Improving the quality of wastewater in traditional chicken slaughterhouses using moringa seed powder

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Abstract. In the city of Denpasar, Bali, Indonesia, there are hundreds of home-based chicken slaughtering industries. The slaughtering process produces liquid waste from washing chicken and equipment mixed with feces, blood, traces of protein, and fat. This waste contains high concentrations of organic material, dissolved solids, pathogenic microorganisms, and other hazardous substances that can pollute the environment. The environmental damage that occurs will adversely affect the health of humans and the surrounding animals. This study aims to determine the effect of Moringa seed powder on the quality of traditional chicken slaughterhouse waste in terms of pH, Total Suspended Solid (TSS), and Total Dissolved Solids (TDS). This study used a randomized group design with a split time pattern. The results showed that the concentration of Moringa seed powder of 200 mg/L with a deposition time of 60 minutes made the pH of wastewater close to normal pH, and also significantly reduced TSS and TDS (P<0.05). It can be concluded that moringa seed powder can improve the quality of traditional chicken slaughterhouse wastewater.

1. Introduction
The increase in the amount of chicken meat consumption is directly proportional to the increase in the poultry business chain, including home industry players engaged in slaughtering chickens. Most of the chicken slaughtering process in Denpasar, Bali, Indonesia is still carried out by traditional home industries. These home industries discharge their waste directly into public waterways. Waste produced from the traditional chicken slaughtering industry consists of two types, namely solid and liquid waste. Solid waste in the form of a mixture of feathers, feces, and disposed body parts. Liquid waste comes from washing water from carcasses and equipment mixed with blood, residual protein, fat, detergent, chlorine, and suspended solids [1][2][3], reported that the slaughterhouse and industrial-scale slaughterhouse waste has caused water pollution in the middle and downstream in the Subak Pakel River, Badung. Another study by Aini et al. [4], mentioned that the contamination of poultry slaughterhouse waste has caused an increase in Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), TSS, ammonia, pH and microbial contamination exceeding the wastewater quality standard.
The traditional chicken slaughterhouses wastewater is characterized by its content (blood, protein, fat, and the remnants of organic matter) which will contribute to the development of pathogenic microorganisms. This causes eutrophication and threatens the aquatic ecosystems within it. The eutrophication process also produces unpleasant odors that pollute the air. Organic and inorganic materials that are suspended and dispersed in it will cause river air pollution [5]. The quality of the water that has been polluted by waste can be measured by physical, chemical, and biological parameters, which reflect the quality of the air. The parameters that are measured include changes in pH, BOD, COD, TSS, and TDS, heavy metals and levels of microbial contamination [6]-[8].

Various wastewater treatment models have been developed to meet the quality standards before they are channeled into the river. The effort was made to reduce the composition of solids, hazardous materials, and microbial contamination before being discharged into the river. Wastewater treatment includes aerobic-anaerobic deposition systems, electrocoagulation, flocculation, coagulation, and sedimentation [9]-[12]. The biocoagulation-flocculation technique is considered safe and environmentally friendly because it utilizes natural ingredients [13]. One of them is a natural coagulant that is produced from moringa seeds [14]. Moringa seeds contain the active compound 4-(α-L-rhamnopyranosyloxy) benzyl isothiocyanate [15]. The use of Moringa seeds for water purification has been widely reported. Hendrawati et al. [16], succeeded in improving the quality of textile wastewater and groundwater. Another study reported that moringa seeds reduced the turbidity of drinking water [17], removed heavy metals [18], and decreased microbial contamination [19]. Due to the cationic polyelectrolyte protein content which can form floc by absorbing and binding colloidal particles and microbes in water [15], [20]. Omodamiro. Based on the descriptions above, further research is needed to determine the effect of Moringa seed powder on the quality standard of traditional chicken slaughtering waste in terms of pH, TSS, and TDS.

