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Application of Chitosan from *Litopenaeus vannamei* and Baglog Waste from *Pleurotus ostreatus* for Decolorizing Batik Wastewater

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Abstract: The batik sector generates wastewater containing pigment residue, posing a significant environmental threat if not managed effectively. *L. vannamei* shells yield chitin, convertible to chitosan for water treatment. Baglogs, comprising mycelium and cellulose, serve as a viable adsorbent. The purpose of this study was to learn about the use shell of *L. vannamei* and *P. ostreatus* baglog waste with different lengths of time to decolorization batik industry waste water. The method for testing involves deproteinization with NaOH 1M, demineralization with HCl 2M and deasetylation with NaOH 60%. Chitosan *L. vannamei* and baglog *P. ostreatus* (2:25 g/g) waste were put into a container containing 500 mL of batik wastewater with colour content with variations of 0, 12, 24, 48, 72, 96, and 120 hours. Further, the capacity of adsorption chitosan and baglog waste are determined using a UV-Vis spectrophotometer during the decolorization process in batik industry waste water using shell of *L. vannamei* and *P. ostreatus* baglog waste are 96 hours with a decolorization rate of 90.05%.

Keywords: Baglog waste *P. ostreatus*; Batik wastewater; Chitosan *L. vannamei*; Decolorization

Introduction

The increasing demand for batik has prompted manufacturers to scale up production, resulting in greater environmental consequences. In Indonesia, the majority of batik production occurs without adequate industrial waste treatment facilities, leading to the discharge of untreated effluents into rivers or drainage systems. This absence of proper treatment facilities compels the batik industry to often release untreated effluents into specialized vessels or directly into water bodies, presenting a significant sectoral challenge. Consequently, the untreated disposal of batik wastewater gives rise to diverse environmental concerns (Triwiswara, 2019).

Batik wastewater contains hazardous components originating from synthetic dyes used in dyeing and washing processes. Synthetic dyes, known for their vivid colors, ease of application, and cost-effectiveness, are resistant to degradation in water, rendering untreated batik wastewater a significant environmental concern (Apriyani, 2018). These dyes, such as indigosol and naphthol dyes, are commonly utilized in the batik production process (Yanto et al., 2021).

The color concentration in batik wastewater is frequently elevated, necessitating further treatment before its release into the environment (Apriyani, 2018). The production of batik relies on dyes derived from synthetic sources that contain heavy metals, which are recognized as one of the most perilous pollutants, capable of posing significant risks to both humans and the environment. It is suggested that the higher the color level of the waste, the higher the concentration of hazardous heavy metals it may contain, thus posing an environmental threat (Astuti et al., 2023). As per Minister of Environment Regulation No. 16 of 2019, the allowable level of color in textile wastewater discharged

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into the environment is approximately 200 Pt-Co units. Numerous research studies have explored methods to remove color from batik wastewater, and one of the most commonly employed techniques is adsorption. Adsorption is widely favored in physicochemical wastewater treatment due to its safety, lack of adverse health effects, simplicity, cost-effectiveness, and recyclability. This method effectively eliminates colorants from the waste stream, making it a reliable choice for addressing this environmental concern (Zakaria et al., 2023).

Chitosan is employed as an adsorbent for textile dye removal due to its environmentally friendly nature, abundant availability, biodegradable characteristics, and cost-effectiveness (Sarode et al., 2018). Chitosan can be synthesized from the biopolymer chitin found in crustaceans such as lobsters, crabs, and shrimp. Shrimp production in Indonesia has been on the rise because, typically, only the meat is consumed, while the shells, tails, and heads are discarded without processing, potentially causing environmental pollution. The shells of whiteleg shrimp (*L. vannamei*) contain 25-40% protein, 45-50% calcium carbonate, and 15-30% chitin (Imtihani & Permatasari, 2020). Chitin and chitosan can both be used as adsorbents, but chitin is less effective because the acetylamide groups ($NHCOCH_3$) in chitin are weaker ligands compared to the amine groups (NH_3) in chitosan, which serve as strong ligands. Therefore, chitosan is more efficient in adsorption processes than chitin (Saheed et al., 2020). Chitosan indeed has the capability to absorb textile dyes such as methylene blue up to 85.05% (Iget et al., 2019) until 90% (Nurlaili et al., 2022) and remazol red up to 100% (Safitri & Rahmayanti, 2020).

