic waste treatment is still limited.

SP seed is used to treat wastewater and is safe for human consumption. The powdered seed of SP contains coagulating qualities that have been employed for different aspects of wastewater treatment, including as turbidity, TDS, and pH. Many important physico-chemical parameters such as EC, Ca^{2+} , Mg^{2+} , and PO_4^{3-} have not been elucidated [18,28,30]. The main objective of the study focuses on evaluating the potential of SP seeds in treating washing machine effluent. Therefore, the novelty of the present work is to determine the various physicochemical parameters using SP seed powder. This knowledge will help to understand the coagulation mechanisms of SP seeds.

2. Materials and methods

2.1. Wastewater collection

The type of wastewater used in this study was washing machine discharge. Samples were collected from the residential area of Alwarthurich located along Tenkasi District, Tamil Nadu, India. The collected washing machine discharge is cloudy and turbid. Coagulation tests were performed on the water discharged from the washing machine.

2.2. Preparation of coagulant

Strychnos potatorum seeds were collected from the Ayurvedic herbal market located in Tirunelveli district of Tamil Nadu, India (Fig. 1). Distilled water was used to remove all dirt on the surface of the seeds. To remove moisture, these partially dried seeds are sun-dried for a week and then in an oven at 80 °C for 12 h. The dried seeds were collected and crushed into powder form using a domestic mixer [31]. The extract was prepared by mixing 1 g of coagulant with 1000 mL of distilled water for 1 h at room temperature, which possess coagulant characteristics. This was done using a magnetic stirrer. Then the solution was filtered using a filter paper [32]. Fig. 2 shows the steps involved in the preparation of natural coagulants.

2.3. Wastewater treatment

The collected sample was used for coagulation study to evaluate the efficacy of SP with the help of Jar test. The initial turbidity of the sample is first noted before adding the coagulant. Beakers were filled with 500 mL of collected wastewater by coagulant dosage of 5 mL for rapid stirring at 150 rpm for 5 min. Then, the speed was reduced to 30 rpm for about 25 min. The water sample was left undisturbed for 1 h to allow all the particles to settle (sedimentation). After 1 h, the clear water sample was collected in a conical flask. It is filtered using filter paper to remove the remaining sediment. Now the turbidity is observed and the sample is stored at 4 °C for further analysis [26,27]. Fig. 3 shows the stages involved in the coagulation process.

Table 1
Research on removing contaminants using SP.

Type of water	Technology	Conditions	Removal efficiency	Reference
Industrial water	Coagulation	150 rpm for 2 min, 30 rpm for 30 min	Turbidity – 50–84%	[26]
Washing machine discharge	Coagulation	150 rpm for 2 min, 30 rpm for 30 min	Turbidity – 65–84%	[27]
Laundry discharge	Adsorption		TDS – 92.86%	[28]
River water	Coagulation	120 rpm for 1 min, 40 rpm for 10 min, 20 rpm for 10 min	Turbidity – 71.42%	[29]
Washing machine discharge	Coagulation	150 rpm for 5 min, 30 rpm for 25 min	Turbidity – 81.54%, PO_4^{3-} – 41.45%	Present study



Fig. 1. *Strychnos potatorum* seeds.

3. Analytical methods

Physical parameters such as turbidity were measured using Digital Nephelo-turbidity meter (Systronics, Model No.132), EC by EC meter (Systronics-MK 509), and pH using Digital pH meter (Scientific tech-ST 2001). The chemical parameters determined in this study include major cations such as Ca^{2+} by flame photometer (Systronics-130), Mg^{2+} using EDTA titrimetric method, and minor anion PO_4^{3-} were measured using colorimeter (Systronics Instruments-113).

Using the following equation, the percentage of removal efficiency can be determined:

Removal efficiency (%) = $\frac{C_i - C_f}{C_i} \times 100\%$ where C_i is the concentration before treatment and C_f is the concentration after treatment.

4. Results and discussion

The physicochemical characteristics for wastewater sample discharged from washing machine, before and after treatment were analyzed and presented in Table 2. The phytochemical analysis for various extracts of SP seed were studied and listed in Table 3.

