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The Potential Use of Basil Seeds as a Biocoagulant in Reducing Water Turbidity

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ABSTRACT

Background: The excessive use of chemical coagulants such as alum can cause health problems including tissue damage, detoxification issues, kidney excretion problems, and nervous system disorders. Research has shown that the consumption of alum at a dose of 800 mg/kg body weight per day can cause cell degeneration in Wistar rats. **Methods:** The research method used is an experiment with a pre-post test control group design. The subject studied was basil seeds used as a bio-coagulant in pond water at Poltekkes Surabaya. The dose variations used in this study were 0.5 ml, 1 ml, 1.5 ml, and 2 ml. Data were analyzed using the Kruskal-Wallis test to examine differences in water turbidity levels. **Results**. Based on the results, the use of basil seed bio-coagulant increased turbidity values with an average of 14.52 NTU for the control, 23.35 NTU for the 0.5 ml dose, 27.95 NTU for the 1 ml dose, 31.6 NTU for the 1.5 ml dose, and 29.4 NTU for the 2 ml dose. **Conclusion:** Based on the results obtained, it is known that no confidence was gained in the use of basil seeds as a bio-coagulant to reduce turbidity.

Keywords: Basil Seed, Biocoagulant, Turbidity

BACKGROUND

Water resources represent one of the most crucial components for the sustainability of life on Earth. Water not only serves as the primary medium for life among living organisms but also plays a decisive role in social, demographic, and economic dynamics (Fina Fakhriyah, 2021). Ecologically, water provides habitats for diverse organisms and acts as a vital medium in the photosynthetic processes of plants, forming the foundation of food chains. Although water is a naturally renewable resource, its availability does not always guarantee quality sufficient to meet human and ecological needs. Therefore, maintaining both water quality and availability is of paramount importance for public health and environmental sustainability (Fina Fakhriyah, 2021).

However, increasing population, urbanization, and industrial activities have led to significant degradation of water quality. Contaminated water can serve as a vector for various diseases, especially gastrointestinal infections such as diarrhea and cholera, which may lead to fatalities—particularly among vulnerable populations (Trisna, 2018). Turbidity, caused by the accumulation of suspended particles and colloids, stands out as one of the most critical indicators of water pollution requiring urgent attention to improve the quality of drinking water. Consequently, water clarification has become a primary focus in sustainable water resource management.

Coagulation-flocculation is one of the most widely used and effective techniques for addressing water turbidity.

This process involves the addition of coagulants capable of neutralizing the surface charges of colloidal particles, enabling previously stable particles to aggregate into larger, settleable flocs (Febrianti et al., 2024). Coagulants may be synthetic chemicals or natural substances (biocoagulants). In practice, aluminum sulfate (alum) remains the most commonly used chemical coagulant. However, concerns about heavy metal residues and environmental side effects have spurred interest in more environmentally friendly alternatives—particularly biocoagulants derived from natural sources, such as basil seeds, moringa seeds, and chitin/chitosan (Febrina, 2019).

The use of biocoagulants is increasingly gaining attention due to their biodegradability, sustainability, and promising efficacy comparable to synthetic coagulants. Basil seeds (*Ocimum basilicum* L.), in particular, naturally contain natural polymers that function as effective natural coagulants in reducing water turbidity. Beyond their environmental benefits, biocoagulants are typically more affordable and accessible from local resources (Febrianti et al., 2024).

Limited access to clean water significantly impacts various aspects of life, especially public health. Poor water quality increases the risk of infectious diseases that threaten community well-being. Regions with restricted access to clean water require low-cost, easily implementable solutions—such as the use of natural biocoagulants—to enhance drinking water quality. Nevertheless, further research and validation are necessary to confirm the effectiveness of biocoagulants derived from basil seeds as a natural coagulant alternative for optimal turbidity reduction.