White oyster mushrooms (*P. ostreatus*) generate solid waste in the form of mushroom growing media or baglogs during their production process. Baglog waste originating from mushroom cultivation has the potential to pollute the environment and has not been fully utilized. So far, baglog waste has been repurposed into organic compost, contributing to environmental cleanliness and serving as a natural fertilizer for plants. However, due to its slow decomposition, additional organic materials and bio activators are necessary to expedite the composting process (Susilowati et al., 2023). To maximize the utility of baglog waste, it can be employed as a decolorizing agent in treating batik wastewater. This is feasible because baglog waste retains mushroom mycelium and cellulose, which can effectively adsorb dye substances present in batik wastewater (Dewi et al., 2023). Baglog waste from *P. ostreatus* is one of the effective materials for decolorizing batik wastewater, achieving a decolorization rate of 85.64% over 60 hours (Wulandari et al., 2014).

The main goal of this research is to explore the decolorization of wastewater from the batik industry using chitosan from *L. vannamei* and baglog waste from *P. ostreatus*, with a focus on assessing their effectiveness at different time intervals. Additionally, the study aims to determine the optimal adsorption duration for both chitosan derived from *L. vannamei* and baglog waste from *P. ostreatus* in the context of decolorizing batik industry wastewater.

Method

The research utilized an experimental approach with authentic experiments. The research procedure follows the flowchart depicted in Figure 1.

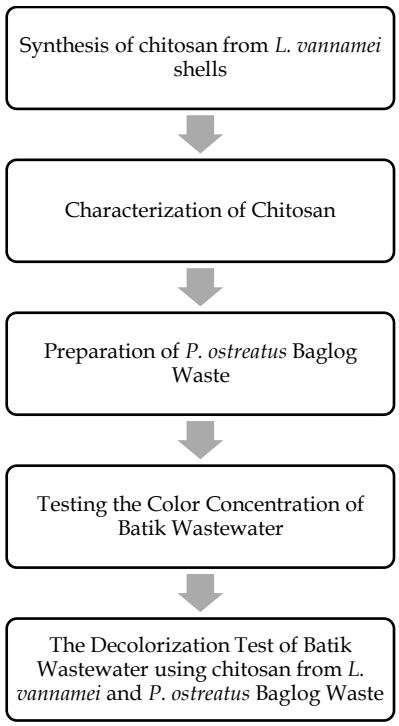

Figure 1. Research flowchart

The materials used in this study include the shells of *L. vannamei*, baglog waste from *P. ostreatus*, and wastewater from the batik industry. For the production and characterization of chitosan, the following materials were used: sodium hydroxide, hydrochloric acid, ninhydrin solution, glacial acetic acid, biuret reagent, and distilled water. The research utilized various laboratory equipment, such as glassware, analytical balances, filter paper, pH meters, ovens, blenders, funnels, reflux apparatus, FTIR spectrophotometer for chitosan characterization, and UV-Vis spectrophotometer for determining the color values in the batik industry wastewater.

Synthesis of chitosan from L. vannamei shells
Deproteinization

Deproteinization is carried out to remove the proteins present in the *L. vannamei* shells. Finely crushed *L. vannamei* shells are mixed with a 1M NaOH solution at a ratio of 1:10 (g/mL). The mixture is stirred for one hour, refluxed for four hours at a temperature of 100°C, then cooled and filtered. The residue obtained from filtration is rinsed with distilled water until it reaches a neutral pH. Afterward, it is dried in an oven at 60°C, and the fine powder is qualitatively tested using a biuret test. The protein-free fine powder is then weighed.

Demineralization

The demineralization process aims to eliminate minerals present in the *L. vannamei* shells. Finely ground *L. vannamei* shells are mixed with a 2M HCl solution at a ratio of 1:10 (g/mL) and stirred for an hour. The mixture is then filtered, and the resulting residue is rinsed with distilled water until it reaches a neutral pH. After rinsing, the residue is dried in an oven at 60°C. The outcome of this phase is chitin, which is subsequently characterized using a FTIR Spectrophotometer to analyze the functional groups of chitins.