4.1. Physical characteristics

Turbidity is actually the amount of dissolved compounds or suspended particles in water. Turbidity in water arises from the presence of very finely separated solids (organic and inorganic). The initial turbidity of the effluent from the washing machine was 65 NTU (before treatment). Particles that create turbidity can interfere with the treatability of water and the effects of excessive turbidity can result in devastating. After treating with SP seed powder, the turbidity value was reduced from 65 to 12 NTU. The removal of turbidity efficiency was reached to 81.54%. These results were in complete agreement with the results of previous studies indicating that turbidity was reduced from wastewater samples after treatment with SP seed powder [26].

TDS in water is the amount of solids dissolved in water. In the present study, TDS increased from 6573 to 7237 mg/L. The initial value of TDS is due to water flow through stones, pipes and chemical detergents used for washing. The increase in TDS value after treatment may be the result of phytoconstituents in the treating agent. If TDS is high, the water will be saline. These obtained results met with the findings of Shan et al. (2017) [34]. While the TDS values did not match those reported by Mangale et al. (2012), which stated that TDS has decreased after treatment with *Moringa oleifera*. This might have occurred as a result of the unique pre-treatment procedure [35].

In general, EC is the ability to conduct electricity. As the concentration of ions increases, the conductivity also increases. The conductivity of the wastewater sample before treatment was 8710 $\mu\text{S}/\text{cm}$. After treatment the conductivity value reaches 8933 $\mu\text{S}/\text{cm}$. The only factor affecting the EC of the colloidal suspension and removal efficacies is the association and dissociation of the coagulant utilised for the coagulation. A high conductivity value is not suitable for the survival of some organisms. On the one hand, there was not much variation in conductivity after treatment [36].

The pH of water is a very important measure of water quality. pH has an impact on removal effectiveness. The pH scale varies from 0 (highly acidic) to 14 (highly alkaline). In the present work, the pH value was varied from 6.81 to 7.11. The pH of the sample was not significantly altered after treatment with SP seed powder. As per BIS standards, pH values should be less than 8.5 and more than 6.5. This demonstrated that SP seed powder did not affect the pH values in wastewater samples, in agreement with published observations [34]. Therefore, no additional step is required to adjust the pH value after treatment. The charge neutralization process was encouraged by this coagulant, which did not change the pH of the colloidal solution.

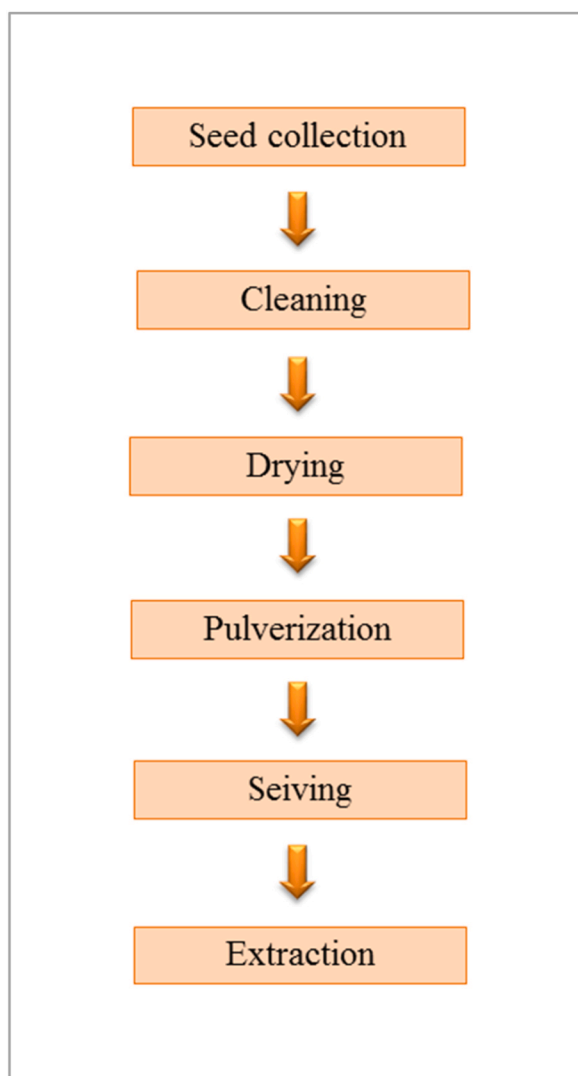


Fig. 2. Steps involved in the preparation of natural coagulant.

