The potential of chitosan from comb-pen \((Atrina pectinata)\) shell waste on the characteristics of hand body cream

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Abstract. The comb-pen shell is one of the sources of chitosan, which is a natural substance derived from chitin and could be found in Indonesia. In the non-food sector, chitosan can be used as antibacterial, antioxidant, and thickening on hand body cream ingredients. This study aimed to determine the effect of the addition of chitosan from comb-pen shell waste to the characteristics of hand body cream. The parameters, including pH, TPC, and Homogeneity were tested on the first day and after fourteen-day storage at room temperature. The pH test results range from 7.8 to 8.0. The TPC test result is <10 CFU/25 g in every sample on both tests. The hand body cream homogeneity has the same homogeneity level between the control and the treatment samples. The viscosity results were obtained from literature studies regarding the manufacture of hand body cream using chitosan from small crab \((Portunus pelagicus)\) shells. The characteristics of hand body cream have a good result on every parameter test shown that are fulfilled the standard of Indonesia Nasional Standards (SNI). The addition of chitosan from comb-pen shell \((Atrina pectinata)\) waste affects the characteristics of hand body cream.

1. Introduction

Comb-pen \((Atrina pectinata)\) shell is one of the fisheries commodities found in many coastal areas in Indonesia. Comb-pen was commonly used as commercial consumption material for coastal communities. One of its uses is as a source of chitin and chitosan [1]. Chitosan is a derivative of chitin obtained from the extraction process by removing the acetyl group by leaving the free amine group [2].

The use of chitosan in the non-food sector, among others, in the fishing industry, agriculture, industrial environment, pharmacy, and health [3]. This is because chitosan is a reactive material and is easy to process [4]. Chitosan is widely used as an antibacterial material because it has affinity properties that can affect the failure of the bacterial DNA synthesis process [4]. Chitosan also has hygroscopic properties, which means it can bind to water so that it will affect the water content of the product. The antioxidant properties of chitosan are also widely used because chitosan can reduce the activity of free radicals by binding to free radicals [5].

The use of chitosan in the non-food sector, especially cosmetics, is as a material for making hand body cream. The hand body cream is an emulsion product with daily topical use. Characteristics of good hand body cream following Indonesia national standards are those that have a pH value between...
4.5-8.0; viscosity values between 2,000-50,000 cPs; homogeneous mapping and has a maximum total plate count of $10^2$ [6].

2. Material and methods

2.1. Material
Comb-pen (A. pectinata) shell waste chitosan obtained from previous Fisheries and Marine Faculty research at Universitas Airlangga.

2.2. Chitosan solution preparation
Chitosan solution was prepared according to the Apriadi [6] method, which is by dissolving chitosan (1 g, 2 g, 3 g) with 1% acetic acid with a ratio of 1:10 (w/v) then adding distilled water to 100 ml volume.

2.3. Making the hand body cream
The hand body cream was made refers to the modified Tribawono [7] method which consists of a water phase consisting of 2.5 g propylene glycol, 2 g triethanolamine, 3.5 g glycerin, 4 g BHT, 0.2 ml Disodium EDTA, 2 g cetyl alcohol, 1 ml chitosan solution, and 25 ml water and the oil phase consisting of 1.5 g of lanolin, 8 g olive oil, and 3 g stearic acid which was homogenized until evenly distributed and stopped at 35°C.

2.4. pH Test
The pH test is carried out by using pH meter. The measurement begins to clean the eyes pH meter using distilled water and wiped with a tissue. Dip the pH meter into the diluted sample until the number that appears on the pH meter is stable [8].

2.5. Total plate count test
Total plate count test is carried out by weighing a sample of 1 g added 85% NaCl solution to a volume of 10 ml and stirring evenly to obtain a $10^1$ dilution. A $10^2$ dilution is obtained by dissolving 1 ml of a $10^1$ dilution sample with the addition of an 85% NaCl solution. The same method is carried out until $10^5$ dilution. Then the sample was inoculated on the media. 1 ml sample solution that has been diluted put into a petri dish then add the nutrient agar to the petri dish as much as 12 ml. Incubation at 37°C for 24 hours. The incubated media was then calculated by counting colonies with a diameter of 0.5 - 3 mm [9].

2.6. Homogeneity test
Homogeneity test is done by weighing 0.1 g of hand body cream and place it on a glass object and covered with a glass cover and then observed using a microscope with a magnification of 40 x 10 or can be seen in plain view [10].

2.7. Viscosity test
The viscosity results were obtained from literature studies regarding the manufacture of hand body cream using chitosan from small crab (Portunus pelagicus) shells [11].

2.8. Data analyze
This study is experimental research using Completely Randomized Design (RAL) with four treatments (Control: without addition of chitosan solution, P1: Addition 1% of the chitosan 1 g solution, P2: Addition 2% of the chitosan 2 g solution, P3: Addition 3% of the chitosan 3 g solution) and five repetitions. The obtained data from pH test were analyzed by ANOVA (Analysis of Variance) and continued with Duncan Multiple Test (DMRT) to determine the very significant differences (P<0.01).
3. Result and discussion

3.1. pH

The obtained pH test result are between 7.8-8.0 both on the first day of testing and after 14 days of storage. The difference pH values in each treatment is due to the use of different amounts of acetic acid 1%. The higher the concentration of chitosan solution, the more acidic the pH value of the hand body cream [11]. The change in pH value during the storage period is very small, so it does not greatly affect the final pH value.

