The utilization of chitosan from Comb-pen shell (*Atrina pectinata*) as an emulsion stabilizer in the production of hand body cream

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Abstract. Chitosan is a natural polymer that can be used as a source of natural materials because it has good characteristics. Chitosan is used in the cosmetic sector as a humectant, thickener, moisturizer, antioxidant, sunscreen cream, and stabilizer. The shell chitosan of the shells is used as an emulsion stabilizer in the Hand Body Cream because the emulsion product has problems in its unstable nature so that oil and water are easily separated. The parameters observed were emulsion stability, pH, and TPC. The result of this study indicated that the chitosan substitution in the shell of the shells gave significantly different result (P<0.01) between the control treatment against the addition of 1%, 2%, and 3% chitosan addition, but there was no significant difference between the 1% addition of chitosan treatments 2% and 3% of emulsion stability values and pH on day 1 and day 14. The TPC test results are <10 cfu/25 g in every sample on both tests.

1. Introduction  
Comb-pen (*Atrina pectinata*) are one of the popular fisheries commodities because they have a high price and high protein content. In general, the shellfish is only consumed commercially, and the remaining shell is obtained as waste. Based on data on the export of Indonesian fishery products in 2004, for the commodity shellfish produced about 2.752 tons [1].

The shell waste has the potential to be used as a high value and useful product, known as waste to product [1]. In general, clamshells are one of the fisheries raw materials that contain chitin so that the shell waste can be further processed into high-value products, namely chitosan [2]. Chitosan in the cosmetic field has been applied as a humectant, thickening agent, moisturizer, antioxidant, sunscreen cream, and emulsion stability [3].

The most preferred hand body cream is a cream product in the form of oil in water (O/W). The problem that arises in the emulsion product is that it is unstable so that the oil and water are easily separated. Time, temperature variation, mixing process, and suitability of the material used have a complex effect on the emulsion product [4]. A stabilizer needs to be added [5] to stabilize the emulsion system.
2. Material and methods

2.1. Material
Chitosan from Comb-pen (A. pectinata) shell waste was made at Fisheries and Marine Faculty Airlangga University.

2.2. Chitosan solution preparation
Chitosan solution was prepared according to the Sidauruk et al. [3] 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.

2.3. Making hand body cream
The hand body cream was made refers to the modified Sidauruk et al. [3] method, which consists of a water phase consisting of 2.5 g propylene glycol; triethanolamine 2 g; glycerin 3.5 g; BHT 4 g; cetyl alcohol 2 g; 1 ml chitosan solution; and water 25 ml for the 1%, 2%, and 3% treatment. While the control treatment without using chitosan solution and replaced with disodium EDTA 0.2 ml. the oil phase consisting of 1.5 g of lanolin; 6 g olive oil; and 3 g stearic acid, which was homogenized until evenly distributed and stopped at 35°C.

2.4. Emulsion stability test
Testing the emulsion stability using the centrifugation method (McClement method) [6]. A sample of 14 ml of hand body cream was put into a centrifuge tube, then put into a centrifuge at 4500 rpm form 5 minutes at room temperature. After the results of the centrifuge are obtained, then measured the volume of cream formed. The creaming index is calculated using the formula according to McMlemen:

\[ CI = \frac{HS}{HE} \times 100\% \]

Note:  
CI = Creaming Index  
HS = Emulsion Height After  
HE = Initial Emulsion Height before Centrifuge

2.5. pH Test
The pH test is carried out by using a 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 [4]. Tests were carried out on day 1 and day 14 [7].

2.6. Total plate count test
Testing of Total Plate Count is carried out by testing the storage temperature. The hand body cream is stored at room temperature ±23°C for 14 days [7]. TPC testing uses a reference from SNI 2332.3: 2015.

2.7. Data analyze
This study is experimental research using Completely Randomized Design (RAL) with four treatments (Control: without the 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 replications. The obtained data from emulsion stability and pH tests were analyzed by ANOVA (Analysis of Variance) and continued with Duncan Multiple Test (DMRT).