4.2. Chemical characteristics

Ca^{2+} and Mg^{2+} ions are commonly present in natural waters and are essential elements for all organisms. The most common divalent cation found in water is calcium, the first major component of hardness. Magnesium is the second major component of hardness. As per BIS standards, calcium concentration can go up to 200 mg/L and magnesium concentration up to 100 mg/L. After treatment, calcium concentration increased from 59 to 64 mg/L and magnesium concentration from 35 to 40 mg/L. As a result, there was no significant change in Ca^{2+} and Mg^{2+} after treatment.

A recent study using nanomaterials to remove phosphate from solutions successfully illustrates the effectiveness of improved phosphate treatment technologies. The primary drawbacks of this sort of treatment approach are the toxicity and high cost of nanomaterials [37]. As per BIS standard, PO_4^{3-} in water has no toxic effect. In the present study using natural coagulant, the PO_4^{3-} concentration was reduced from 15.97 to 9.35 mg/L. This is correlated with the charge neutralization mechanism [38]. The efficiency of PO_4^{3-} removal from wastewater is determined to be 41.45%.

FT-IR:

Fig. 4 shows the FT-IR spectra of the coagulant recorded in the range of 4000-500 cm^{-1} (Nicolet IS5R). The chemical nature of the functional groups in the SP seed is a primary determinant of its ability to coagulate. The broad peak at 3286.41 cm^{-1} is due to the stretching vibration of amine and hydroxyl groups [39]. The peak at 2922.28 cm^{-1} is attributed to the $-\text{CH}_2$ stretching vibration of the alkyl group. The peak at 2349.75 cm^{-1} is responsible for $\text{C}\equiv\text{N}$ stretching [40]. A bending vibration at 1596.42 cm^{-1} confirmed the existence of water. At 1415.76 cm^{-1} , the CH_2 bending vibration was recorded. At 1241.93 cm^{-1} , the presence of C-N stretching vibration was verified. The $-\text{CO}$ stretching vibration of the ether group resulted in an intense peak at 1015.54 cm^{-1} [39]. As a result, the

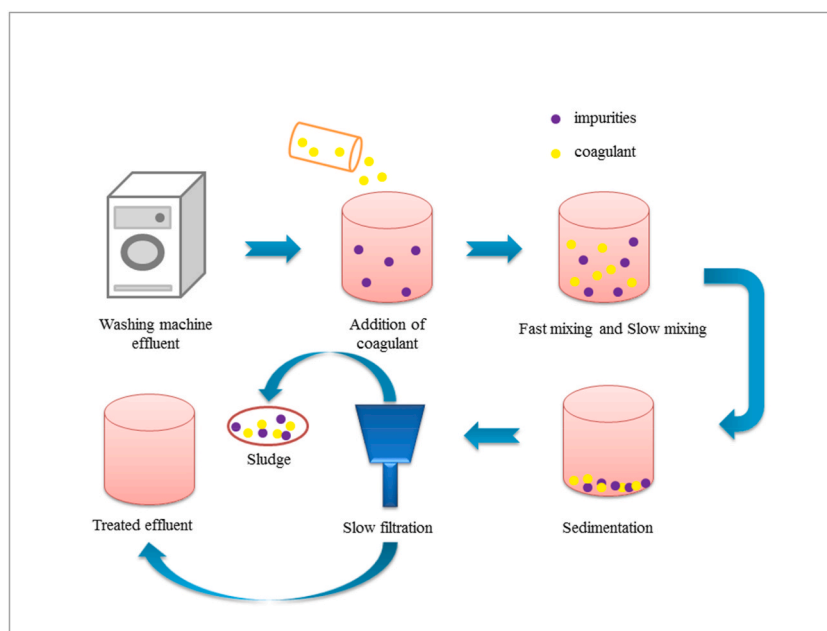


Fig. 3. Stages of coagulation process.