Deacetylation

Deacetylation is performed to remove the acetyl groups ($NHCOCH_3$) from chitin, converting them into amine groups (NH_2) in chitosan. The chitin powder is combined with a 60% NaOH solution at a ratio of 1:10 (g/mL) and refluxed for two hours. The mixture is then cooled and filtered, with the residue being rinsed until it achieves a neutral pH. After rinsing, the residue is dried in an oven at 60°C. The output of this stage is chitosan, which is subsequently characterized using a FTIR Spectrophotometer to analyze the functional groups of chitosan.

Characterization of Chitosan
Chitosan Solubility Test

One gram of chitosan is added to 100 ml of a 2% glacial acetic acid solution. If the solubility of chitosan results in a solution with the same color as the solvent, it meets the maximum standard for solubility.

Ninhydrin Test

One gram of chitosan is sprayed with a ninhydrin solution and left for 5 minutes. A positive reaction is indicated by the appearance of a purple color. Ninhydrin is a strong oxidizing agent that reacts with the amine groups (NH_2) in chitosan.

Moisture Content Test

Two grams of chitosan are placed in an empty dish with a known weight, dried in an oven at 100-105°C for 3 hours, and then placed in a desiccator until a constant weight is achieved. The acceptable moisture content for chitosan is ≤ 10%.

Ash Content Test

Two grams of chitosan are placed into a clean crucible that has been preheated. The chitosan is then heated to a temperature of 600-650°C. After heating, the crucible is placed in a desiccator until a constant weight is achieved. The acceptable ash content for chitosan is ≤ 2%.

Preparation of P. ostreatus Baglog Waste

The *P. ostreatus* baglog waste used in this study is sourced from baglogs that have undergone 2-5 harvest cycles. The baglog waste is then finely ground into a powder.

Testing the Color Concentration of Batik Wastewater

The determination of the color concentration in batik wastewater is carried out using a UV-Vis spectrophotometer. The batik wastewater is first prepared as a stock solution, which is then diluted to create several concentration series. The maximum wavelength is determined, and for each concentration series, the absorbance is measured at specific wavelengths. The absorbance values are then plotted in Excel to create a calibration curve, allowing for the determination of the color concentration, expressed as Pt-Co units (Platinum-Cobalt scale). Decolorization was monitored by a spectrophotometer and expressed as the percentage of removed dye. Percentage of decolorization was calculated as follows equation (1) (Pratiwi et al., 2017).

$$\text{Decolorization (\%)} = \frac{(\text{Abs}_0 - \text{Abs}_t)}{\text{Abs}_0} \times 100 \qquad (1)$$

Where the absorbance at time 0 is Abs_0 and Abs_t is the absorbance at time t, and λmax is measured at the maximum visible wavelength of dye.

The Decolorization Test of Batik Wastewater using chitosan from L. vannamei and P. ostreatus Baglog Waste

For the testing of decolorization of batik waste, chitosan, chitosan from *L. vannamei* and *P. ostreatus* baglog waste (in a ratio of 2:25, g/g) are introduced into a container containing 500 mL of batik industry wastewater with a known color concentration. The mixture is left to stand for various durations: 0, 24, 48, 72, 96, and 120 hours. The results of decolorization for each time variation are determined by measuring the color concentration of the batik industry wastewater using a UV-Vis spectrophotometer.

Result and Discussion

The Synthesis of Chitosan from L. vannamei Shells

The synthesis of chitosan from *L. vannamei* shells involves three stages, namely deproteinization, demineralization, and deacetylation. The deproteinization stage aims to remove the proteins present in L. vannamei shells. This is achieved by using a strong base, NaOH 1M, and heating it to 100°C for 6 hours. The proteins will react with Na+ ions to form soluble Na+ proteinates in water. The Na+ ions bind to the negatively charged ends of the protein chains, causing them to precipitate (Dompeipen et al., 2016).