RESEARCH METHODS

This study aims to determine the effectiveness of biocoagulants derived from basil seeds in reducing water turbidity. Different dosages of the coagulant are tested to identify the optimal dosage that yields water quality meeting regulatory standards. Thus, this research is expected to contribute significantly to the development of environmentally friendly, scalable, and sustainable natural water treatment technologies.

The findings of this study offer valuable contributions to scientific knowledge, particularly in the field of environmental sanitation, by providing an effective and affordable alternative water treatment method. Moreover, the application of basil seed biocoagulants can enhance community self-reliance in managing local water resources sustainably and reduce dependence on hazardous chemical coagulants in water treatment processes.

Basil seeds possess hydrophilic properties, meaning they can absorb water effectively. These seeds contain a significant amount of hemicellulose and cellulose. When soaked in water, their outer pericarp expands into an attractive gelatinous mass, due to the presence of a polysaccharide layer that coats the seeds. The polysaccharides in basil seeds contain hydroxyl (-OH) groups that can interact with colloidal particles through hydrogen bonding and electrostatic interactions, thereby facilitating the coagulation and flocculation processes (Zhao *et al.*, 2025).

This study employs a pure experimental design using a pretest-posttest control group design. This design enables the researcher to observe changes in measurements before and after the intervention, while also incorporating a control group to compare the outcomes of the treatment.

The study was conducted at the Environmental Health Laboratory of the Health Polytechnic of the Ministry of Health Surabaya and the Agency for Standardization and Industrial Service Policy. The research took place from January to June 2025. The main focus of this study was the use of a natural biocoagulant derived from basil seeds (*Ocimum basilicum* L.) to reduce water turbidity.

1. Preparation of Basil Seed Biocoagulant
   a. Basil seeds were ground using a blender until a fine powder was obtained.
   b. The basil seed powder was then sieved.
   c. The prepared basil seed powder was added to a NaCl solution at a ratio of 1:10.
   d. The mixture was stirred for 30 minutes.
   e. Residual solids were separated using a filter.
   f. The basil seed coagulant was ready to be used as a biocoagulant.

2. Coagulation-Flocculation Using the Jar Test Method (SNI19-6449-200)
   a. Six beakers were prepared.
   b. Water samples were poured into each beaker.
   c. The coagulant was added to each beaker according to the experimental requirements.
   d. The impeller speed was set to a rapid mix (150 rpm) for 60 seconds.
   e. The impeller speed was then reduced to a slow mix (60 rpm) for 30 minutes.
   f. The mixer was turned off and the stirring rods were lifted simultaneously.

The turbidity data obtained were analyzed quantitatively to determine the effectiveness of basil seed biocoagulant dosages in reducing water turbidity. This study received ethical approval from the Health Polytechnic of the Ministry of Health under approval number.

RESULTS AND DISCUSSION

The measurement of water turbidity using basil seed biocoagulant was carried out at the Laboratory of the Agency for Standardization and Industrial Service Policy, Ministry of Industry. The turbidity level was measured using the Nephelometric method with biocoagulant doses of 0.5 mL, 1 mL, 1.5 mL, and 2 mL.

Table 1.

Water Turbidity with 0.5mL Dose of Basil Seed Biocoagulant

Dose	**Water Turbidity in the Repeat Test**				**Average**
	I	**II**	**III**	**IV**	
Control	15,4	12,3	15,3	15,1	14,52
0,5 ml	23,7	22,8	22,6	24,3	23,35

In Table 1 an increase in turbidity was observed with the use of basil seed biocoagulant at a dose of 0.5 mL. The turbidity increased in each repetition of the 0.5 mL dose, with the average turbidity value rising from 14.52 NTU to 23.35 NTU, representing a 60.81% increase in turbidity.

Table 2.

Water Turbidity with 1mL Dose of Basil Seed Biocoagulant

Dose	**Water Turbidity in the Repeat Test**				**Average**
	I	**II**	**III**	**IV**	
Control	15,4	12,3	15,3	15,1	14,52
1 ml	28,3	28,2	28,2	27,4	27,95

Based on the results presented in Table 2, an increase in turbidity was observed with the use of basil seed biocoagulant at a dose of 1 mL. The turbidity increased in each repetition of the 1 mL dose, with the average turbidity value rising from 14.52 NTU to 27.95 NTU, representing a 92.49% increase in turbidity.