Table 2
Water quality analysis.

Parameters	Acceptable limit	Permissible limit in the absence of alternate source	Washing machine discharge	
	BIS 10500:2012 [33]		Before treatment	After treatment
Turbidity (NTU)	1	5	65	12
TDS (mg/L)	500	2000	6573	7237
EC ($\mu\text{S}/\text{cm}$)	–	–	8710	8933
pH	6.5–8.5	6.5–8.5	6.81	7.11
Ca^{2+} (mg/L)	75	200	59	64
Mg^{2+} (mg/L)	30	100	35	40
PO_4^{3-} (mg/L)	–	–	15.97	9.35

FT-IR spectrum demonstrates that the SP seed mostly contains an aliphatic group with ether linkages and amine bonds. Therefore, SP contains various organic compounds (such as alkaloids and carbohydrates) that have $-\text{COOH}$ and free $-\text{OH}$ surface groups which improve coagulation ability [30,41].

4.3. Phytochemical screening

Phytochemical study characterizes the phytoconstituents present in this seed. Phytochemical analysis of plant-based products identifies whether a plant has coagulating or anticoagulant qualities. The plant extracts of ethanolic, methanolic and aqueous solutions were assessed for the existence of the phytochemical analysis of SP seed powder by using the standard methods [42]. These extracts were utilised to conduct a preliminary screening for phytochemicals such as alkaloids, flavonoids, sterols, terpenoids, anthraquinone, anthocyanin, proteins, phenolic compounds, quinones, carbohydrates, tannin, saponin, cardiac glycosides, lignin, and coumarins. Any change in colour or formation of a precipitate was used to indicate a positive response to these tests.

4.4. Test for alkaloids

Mayer's test: A few drops of Mayer's reagent (potassiummercuric iodide) was added to a small amount of extract, the formation of a cream coloured precipitate indicated the presence of alkaloids (Figs. 6a and 7a).

Dragendorff's test: The extract was treated with a few drops of Dragendorff's reagent (bismuth potassium iodide), formation of a reddish-brown precipitate indicating the presence of alkaloids (Figs. 6b and 7b).

Table 3
Phytochemical analysis for the various extracts of *Strychnos potatorum* seed.

S.No.	Metabolite	Test applied	Aqueous Extract	Ethanol Extract	Methanol Extract
1.	Alkaloid	Mayer's test	-	+	+
		Dragendorff's test	-	+	+
2.	Flavonoid	Alkaline reagent test	-	-	-
		Lead acetate test	-	-	-
		Shinoda test	-	-	-
		Libermann-Burchard test	+	+	+
3.	Sterols	Libermann-Burchard test	+	+	+
4.	Terpenoids	Salkowski test	-	-	-
5.	Anthraquinone	Ammonium Hydroxide test	+	-	-
6.	Anthocyanin	2 M NaOH test	-	-	-
7.	Proteins	Ninhydrin test	-	-	-
		Biuret test	-	-	-
		Xanthoproteic test	-	-	-
		Ferric Chloride test	-	-	-
8.	Phenolic compounds	Ferric Chloride test	-	-	-
9.	Quinones	Concentrated HCl test	-	-	-
		Alcoholic KOH test	-	-	-
10.	Carbohydrates	Molisch's test	+	+	+
		Fehling's test	+	+	+
11.	Tannin	Ferric chloride test	-	-	-
12.	Saponin	Foam test	+	-	+
13.	Cardiac glycosides	Baljet's test	-	-	+
		Bromine water test	-	-	+
		Keller-killani test	-	-	+
		Labat test	-	-	-
14.	Lignin	Labat test	-	-	-
15.	Coumarins	NaOH test	-	-	-