In this research, the deproteinization yield obtained was 60.09% (Table 1), which is quite high compared to the deproteinization yield from tiger shrimp (*Penaeus monodon*), which is 42% (Jaya et al., 2017). The deproteinization yield value from *L. vannamei* shells in another study was 29.96% (Mittal et al., 2021). Qualitative testing of protein-free samples was conducted using the biuret reagent to determine whether the samples still contained proteins or if they had been removed. A positive reaction between the sample and the biuret reagent results in a purple color formation. This color change occurs because the peptide bonds in the proteins form Cu^{+} ion complexes with the biuret reagent, leading to a positive reaction (Li et al., 2020). In this study, the sample showed a negative reaction as it did not exhibit a purple color, indicating that the protein content in the sample had been eliminated.

Table 1. The results of the chitosan synthesis stages in *L. vannamei*

Treatment Stage	Materials	Result (g)	Yield (%)
L. vannamei shell	-	500	-
Deproteinization	-	300.45	60.09
Demineralization	Chitin	250.35	83.32
Deacetylation	Chitosan	110.6	44.19

The protein-free sample was then subjected to the demineralization stage to remove minerals or inorganic salts using a 2M HCl solution. Minerals can dissolve in HCl, allowing them to be separated. The separation process is indicated by the formation of CO_2 gas in the form of air bubbles when HCl is added to the sample (Borja-Urzola et al., 2020).

The protein-free and mineral-free sample consists of chitin. The chitin obtained in this study has a fairly high yield of 83% (Table 1), when compared to the chitin yield from tiger shrimp (*Penaeus monodon*), which is 47% (Jaya et al., 2017). In another study, the chitin yield from *L. vannamei* shells was 47.5%. The formed chitin was characterized using an FTIR spectrophotometer to determine the functional groups present in chitin. The FTIR spectrum of chitin shows peaks at 3255 cm^{-1} due to the stretching vibration of O-H groups. Absorption bands at 1069 cm^{-1} and 1010 cm^{-1} indicate stretching vibrations of C-O groups. Absorption bands at 2931 cm^{-1} and 2877 cm^{-1} correspond to CH_2 and CH_3 groups. The presence of CH_3 groups bound to amide ($NHCOCH_3$) is supported by the absorption band at 1418 cm^{-1}. Peaks at 1619 cm^{-1} and 1653 cm^{-1} represent stretching vibrations of C=O groups, and the bending vibration of the NH amide is located at 1552 cm^{-1}.

The chitin produced is subsequently treated with 60% NaOH to eliminate the acetyl groups ($NHCOCH_3$) in chitin, converting them into amino groups (NH_2) in chitosan. The use of a high concentration of NaOH aims to disrupt the bonds between the acetyl groups and the nitrogen groups, thus transforming them into amino groups (NH_2). The higher the NaOH concentration, the greater the number of substances that react, increasing the likelihood of collisions. The formation of chitosan from chitin occurs through the hydrolysis reaction of an amide by a strong base. In this process, chitin acts as an amide, while NaOH functions as its base. The reaction initially involves an addition reaction, where the -OH group is incorporated into the $NHCOCH_3$ group. Subsequently, an elimination reaction occurs, removing the CH_3COO group, resulting in the formation of an amino group (NH_2), which is chitosan (Cahyono, 2018).