Table 3.

Water Turbidity with 1,5mL Dose of Basil Seed Biocoagulant

Dose	Water Turbidity in the Repeat Test				Average
	I	II	III	IV	
Control	15,4	12,3	15,3	15,1	14,52
1 ml	38,3	35,2	34,2	18,7	31,6

In Table 3, an increase in turbidity was observed with the use of basil seed biocoagulant at a dose of 1.5 mL. The turbidity consistently increased in each repetition of the 1.5 mL dose, with the average turbidity value rising from 14.52 NTU to 31.6 NTU, representing a 117.79% increase in turbidity.

Table 4.

Water Turbidity with 2mL Dose of Basil Seed Biocoagulant

Dose	Water Turbidity in the Repeat Test				Average
	I	II	III	IV	
Control	15,4	12,3	15,3	15,1	14,52
2 ml	17,7	41,7	18,5	37,5	29,4

Based on the results presented in Table 4, an increase in turbidity was observed with the use of basil seed biocoagulant at a dose of 2 mL. The turbidity increased consistently in each repetition of the 2 mL dose, with the average turbidity value rising from 14.52 NTU to 29.4 NTU, representing a 102.47% increase in turbidity

Table 5.

Analysis of Turbidity Changes Using Basil Seed Biocoagulant at Doses of 0.5mL, 1mL, 1,5mL, and 2mL

Code of Conduct	Dose	Water Turbidity in the Repeat Test				Average
		I	II	III	IV	
Ar	0,5	23,7	22,8	22,6	24,3	23,35
Br	1	28,2	27,9	27,4	28,3	27,95
Cr	1,5	34,2	38,3	18,7	35,2	31,6
Dr	2	17,7	41,2	18,5	37,7	29,4

Based on the results presented in Table 5, an overall increase in turbidity was observed compared to the control values. Although turbidity increased at all applied biocoagulant doses, a decrease in turbidity was also noted at certain repetitions within specific doses. At the 1.5 mL dose, the third repetition showed a reduction in turbidity with a value of 18.7 NTU, whereas the other repetitions at the same dose ranged between 30 and 38 NTU. A similar decrease was observed at the 2 mL dose in the first and third repetitions, with turbidity values of 17.7 NTU and 18.5 NTU, respectively, compared to the second and fourth repetitions, which recorded 41.7 NTU and 37.5 NTU, representing a 6.96% decrease in turbidity.

To determine whether basil seed biocoagulant produced a significant difference in turbidity levels across different dosage variations, a statistical analysis was performed. The analysis employed One- Way ANOVA when the data were normally distributed and the Kruskal–Wallis test when the data were not normally distributed. A Post Hoc test was subsequently conducted to determine whether significant differences existed among more than two groups.

Based on the normality test results, the control variable showed a p-value < 0.05, indicating that the data were not normally distributed. Therefore, the Kruskal–Wallis test was performed, yielding a significance value ($p \leq 0.028$), which indicates a difference among the dosage variables used. To further identify whether differences existed between individual dosage groups, the Mann–Whitney test was conducted, resulting in a p-value > 0.05, suggesting that there was no significant difference among the individual biocoagulant dosages tested.

DISSCUSION

Based on the results obtained, the protein, polysaccharide, and flavonoid contents in basil seeds were found to be ineffective as biocoagulants in reducing water turbidity. This may be due to the limited binding ability of these components to form stable flocs. Therefore, when using basil seeds as a biocoagulant, it is essential to consider the polysaccharide and flavonoid content to determine the optimal dosage required for effective coagulation.