2. Methods
   2.1. Preparation of moringa seed biocoagulant
   The matured Moringa seeds were peeled and dried to a moisture content of 2%, then crushed to powder. Then, this powder was sieved using a 60 mesh sieve to obtain homogeneous simplicia [21]. Moringa seed biocoagulant was created by dissolving 1 gram of Simplicia into aquadest until the volume reaches 200 ml. Therefore, 5,000 mg/l (w/v) of Moringa seed biocoagulant preparations were obtained. These biocoagulant preparations were then used as a treatment for wastewater with various concentrations of 0 mg/L, 50 mg/L, 100 mg/L, 150 mg/L, and 200 mg/L adopted by Wibawarto et al. [15] and Hendrawati et al. [16].

   2.2. Wastewater flocculation-coagulation process
   A total of 5 beaker glass were each given a wastewater sample, and the moringa seed biocoagulant was added until the volume reached 500 ml. The amount of biocoagulant added to get the treatment concentration of 50 mg/L, 100 mg/L, 150 mg/L, and 200 mg/L is 5 ml, 10 ml, 15 ml, and 20 ml of coagulant, respectively. After the initial conditions are obtained, followed by a rapid stirring process at 200 rpm for 2 minutes and continued with slow stirring at 60 rpm for 5 minutes. Followed by the flocculation process with a variation of time 0, 20, 40, and 60 minutes to form a floc layer. Wastewater samples that have passed the coagulation-flocculation process were then measured (pH, TSS, and TDS) based on the determined concentration and deposition time.

   2.3. Measurement of the pH
   The degree of acidity (pH) of wastewater was measure using a pH meter (Senz pH, Trans Instruments Pte Ltd). The pH meter was calibrated and standardized before testing.

   2.4. Measurement of Total Suspended Solid
   Total Suspended Solid analysis uses the gravimetric method, referring to SNI-06-6989.3-2004 [22]. Filter paper with a size of 0.45 μm was placed into a porcelain cup and then dried in the oven at 103-
105°C for 1 hour, let stand for 5 minutes in a desiccator, and then weighed (WA). Furthermore, the filter paper is used to filter 20 ml of waste samples that have been treated. The residue retained in the filter is dried at a temperature of 103°C-105°C for 1 hour until it reaches a constant weight, let to cool down for 5 minutes in a desiccator, and then weighed (WB). The weight gain on the filter paper is marked as the total suspended solids. To determine the value of TSS, calculated using the following formula:

\[ TSS = \frac{(W_B - W_A) \times 1000}{Volume \ of \ sample \ test} \]

Note: TSS is total suspended solids (mg/L); WA is filter paper weight (mg); WB is filter paper weight with dried residue (mg).

2.5. Measurement of Total Dissolved Solid

Total dissolved solid analysis uses the gravimetric method, referring to SNI 06-6989.27-2005 [23]. A total of 50 ml sample of flocculation-coagulation waste was placed into a filter that had been equipped with a vacuum pump and filter paper. After perfect filtering, filtered waste was then moved to a cup with fixed weight (W0). The filtered waste was then evaporated to dryness over a water bath, then placed into the oven at 180°C temperature for 1 hour. The cup was allowed to stand in a desiccator for 5 minutes, then weighed. Repeat the heating process in the oven until a fixed weight (W1) is obtained. To determine the value of TSS, calculated using the following formula:

\[ TDS = \frac{(W_2 - W_1) \times 10^6}{Volume \ of \ sample \ test} \]

Note: TDS is total dissolved solids (mg/L); W1 is weight remains empty cup after heating 180°C; W2 is fixed weight of the cup containing TDS after 180°C heating

2.6. Statistic analysis

Quantitative research data on pH, TSS, and TDS were analyzed using one-way Analysis of Variance, followed by Duncan test if there are significant differences between treatments. Variations between treatment groups of p<0.05 were considered to be statistically significant. Quantitative research data on pH, TSS, and TDS will be analyzed using one-way Analysis of Variance, followed by Duncan test if there are significant differences between treatments. Variations between treatment groups of p<0.05 were considered to be statistically significant.