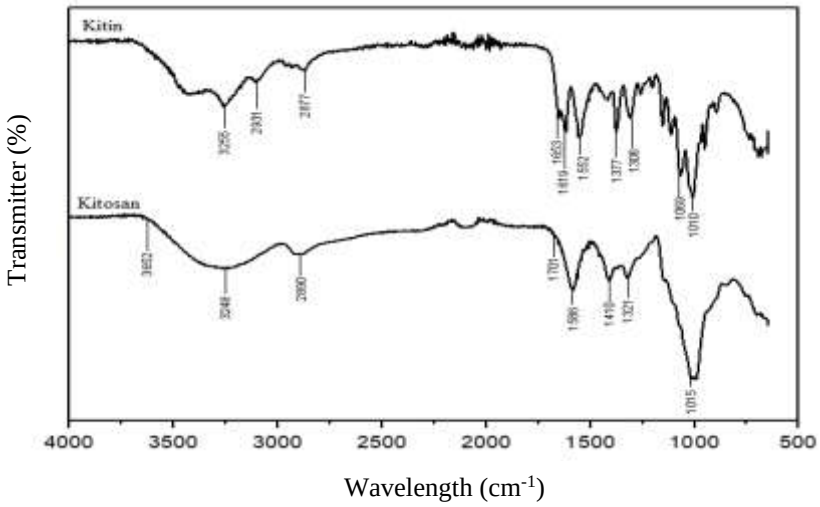

Figure 1. FTIR Spectra of Chitin and Chitosan

Based on the results of the chitosan FTIR spectrum, the degree of deacetylation can be calculated. The degree of deacetylation is a parameter used to determine the quality of chitosan by showing the percentage of acetyl groups that can be removed from the chitin yield to become chitosan. The difference between chitin and chitosan lies in the ratio of amino groups (NH_2) to acetyl groups ($NHCOCH_3$), which is referred to as the degree of deacetylation (Aulia & Rahayu, 2015).

The degree of deacetylation can be calculated using the baseline method from the analysis of chitosan using an FTIR spectrophotometer. Chitosan typically has a degree of deacetylation of less than 70% (Aulia & Rahayu, 2015). In this study, the degree of deacetylation of chitosan was found to be 87% (Table 2), indicating that the obtained sample is indeed chitosan. This degree of deacetylation in this study is better compared to the degree of deacetylation of chitosan from fishery waste, which was 61% (Ahmed, 2015) and from other *L. vannamei* shells, which was 76.24% (Imtihani & Permatasari, 2020). Azizati (2019) stated that the degree of deacetylation of chitosan from freshwater prawn shells is 93.47%. According to Aulia & Rahayu (2015), the higher the degree of deacetylation of chitosan, the lower the acetyl groups, leading to stronger interactions between ions and hydrogen bonds. The synthesis of chitosan from chitin occurs due to the breaking of bonds between the acetyl groups of chitin and nitrogen atoms (deacetylation), resulting in the formation of amino groups (NH_2).

Characterization of Chitosan

Chitosan characterization includes assessing its solubility, ninhydrin test, water content, and ash content (Table 2). Chitosan solubility is a key quality standard. Chitosan is dissolved in 2% glacial acetic acid, and the reaction demonstrates that chitosan completely dissolves in glacial acetic acid. This can be observed by comparing the clarity of the chitosan solution to that of the solvent (Jaya et al., 2017).

The ninhydrin test is conducted to qualitatively determine the presence of amino groups (NH_2) in chitosan. In this study, chitosan is sprayed with a ninhydrin solvent, resulting in a positive reaction characterized by the formation of a purple color (Ningrum et al., 2022). Ninhydrin is a strong oxidizing agent that reacts with the amino groups in chitosan, producing a compound resulting from the bond between hydrindantin and ninhydrin through a nitrogen bridge, which appears purple.

In this study, the moisture content of chitosan derived from *L. vannamei* shells is found to be 5,76% (Table 2). This aligns with the quality standard for chitosan moisture content, which specifies a moisture content of ≤ 10%. The moisture content present in chitosan represents water bound to the functional groups of chitosan (Al-Manhel et al., 2018). The moisture content in chitosan can be influenced by the duration of the drying process, the quantity of chitosan being dried, and the surface area of the chitosan being dried.

The ash content of chitosan in this study is determined to be 8.98% (Table 2). This indicates that the ash content produced does not meet the quality standard, which specifies an ash content of ≤ 2%. The high ash content suggests the presence of mineral content in the material. The ash content can be influenced by the washing process and the duration of agitation (de Queiroz Antonino et al., 2017).

Decolorization of Batik Industry Wastewater Using Chitosan from L. vannamei and P. ostreatus Baglog Waste

The color value of batik industry wastewater in this study is 5800 Pt-Co, which exceeds the established quality standard of 500-2500 Pt-Co. Such a high color

value indicates that the wastewater exceeds the permissible limit and can potentially pollute aquatic environments. Therefore, it is essential to undergo a treatment process before discharging it directly into water bodies. The color of batik industry wastewater can be reduced through adsorption processes.

In this research, chitosan from *L. vannamei* and waste from *P. ostreatus* baglogs are used as adsorbents for decolorization. Adsorption is the process of fluid adherence to the surface of the adsorbent material. The study explores different time variations for the decolorization process, including 0, 24, 48, 72, 96, and 120 hours.