Flavonoids, a group of phenolic compounds abundantly present in basil seeds and many other plants, operate through distinct yet complementary mechanisms. Compounds such as quercetin and kaempferol can interact with hydroxyl and carbonyl groups on particle surfaces, enhancing floc stability through hydrogen bonding and van der Waals interactions. According to research conducted by (Lwasa et al., 2024) on *Moringa oleifera* extract, flavonoids contributed approximately 23% of the total coagulation efficacy.

Similarly, a study by D et al., (2021) reported that the molecular structure of polysaccharides— composed of repeating monosaccharide units—enables them to function as multifunctional biocoagulants, primarily through charge neutralization and the bridging effect. At neutral pH, the hydroxyl (-OH) and carboxyl (-COOH) groups in polysaccharide chains, such as modified cellulose, form stable complexes with negatively charged colloidal particles, reducing the zeta potential by 15–20 mV within 5 minutes.

Based on the results of using basil seeds as a biocoagulant, an increase in turbidity was observed compared to the control. Turbidity levels increased across all applied doses, with turbidity values recorded as 14.52 NTU for the control, 23.35 NTU for 0.5 mL, 27.95 NTU for 1 mL, 31.6 NTU for 1.5 mL, and 29.4 NTU for 2 mL. The increase in turbidity observed with the use of basil seed biocoagulant may be attributed to the characteristics of the raw water used as the sample. One of the key factors influencing the performance of the biocoagulant is the colloidal nature of the raw water. Colloids play a central role in water treatment processes through electrochemical destabilization mechanisms. The structure of natural water colloids can generally be classified into two main categories: organic colloids (such as humic substances and microbial exopolymers) and inorganic colloids (such as silicate clays or iron oxides).

Colloids in water are defined as dispersed systems of particles ranging from 1 to 1000 nanometers, which are not completely soluble yet remain suspended in the liquid medium due to surface charge and Brownian motion. These unique characteristics make colloidal particles difficult to settle naturally, thereby constituting the main source of turbidity in surface waters such as rivers and lakes. Colloidal particles are typically amphiphilic, with negatively charged surfaces resulting from ionized carboxyl or hydroxyl groups, forming an electric double layer that prevents aggregation.

According to the study conducted by Worms et al., (2010), natural polysaccharides such as chitosan derived from basil seeds have been shown to form hydrogen bridges with –OH groups on the surface of clay colloids. Fourier-transform infrared spectroscopy (FTIR) analysis revealed a shift in the absorption band from 3400 cm^{-1} to 3200 cm^{-1}, indicating a strong interaction between the hydroxyl groups of the polysaccharide and the colloidal surface.

In general, compared with the control, turbidity levels increased with the use of basil seed biocoagulant; however, the 2 mL dose exhibited a decrease in turbidity relative to the 1.5 mL dose (from 31.6 NTU to 29.4 NTU). The increase in turbidity may have been influenced by several factors, including the use of NaCl as a solvent, the biocoagulant dosage, and the characteristics of the raw water, such as pH, temperature, and organic matter content.

NaCl, as a solvent, plays a crucial role in the chemical interactions between proteins, sodium chloride, and other seed components. NaCl increases the ionic strength of the solution, which can weaken intermolecular interactions among proteins, facilitating their release. Furthermore, NaCl influences the "salting-in" and "salting-out" phenomena. At low concentrations, NaCl enhances protein solubility (salting-in), whereas at higher concentrations, it causes protein precipitation (salting-out), aiding the coagulation process necessary for biocoagulant formation.

At relatively low salt concentrations, salting-in occurs as NaCl ions interact with protein and water molecules, reducing intermolecular attractions that cause aggregation and allowing proteins to remain more soluble. Conversely, at higher salt concentrations, NaCl ions compete with proteins for water molecules, reducing hydration and leading to protein precipitation due to restored intermolecular attractions.