3. Result and discussion

3.1. Emulsion stability
The value of the emulsion stability produced on the first-day test ranged from 82.42-99.57%, while on the 14 days, the test ranged from 81.28-99.28%. The results on the emulsion stability value of the hand
body cream were obtained based on calculations using the emulsion stability calculation formula. The value data presentation of the emulsion stability of the hand body cream can shown in Table 1.

Table 1. Emulsion stability test

| Sample | Average Emulsion Stability ± SD | First-day Storage | After Fourteen-day Storage |
|--------|--------------------------------|------------------|----------------------------|
| Control | 82.42b ± 3.05 | 81.28b ± 3.28 |
| P1     | 99.28a ± 0.71 | 98.57a ± 0.50 |
| P2     | 99.42a ± 0.59 | 99.14a ± 0.59 |
| P3     | 99.57a ± 0.63 | 99.28a ± 0.71 |

Note: The same superscript in the same column does not show a significant difference (P<0.01).

Emulsion stability is an important parameter in emulsion products. The emulsion stability level indicates the resistance of the emulsion product to certain conditions and within a certain time span [7]. Testing the stability of emulsion in this study using a mechanical test method with a centrifuge. The centrifuge test is an indicator of the physical stability of semisolid preparations [8].

The standard value of commercial hand body cream is 98% [9]. The stability of the hand body cream emulsion on the first day and after fourteen days had significantly different results between the control treatments with the addition of chitosan treatment 1%, 2%, and 3%. While the emulsion stability of hand body cream between treatments of 1%, 2%, and 3% chitosan addition was not significantly different.

The addition of chitosan to a level of 3% can be balanced with an increase in the value of emulsion stability, which increases. The emulsion stability of the hand body cream is not influenced by the concentration of chitosan used, so it is assumed that the value obtained is influenced by the composition of the constituent ingredients used [3].

3.2. pH

The result of the pH test have been shown in Table 2.

Table 2. pH test

| Sample | Average pH ± SD | First-day Storage | After Fourteen-day Storage |
|--------|----------------|------------------|----------------------------|
| Control | 7.88a ± 0.05 | 7.88a ± 0.05 |
| P1     | 8.13b ± 0.03 | 8.15c ± 0.01 |
| P2     | 8.07b ± 0.07 | 8.05b ± 0.06 |
| P3     | 7.94a ± 0.03 | 7.94a ± 0.03 |

Note: The same superscript in the same column does not show a significant difference (P<0.01).

pH measurement aims to determine the degree of acidity or basicity owned by a preparation that can affect comfort when the preparation is applied [8]. pH value with the addition of chitosan 1% and 2% on the first day and after fourteen days had a pH value exceeding 8. Product with a pH that is far from the standard will more easily irritate the skin. The skin is covered by an acid mantle, which is a moist, acidic layer on the surface of the coolie [10].

The test result shows that the control treatment and as much as 3% of the addition of chitosan on the first and after fourteen days get the pH value according to the SNI. The higher the addition of chitosan, the lower the pH value will be. This is due to the added chitosan is chitosan dissolved in acetic acid in more quantities than other treatments, thus affecting the pH value of the product produced [11].
3.3. Total plate count
The result of the TPC test can be seen in Table 3.

|       | Control | 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.

Total Plate Count (TPC) is important because microbial contamination can cause the separation of the water phase and the oil phase, which affects the stability of the emulsion, severe shrinkage, and the generation of unpleasant odors [7].

The result of TPC tests on the first day and after fourteen days of all treatments showed good results, at 10<sup>-1</sup> dilution, no microorganisms were found to grow on the agar medium to which the sample was added. The colony number of all plates was less than 10 or plates without colonies. The test results show that the aseptic process has been achieved and the addition of active compounds that are capable of being anti-microbial and able to inhibit the growth of microorganisms [7].

4. Conclusion
The addition of chitosan *A. pectinata* had a good effect on the stability of the emulsion of the hand body cream, but it was not affected by different concentrations of the chitosan used. The result is also supported by the supporting test values of pH and TPC which are also good.

5. References
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