+ = presence; - = absence

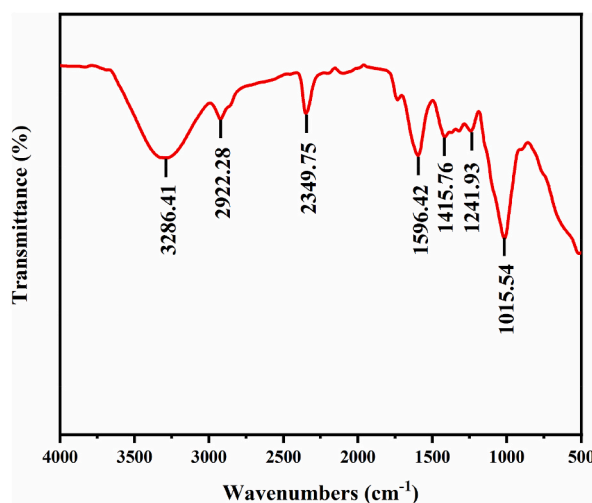


Fig. 4. FT-IR spectra of SP seed.

4.5. Test for flavonoids

Alkaline reagent test: The extract was taken in a test tube and a few drops of NaOH solution were added; an intense yellow colour was produced, which became colourless when a few drops of dilute HCl were added to the mixture. It showed the presence of flavonoids.

Lead acetate test: The extract was added with a few drops of lead acetate solution, and the appearance of a white precipitate indicates the presence of flavonoids.

Shinoda test: A few drops of concentrated HCl were added to extract. Then magnesium turnings were put into the solution and observed for the appearance of crimson red colour.

4.6. Test for sterols

Libermann-Burchard test: A chloroform solution with concentrated sulphuric acid and acetic anhydride produces a blue-green

colour, indicating the presence of sterols (Figs. 5a, 6c and 7c).

4.7. Test for terpenoids

Salkowski test: The extract was treated with chloroform; followed by concentrated sulphuric acid to form two distinct layers, the upper layer (chloroform) acquires a bluish red to violet, while the layer (sulphuric acid) changes from yellow to green, with a greenish glow being visible.

4.8. Test for anthraquinone

Ammonium Hydroxide test: A drop of concentrated ammonium hydroxide was added to the prepared extract solution, previously dissolved in isopropyl alcohol. After 2–3 min, the formation of reddish orange colour indicates the presence of anthraquinone (Fig. 5b).

4.9. Test for anthocyanin

2M NaOH test: The presence of anthocyanin has been demonstrated by adding 2 M NaOH to the extract, resulting in the formation of a blue green colour.

4.10. Test for proteins

Ninhydrin test: Take the sample solution in a test tube, pour 2% Ninhydrin reagent and heat in a water bath for 3–5 min, the development of purple colour indicates the presence of proteins.

Biuret test: The extract is treated with one drop of 2% copper sulphate solution. 1 ml of ethanol (95%) is added to this solution, followed by potassium hydroxide pellets. A pink colour in the ethanolic layer indicates the presence of proteins.

Xanthoproteic test: Concentrated nitric acid is added to a sample and the mixture is heated until it turns yellow.

4.11. Test for phenolic compounds

Ferric Chloride test: To the extract, a few drops of neutral 5% ferric chloride solution are added. A dark green colour indicates the presence of phenolic compounds.

4.12. Test for quinones

Concentrated HCl test: A small amount of the extract was treated with concentrated HCl and observed for the formation of a yellow precipitate.

Alcoholic KOH test: When alcoholic KOH is added to the test sample, the colour changes from red to blue.

4.13. Test for carbohydrates

Molisch's test: The test sample is mixed with a small amount of Molisch's reagent (α -naphthol dissolved in ethanol), then the mixture is shaken well and a small amount of concentrated sulphuric acid is added slowly to the sides of the inclined test tube and allowed to stand to form a layer. A positive reaction of carbohydrates is indicated by the appearance of violet rings (Figs. 5c, 6d and 7d).