This observation aligns with the findings of Zhang et al., (2025), who reported that salting-in occurs below 50 mM NaCl, where hydrated Na^+ ions form loose solvation layers around proteins, reducing aggregation. For example, pea legumin protein exhibited a 1.8-fold increase in solubility at 30 mM NaCl compared to salt-free conditions. On the other hand, N.A. Deak, P.A. Murphy, and L.A. Johnson (2006) reported that salting-out becomes dominant above 100 mM NaCl, as Cl^- ions withdraw water from the protein hydration shell, leading to destabilization and precipitation. Their study on soybean protein fractionation showed a 60% reduction in β-conglycinin solubility as NaCl concentration increased from 50 mM to 200 mM.

The performance of NaCl as a solvent is highly dependent on the type of protein present. Hydrophobic proteins, such as corn zein, tend to precipitate even at low NaCl concentrations due to enhanced hydrophobic interactions induced by Cl^- ions. In contrast, hydrophilic proteins, such as legumin from legumes, remain more stable in saline solutions. The distribution of Na^+ and Cl^- ions in grain solutions is influenced by the surface charge of specific proteins. For instance, glutenin in wheat, rich in glutamine groups, forms stable dipole–ion complexes with Na^+, reducing free ion availability for starch solubilization. Rheological studies have shown a 40% reduction in NaCl ionic activity in 5% gluten solutions compared to pure water at 0.1 M NaCl. Conversely, corn albumin proteins, which contain abundant aspartate residues, promote NaCl dissociation by forming weak complexes with Cl^-, resulting in a 25% increase in electrical conductivity under similar conditions.

The biocoagulant dosage plays a critical role in determining the effectiveness of turbidity removal. An optimal dose enhances the sedimentation of suspended particles, thereby significantly reducing turbidity. Proteins act as biological macromolecules capable of binding suspended particles via electrostatic interactions and molecular bridging. They contain functional groups such as amino and carboxyl groups that neutralize negatively charged colloids, facilitating floc formation and aggregation into larger, settleable particles. The advantages of protein-based biocoagulants include biodegradability, environmental friendliness, and renewability. Proteins are particularly effective against colloid-induced turbidity through adsorption, charge neutralization, and bridging mechanisms, which enhance coagulation–flocculation efficiency in water treatment.

Colloidal particles, stabilized by their surface charge and electrical double layer (EDL), resist aggregation. The EDL comprises a compact layer of ions with the same charge as the colloidal particle and a diffuse layer of oppositely charged ions, conferring additional stability. Colloids also exhibit the Tyndall effect, scattering light that passes through, which accounts for the cloudy appearance of turbid water. Their high surface area allows significant chemical and physical interactions, including adsorption of dissolved substances, pollutant transport, and floc formation with biocoagulants. However, the relationship between dosage and turbidity reduction is not linear — exceeding the optimal dose can reduce effectiveness or cause restabilization of colloids.

This observation aligns with Fakhara Shahzadi (2024), who emphasized that selecting an optimal dosage is crucial for maximizing turbidity removal while maintaining material efficiency. Her findings on sorghum-based biocoagulants demonstrated that proper dosage enables efficient natural material use, minimizing the need for synthetic chemicals and promoting sustainable, eco-friendly water treatment. Similarly, Mukheled Al-Sameraiy (2024) stated that the strategic use of natural biocoagulants can significantly reduce chemical input, processing time, and improve treated water quality. Underdosing results in ineffective coagulation, while overdosing wastes material and may increase residuals or lower water quality.

The effectiveness of a biocoagulant is also influenced by the characteristics of the raw water. Parameters such as pH, temperature, and organic matter, especially humic and fulvic substances from natural decomposition, can interfere with coagulation–flocculation mechanisms. These organic compounds may form a stable coating around colloidal particles, preventing effective biocoagulant binding. Consequently, the formed flocs are weak and easily redispersed, limiting turbidity reduction.

Hydrophobic interactions also play an important role in the coagulation mechanism. Basil seed proteins contain hydrophobic domains that can interact with non-polar contaminants or hydrophobic regions of particle surfaces, promoting particle aggregation. Their amphiphilic nature allows proteins to act as linking agents, binding diverse particles into larger flocs. The mucilaginous polysaccharide matrix associated with basil seed proteins enhances this mechanism by increasing solution viscosity and creating a gel-like environment that traps suspended solids.