Table 2. Characterization of chitosan from *L. vannamei*

Parameter	Yield of Chitosan (%)	Standard Chitosan (Jaya et al., 2017) (%)
Water content	5.76	≤ 10
Ash content	8.98	≤ 2
Dissolving in 2% glacial acetic acid	dissolve	dissolve
Ninhydrin test	Positive purple	Positive purple
Degree of deacetylation	87	≥ 70

Tabel 3. Decolorization of batik wastewater using chitosan from *L. vannamei* and waste from *P. ostreatus* baglogs

Time variation (hours)	Color value before treatment (Pt-Co)	Color value after treatment (Pt-Co)	pH value	Percentage of Decolorization (%)
12		2827	7.1	51.26
24		762	5.2	86.89
48	5800	1034	6.8	82.17
96		577	4.4	90.05
120		898	4.8	84.52

The results of the various time treatments for the decolorization of batik industry wastewater using chitosan from *L. vannamei* and waste from *P. ostreatus* baglogs (Table 3) show varying color values and decolorization percentages, ranging from 51.26% to 90.05%. The lowest decolorization value is obtained at 24 hours, which is 2827 Pt-Co with a decolorization percentage of 51.26%, while the highest decolorization value is achieved at 96 hours, which is 577 Pt-Co with a decolorization percentage of 90.05%. These results are lower than the decolorization achieved using the fungal mycelial-light expanded clay aggregate composite, which yielded a decolorization effectiveness still above 96% (Yanto et al., 2023), and also lower than the electrocoagulation method, which resulted in a decolorization value of up to 99.33% (Rusdianasari et al., 2020). The hydrodynamic cavitation and Ozonation with coagulation-flocculation pretreatment method also resulted in a higher decolorization value of 96.42% (Saputra et al., 2021). However, the results of this study are better compared to the immobilization of papaya laccase in chitosan method, which can degrade color up to a maximum of 67% (Jaiswal et al., 2016), 60.53% using *Ganoderma lucidum* with bioremediation process (Pratiwi et al., 2017), 88.2% using solar catalytic processes (Khalik et al., 2015).

The variation in adsorption time significantly influences the decolorization process of batik industry wastewater. The longer the decolorization process, the more color substances are absorbed by the adsorbent. This can be observed in Table 3, where the decolorization percentage increases as the duration of decolorization time extends. However, the increase in decolorization percentage may plateau once the adsorption capacity of chitosan from *L. vannamei* and waste from *P. ostreatus* baglogs reaches saturation (Wulandari et al., 2014).

The decolorization mechanism of batik industry wastewater using chitosan occurs because chitosan possesses amino groups (NH_2) within its carbon chain and carries a positive charge, rendering the molecules resistant to mechanical stress. The amino groups can bind to the colloidal particles present in the color substances of batik industry wastewater. Typically, colloidal particles in water are negatively charged, resulting in repulsive forces between particles of like charges. Chitosan, as an adsorbent, has the capability to stabilize these colloidal particles in water by forming larger flocs that can subsequently settle (Lichtfouse et al., 2019).

The reduction in color content through chitosan adsorption occurs when the particles in the water undergo destabilization with chitosan, reaching a maximum at a certain time. However, under certain conditions, the particles that have been destabilized and formed flocs become unstable with prolonged exposure, causing the flocs to break apart in the wastewater. Consequently, the color substances trapped within the flocs become dispersed once again (Aulia & Rahayu, 2015).

The decolorization process with the optimum contact time represents the point where the adsorption or diffusion of adsorbate molecules occurs most effectively. This process continues until it reaches a state of saturation (equilibrium), which should be marked by no significant further change in the concentration of color substances. The decrease in color substances after reaching the optimum time is due to a desorption process. Desorption can occur because the adsorption of color substances by chitosan is driven by ionic interactions, which result in the protonation of amino groups (NH_2) into amino ions (NH_3^+), which are basic in nature. As a result, as the adsorption time increases, the environment becomes more alkaline. An alkaline environment tends to promote desorption, which means the release of color substances that were previously bound to chitosan (Kyzas et al., 2017).