A good pH value of hand body cream was neutral or 7. This is because if the pH value is too acidic it will irritate the skin, whereas if it is too alkaline the skin will dry so that it will eliminate the function of the hand body cream as a skin moisturizer [12]. Analysis of variance in pH test result showed very significant differences between treatments. This shows that chitosan affects pH value in each treatment. The pH test result can be seen in Table 1.

| Sample | Average pH ± SD | First-day Storage | After Fourteen-day Storage |
|--------|-----------------|------------------|---------------------------|
| Control | 7.86 ± 0.06     |                  | 7.84 ± 0.06               |
| P1     | 8.02 ± 0.04     |                  | 8.00 ± 0.03               |
| P2     | 7.94 ± 0.02     |                  | 7.91 ± 0.03               |
| P3     | 7.87 ± 0.02     |                  | 7.84 ± 0.03               |

Note: Different letter in the same column indicate very significant difference (P<0.01).

3.2. Total plate count

Result of the TPC test can be seen in Table 2.

| Sample | P1          | P2          | P3          |
|--------|-------------|-------------|-------------|
| I      | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g |
| II     | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g |
| III    | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g |
| IV     | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g |
| V      | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g | <10 cfu/ 25 g |

Note: The unit weight of the test sample is in accordance with SNI 01-2332.3-2015.

In the test media with 10^1 dilution, there were no bacteria colonies that grew, so the next dilution was not tested. Based on the rules of writing microbiological test results following national standards, it is not allowed to write 0 (blank). Then the result wrote using the formula 1/d, where “d” is the dilution factor used, it was 10^1. The symbol less than is a sign that there is no colony growing on the media test.

Test results after 14 days of storage also showed a good result. This shows that the antibacterial properties of chitosan can work on hand body cream. The antibacterial properties of chitosan are due to the affinity of its ability to bind to bacterial DNA and influence the bacterial protein synthesis process [4]. The affinity of chitosan is influenced by the value of the degree of diacetyl, the higher the value of the degree of diacetyl, the higher the antibacterial properties of the chitosan which means the more positive ion on the chitosan [4].
According to Zheng et al. [13] antibacterial activity on chitosan against bacteria both gram-negative or gram-positive bacteria has two possible mechanisms, the first is that chitosan binds to the surface of the bacterial cell and forms a polymer membrane that can prevent the entry of nutrients into the cell so that over time the cell will die, the second low molecular weight chitosan will enter the cell and include the cell because chitosan can adsorb the cell's electronegative substance and makes the cell float and will disrupt the psychology of cell activity so that over time the cell will die. Electronegative cells in gram-positive bacteria are found in cell walls, namely lipoteichoic acid (LTA). LTA is part of the bacteria that will later bind with polycationic to chitosan [14].

3.3. Homogeneity

Homogeneity test result can be seen in Figure 1, Figure 2, Figure 3, and Figure 4.

![Figure 1. Homogeneity of control sample.](image1)

![Figure 2. Homogeneity of P1 sample.](image2)

![Figure 3. Homogeneity of P2 sample.](image3)

![Figure 4. Homogeneity of P3 sample.](image4)

Homogeneity of hand body cream test results show the same level of homogeneity both on the control and treatment samples. Homogeneity measurements were carried out directly on the visible eye and applied to the skin. Each sample has a good homogeneity. This is shown when applying to the skin does not require a long time to absorb, there are no lumps of material that are not mixed with other ingredients and does not leave an oily effect on the skin. Erungan et al. [15] state that the good homogeneity of the emulsion can be seen from the absence of separation or lumps of material used in the emulsion.

Homogeneity was influenced by the ingredients and the stirring process used at the time of manufacture. In this study the use of chitosan that slightly does not affect the homogeneity of hand body cream. Homogeneity quality of good hand body cream according to Indonesia's national standards is homogeneous.
3.4. Viscosity
The viscosity results were obtained from literature studies regarding the manufacture of hand body cream using chitosan from small crab (Portunus pelagicus) shells [11]. Based on this research, it shows that the difference in the use of chitosan concentration affects the viscosity level very significantly. The use of 3% chitosan concentration had the highest value compared to other concentrations, it was 48.747 cPs, while the viscosity value of the 0% concentration of chitosan was the lowest, and it was 21.653 cPs. This shows that chitosan functions as a thickener in the manufacture of hand body cream. The ability of chitosan as a thickening agent is because chitosan is a hygroscopic material, which means it can bind to water and will increase the viscosity of the hand body cream.

4. Conclusion
The addition of chitosan with different concentrations affects the characteristics of hand body cream. Each parameter shows good results and meets the quality requirements following Indonesia's national standards SNI 16-4399-1996.

5. References
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