Fehling's test: Take the sample solution in a clean test tube and add Fehling's solution A and Fehling's solution B to it. Place the

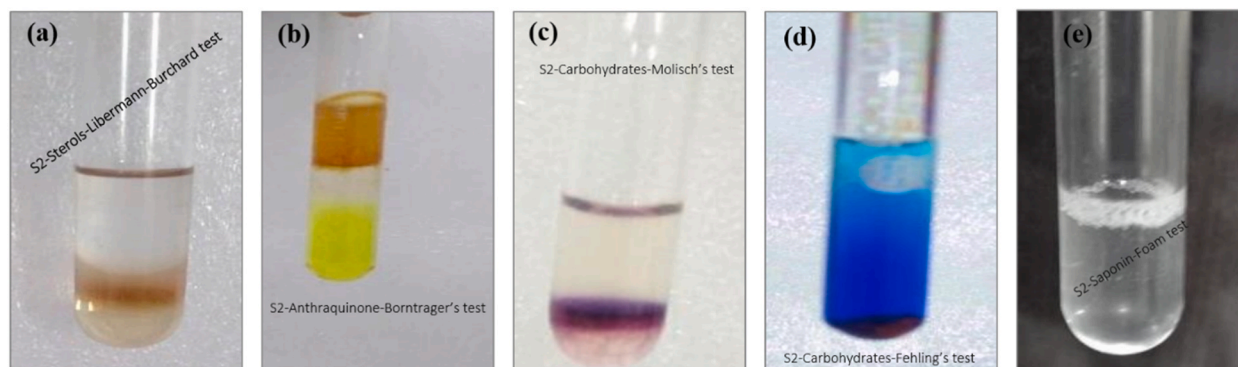


Fig. 5. Confirmation of the presence of metabolites for aqueous extract of SP seed powder.

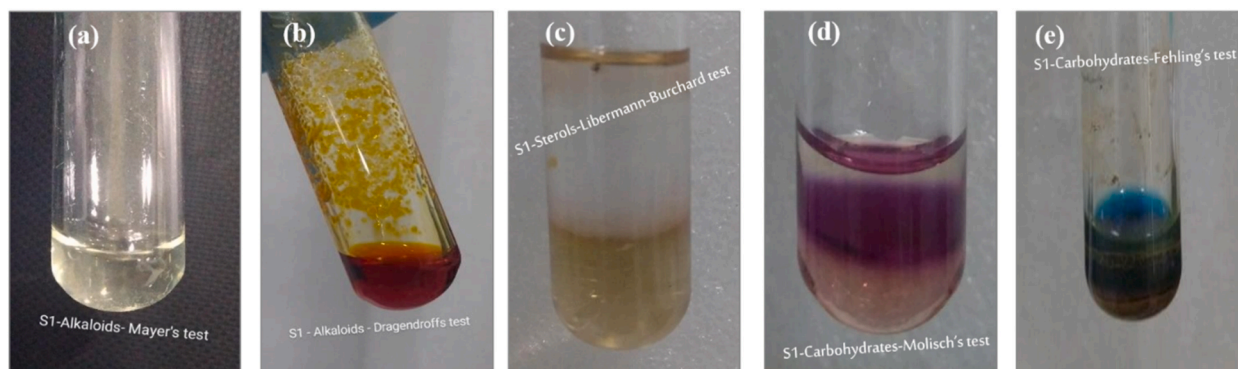


Fig. 6. Confirmation of the presence of metabolites for ethanol extract of SP seed powder.

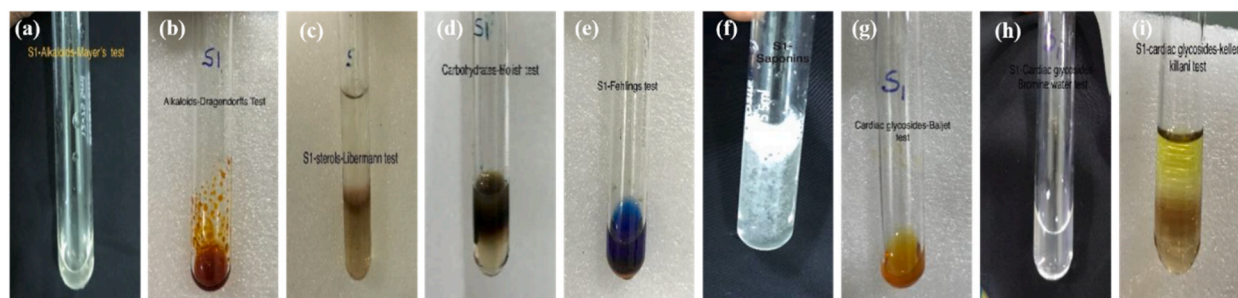


Fig. 7. Confirmation of the presence of metabolites for methanol extract of SP seed powder.

solution in a boiling water bath for about 10 min to observe the formation of a red precipitate confirming the presence of carbohydrates (Figs. 5d, 6e and 7e).