The decolorization process of batik industry wastewater using *P. ostreatus* baglog waste as an adsorbent is driven by multiple factors related to the fungal's components. The cell walls of *P. ostreatus* contain an extracellular matrix composed of various organic compounds, such as enzymes, proteins, and polysaccharides, along with a gel that acts as an adhesive substance, enabling the absorption of dyes from the wastewater. Additionally, the hydrophobic properties of the mycelium present in the waste interact with the hydrophilic nature of color substances, facilitating the adsorption process. Furthermore, wood powder in the waste is assumed to contain cellulose, whose hydroxyl groups react with nitrogen atoms in the colorants, forming hydrogen bonds and allowing colorants to bind to cellulose fibers. These combined mechanisms contribute to the effective decolorization of batik industry wastewater and result in high decolorization percentages (Wulandari et al., 2014). Using filamentous fungi for decolorization offers several advantages. Firstly, it's cost-effective, which is crucial for widespread application. Additionally, these fungi possess the ability to completely break down dyes, ensuring thorough treatment. Their ubiquitous presence in the environment highlights their adaptability, a trait vital for survival and effective bioremediation. Moreover, filamentous fungi produce ligninolytic enzymes, which are not selective towards aromatic structures, enabling them to degrade diverse mixtures of aromatic compounds efficiently. This versatility underscores their potential in environmental remediation processes (Zuraida et al., 2015). The mycelium of *P. ostreatus* has the ability to produce various ligninolytic enzymes that can break down complex organic compounds, including color pigments found in wastewater. This ligninolytic immobilized material has successfully decolorized approximately 94.867% of batik wastewater within a 24-hour period (Dewi et al., 2019).

The decolorization percentage of batik industry wastewater fluctuates from 24 hours to 120 hours, and this variation is influenced by the pH values of the solution (Table 3). The lowest pH value, 4.4, occurs at 96 hours, while the highest pH value occurs at 24 hours. The results differ from previous research that utilized bamboo charcoal as an adsorbent, which could raise the water pH value from 4.5 to 7.7 (Lensoni et al., 2023). pH values have a significant impact on the adsorption process (Nurafriyanti et al., 2017). In the pH range of 4-5, color substances exhibit a higher affinity to bind with the active groups, namely NH_2, on chitosan, leading to an increased amount of ionized color substances being adsorbed. However, as the pH value increases to the range of 6-7, the adsorption process decreases. This decline happens because the solubility of color substances in the solution decreases, making the free electron pairs in the active groups like amines and hydroxides in the adsorbent less capable of binding to the color substance ions. This occurs because the NH_2 groups become protonated into NH_3^+ ions due to the increase in the pH value of the solution. The higher the concentration of H^+ ions, the greater the tendency for protonation, causing a decrease in the adsorption process (Sheth et al., 2021).

After the decolorization process, batik waste yields a pH ranging from approximately 4.4 to 7.1 over a 120-hour exposure period. This indicates that the decolorized waste still tends to be acidic. Similar findings were observed in a study on batik waste discharge into the Simbangkulon river, where the water's acidity level (pH) at three stations ranged between 6.70 and 6.94. This suggests that the Simbangkulon river tends to be acidic, indicating the presence of batik waste pollution. The pH value is closely associated with carbon dioxide and alkalinity. At pH < 5, alkalinity can reach zero. The higher the pH value, the higher the alkalinity value, and the lower the level of free carbon dioxide. pH also influences the toxicity of a chemical compound. Ionizable ammonium compounds are commonly found in waters with low pH levels (Zammi et al., 2018).

Conclusion

The decolorization process using *L. vannamei* chitosan and *P. ostreatus* baglog waste with different time variations resulted in varying degrees of decolorization of batik industry wastewater. The most optimal time for the decolorization process of batik industry wastewater was found to be 96 hours, achieving a decolorization percentage of 90.05%.

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Author Contributions
Conceptualization, S. O., S. R., and C. N.; methodology, S. R.; validation, C. N.; formal analysis, C. N.; investigation, S. R.; resources, S. O., and S. R.; data curation, S. R.; writing—original draft preparation, S. O. and S. R.; writing—review and editing; S. O., S. R. and C. N. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest
The authors declare no conflict of interest.

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