However, pH strongly affects protein stability and function. Deviations from the optimal pH can cause protein denaturation, disrupting secondary, tertiary, and quaternary structures without breaking peptide bonds. This leads to loss of biological function and reduced coagulation activity. At alkaline pH, floc disintegration and reduced removal of compounds such as anionic surfactants and chromate ions occur, diminishing charge neutralization and electrostatic attraction.

Temperature also has a close relationship with protein stability. Hydrogen bonds, crucial for maintaining protein structure, are highly sensitive to temperature changes. Elevated temperatures can weaken these bonds, causing denaturation and reduced coagulation efficiency. Kurniawan et al., (2020) reported that at high temperatures (above 40–60°C), biocoagulant proteins lose structural integrity, leading to drastic decreases in flocculation efficiency, while low temperatures slow down molecular motion, reducing aggregation and floc formation.

Similarly, Abdullahi Sanda Ndayako et al. (2023) reported that heavy metals and organic pollutants interfere with biocoagulant binding sites, competing for active sites or altering surface charges. These interactions hinder floc formation and reduce turbidity removal efficiency. A. Lwasa (2024) further noted that heavy metals such as lead, cadmium, and chromium can strongly bind to hydroxyl, carboxyl, and amine groups of biocoagulant molecules, blocking active sites and inhibiting particle aggregation. In addition, Setyo Budi Kurniawan et al. (2023) highlighted that pretreatment and preparation methods of plant-based biocoagulants significantly influence their chemical composition, physicochemical properties, and overall performance in water treatment.

Bioactive compounds such as glycosides, phenolics, tannins, flavonoids, alkaloids, and proteins are central to coagulation–flocculation processes. These compounds contain hydroxyl, carboxyl, and amine groups that interact with suspended particles and contaminants. Solvent extraction can selectively enrich phenolic and flavonoid content, improving charge neutralization and floc formation, while pretreatment can modify surface chemistry and enhance adsorption and bridging capabilities.

Therefore, in applying biocoagulants, it is essential to adjust the dosage and consider the characteristics of the raw water. The biocoagulant dose should correspond to the initial turbidity level and the specific type of biocoagulant used. Insufficient dosage may fail to neutralize colloidal particles, while excessive dosage may lead to floc overaccumulation, resulting in restabilization or ineffective turbidity reduction. Based on the results obtained, the use of basil seed (Ocimum basilicum) as a biocoagulant did not show a significant reduction in turbidity levels; instead, an increase in turbidity was observed across all tested dosages (0.5 mL, 1 mL, 1.5 mL, and 2 mL) compared to the control. This increase may be attributed to factors such as the physicochemical characteristics of the raw water (including pH, temperature, and organic content), the ionic strength introduced by NaCl as a solvent, and the molecular behavior of the proteins, polysaccharides, and flavonoids present in basil seeds.

CONCLUSION

The results suggest that the protein–polysaccharide–flavonoid complex in basil seeds lacks sufficient coagulative interaction potential under the experimental conditions used. Variations in dosage did not produce statistically significant differences in turbidity reduction ($p > 0.05$), indicating that the active compounds in basil seeds may not be effectively optimized for floc formation in this system.

Furthermore, environmental factors such as pH and temperature strongly influence protein stability and denaturation, which in turn affect coagulation efficiency. The findings emphasize the necessity of optimizing both the extraction and pretreatment processes of basil seed biocoagulants, as well as the adjustment of dosage and environmental conditions, to enhance their coagulative efficacy in water treatment applications.

Overall, while basil seeds contain bioactive compounds with potential coagulation properties, their current performance under the tested parameters demonstrates limited effectiveness. Further research is recommended to refine extraction techniques, explore synergistic agents, and determine optimal operating conditions to realize the potential of basil seeds as sustainable natural biocoagulants.

CONFLICT OF INTEREST

The authors declare no conflicts of interest regarding this manuscript.

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