3. Results and Discussion

3.1. Results

In the current study, wastewater treatment using Moringa seed powder succeeded in improving the quality of wastewater in a traditional chicken slaughterhouse in terms of pH, TSS, and TDS. The best results are obtained at a dose of 200 mg/L with a deposition time of 60 minutes. The average value of pH, TSS, and TDS are shown in Tables 1, 2, and 3. Statistical analysis shows that treatment of Moringa seed powder and deposition has a significant effect (P<0.05) on pH, TSS, and TDS of wastewater.

| Concentration (mg/L) | pH     | Sig.   | Deposition time (minutes) | pH     | Sig.   |
|----------------------|--------|--------|---------------------------|--------|--------|
| 0                    | 6.05 ± 0.53\textsuperscript a | 0.000  | 0                         | 6.51 ± 0.61\textsuperscript a | 0.018  |
| 50                   | 6.25 ± 0.29\textsuperscript b |        | 20                        | 6.58 ± 0.63\textsuperscript b |        |
| 100                  | 6.55 ± 0.19\textsuperscript c |        | 40                        | 6.72 ± 0.61\textsuperscript c |        |
| 150                  | 7.16 ± 0.14\textsuperscript d |        | 60                        | 6.82 ± 0.62\textsuperscript d |        |
| 200                  | 7.38 ± 0.14\textsuperscript d |        |                           |        |        |

Note: Different superscript letters towards the column show significantly different (P <0.05)
3.2. Discussion

The degree of acidity (pH) is the most important factor affecting the coagulation process. When the coagulation process is not carried out in the optimal pH range, it can cause to fail flocculation and result in poor water purification quality [16]. The pH value is used to measure whether the condition of wastewater is acidic (<7) or alkaline (>7), whilst the optimum pH ranges from 6-8. Moringa seed biocoagulant is a polyelectrolyte cationic protein. At optimum pH the amino acids are ionized to produce carboxylic ions and protons. The composition of chicken slaughtering wastewater consists of suspended or dissolved organic material in the form of negatively charged particles that cause turbidity in water. The charge of protons (positive ions) will attract negatively charged colloids (electrons) in the waste to form a neutral group and then produce flocs [16][19][24].

Table 2. The average of total suspended solids after moringa seed flocculation

| Concentration (mg/L) | TSS (mg/L) | Sig. | Deposition time (minutes) | TSS (mg/L) | Sig. |
|----------------------|------------|------|--------------------------|------------|------|
| 0                    | 883 ± 94a  | 0.000| 0                        | 677 ± 214a | 0.0018|
| 50                   | 721 ± 96b  |      | 20                       | 573 ± 217b |      |
| 100                  | 596 ± 89c  |      | 40                       | 540 ± 223c |      |
| 150                  | 446 ± 78d  |      | 60                       | 533 ± 273d |      |
| 200                  | 258 ± 95e  |      |                          |            |      |

Note: Different superscript letters towards the column show significantly different (P <0.05)

Table 3. The average of total dissolved solid after moringa seed flocculation

| Concentration (mg/L) | TDS (mg/L) | Sig. | Deposition time (minutes) | TDS (mg/L) | Sig. |
|----------------------|------------|------|--------------------------|------------|------|
| 0                    | 9557 ± 225a| 0.000| 0                        | 8303 ± 1457a| 0.001|
| 50                   | 9385 ± 225b|      | 20                       | 8265 ± 1512b|      |
| 100                  | 9219 ± 130c|      | 40                       | 8182 ± 1500c|      |
| 150                  | 6721 ± 257d|      | 60                       | 8078 ± 1617d|      |
| 200                  | 6154 ± 161d|      |                          |            |      |

Note: Different superscript letters towards the column show significantly different (P <0.05)