4.14. Test for tannin

Ferric chloride test: A quantity of extract was added with 10% alcoholic ferric chloride for the formation of greenish color, indicating the presence of tannins.

4.15. Test for saponin

Foam test: A small amount of the extract was diluted with distilled water. The mixture was shaken vigorously and then observed standing for the appearance of stable foam (Fig. 5e and 7f).

4.16. Test for cardiac glycosides

Baljet test: A few drops of Baljet's reagent (95 mL 1% picric acid + 5 mL 10% NaOH) are added to the extract, the formation of yellow orange colour indicates the presence of cardiac glycosides (Fig. 7g).

Bromine water test: The extract is added to bromine water, the formation of a yellow precipitate indicates a positive test for cardiac glycosides (Fig. 7h).

Keller-killani test: To the seed extract, 2 mL of glacial acetic acid and one drop of ferric chloride were added. A brown ring indicates the presence of cardiac glycosides (Fig. 7i).

4.17. Test for lignin

Labat test: When gallic acid is added to the extract, it produces an olive green colour.

4.18. Test for coumarins

NaOH test: To the extract, 3 mL of 10% NaOH and CHCl_3 were added to obtain a yellow colour.

Aqueous seed extracts of this species showed the presence of sterols, anthraquinone, carbohydrates, and saponin. Ethanolic seed

extracts of this species showed the presence of bioactive constituents of alkaloids, sterols, and carbohydrates. Methanolic seed extracts showed the presence of alkaloids, sterols, carbohydrates, saponins, and cardiac glycosides. Singh et al. (1975) revealed that the seeds of SP contain alkaloids mainly diabolin [43]. In order to remove the colloids from the colloidal suspension, plant parts with coagulant features, like alkaloids are utilised. Therefore, preliminary screening tests are helpful in determining bioactive components, which may result in discovery and development of drugs.

5. Conclusion

As reported in a considerable number of research articles, the present study also demonstrates the ability of natural coagulant to improve coagulation performance. The powdered seed of SP containing anionic polyelectrolytes has coagulant properties used for water quality testing which results to clarify turbid water. As a result, after treatment, this species showed excellent performance in removing turbidity. Conductivity and TDS levels increased slightly and showed minor role in PO_4^{3-} removal. There is no significant change in Ca^{2+} , Mg^{2+} and pH but a slight variation. Coagulation outcomes for the elimination of turbidity and PO_4^{3-} were found to be 81.54 and 41.45%, respectively. Functional groups such as carboxyl, amine and hydroxyl groups responsible for coagulation were identified by FT-IR spectra. The phytochemical elements found in plant materials exhibit coagulating capabilities which boost the efficiency of coagulation. The study comes to the conclusion that SP seed powder is a low-cost material with great promise for wastewater treatment. To obtain knowledge of turbidity removal, the amount of coagulant (concentration of SP seed powder) should be varied. For a detailed analysis of water quality, besides the found parameters, heavy metals, radio nuclides should also be analyzed. It is also advised to investigate the use of various coagulant extraction.

Author contribution statement

N L Sheeba - performed the experiments, analyzed and interpreted the data, wrote the paper; E Selva Esakki - contributed reagents, materials, analysis tools or data, analyzed and interpreted the data; R Sarathi - contributed reagents, materials, analysis tools or data, performed the experiments; A Esaiarasi - contributed reagents, materials, analysis tools or data, wrote the paper; S Meenakshi Sundar - conceived and designed the experiments, wrote the paper.

Data availability statement

The data that has been used is confidential.

Additional information

No additional information is available for this paper.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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