The flocculation rate will increase significantly, followed by the deposition of colloidal particles. This causes the water to turn clear, which is directly proportional to the duration of deposition. In the process of flocculation, the microbial is also coagulated. The flocculation process was able to eliminate about 80-90% of bacteria that generally attach themselves to organic material in the form of colloidal particles [25]. In this study, coagulation of Moringa seeds produced an optimum condition which caused a change in waste pH from 6.05±0.53 to 7.38 ± 0.14, while letting the deposition take part for 1 hour caused a change in pH from 6.51±0.61 to 6.82±0.62. These results are in line with the study of Sari et al. [26], which reported the cationic protein of Moringa seeds caused the pH of initially alkaline brackish water samples to change near-neutral pH because protons attract hydroxyl and colloid groups with negative charges. The duration of deposition also affects the change in pH [27]. Electron absorption by cationic proteins requires sufficient time until the maximum amount of floc is formed, or reaches its saturation point. If the interaction lasts too long, it can interfere with the stability of the floc, as occurs in the bond of lead metal with the Moringa seeds, which are re-released due to the flocculation duration being too long [18], [28].

This wastewater contains very high concentrations of TSS and TDS. TSS consists of organic material with a maximum particle size of 2 µm, including microbes derived from feces. This TSS component first forms a deposition that will hinder the ability to form O2 production in water, which explains why the TSS parameter became an important parameter to determine the initial environmental conditions [29]. TDS components are smaller in size compared to suspended solids. TDS consists of organic and inorganic materials as well as hazardous materials remnants of cleaning and sanitizing.
solution. The high variety of chemical substances contained in wastewater produces unpleasant colors, tastes, and odors. Some dissolved chemicals may be toxic or carcinogenic. Total dissolved solids are dissolved materials (ϕ <1 μm) and colloids (ϕ 1-100 μm) [30]. In this study, the initial TSS yield was 883 ± 94 mg/L and after biocoagulant treatment, the number became 258±95 mg/L (70.78% decrease). Deposition treatment turns the TSS yield from 677±214 to 533±273 mg/L (21.27% decrease). On the other hand, biocoagulant treatment causes the TDS yield to decrease 35.60% from a mean of 9557±225 to 6154±161 mg/L, whereas with the deposition treatment TDS was relatively constant from 8303±1457 to 8078±1617 mg/L.

Turbidity is closely related to the TDS value in water; the higher the TDS value in water, the higher the turbidity value of water. Turbidity prevents light absorption, which causes the process of photosynthesis and other vegetation in the water to be disrupted. TDS can be used as one parameter to measure water quality. This parameter is used to determine whether a water source is suitable as a source of drinking water. Based on the Regulation of the Minister of Health of the Republic of Indonesia, the requirement for clean water quality is to have a TDS value of not more than 1,500 mg/L. Cationic protein can decrease repulsion between colloidal particles in wastewater, so colloidal particles will settle and form flocs [18]. According to Li et al. [31], flocs formed from TSS and TDS can result in changes in the density of wastewater. The use of a suitable biocoagulant will produce a dense floc, resulting in a stable solution [32]. Excessive or insufficient concentration of coagulant can reduce the efficiency of flocculation of dissolved solids. Lack of coagulant causes relatively small floc size and affects the turbidity of wastewater, which will require longer deposition time [13], [33]. Likewise, the use of excessive coagulant causes the solution to become overly saturated with impurities as a result. High concentrations of TSS and TDS contained in traditional chicken slaughterhouse wastewater will cause water pollution [5], [34]. Biocoagulant treatment is able to reduce TSS and TDS significantly, but it still does not meet the quality standards of wastewater set for TSS (100-200 mg/L) and TDS (1000-2000 mg/L). Based on these facts, further studies are needed to overcome the potential of pollution from traditional chicken slaughterhouse wastewater.

4. Conclusion
In this study, we concluded that the biocoagulation of Moringa seed powder can improve the quality of traditional chicken slaughtering wastewater. The best results were obtained at the concentration of 200 mg/L moriga seed powder with a deposition time of 60 minutes.

5. References
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