Microcapsule efficiency of ethanol extract of rosella petal flower (*Hibiscus sabdariffa* Linn) coated crude carrageenan (*Eucheuma cottony*)

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Abstract. The research of microcapsule efficiency of ethanol extract of sepal flower (*Hibiscus sabdariffa* Linn) coated crude carrageenan has been done. The aim of research is to determine the highest efficiency microencapsulation based on the ratio of ethanol extract of the sepal flower toward seaweed crude carrageenan (*Eucheuma cottony*) as coating substance. The microencapsulation was carried out by using freeze drying technique. This encapsulation may preserve the active compound of ethanol extract. Ratio of ethanol extract of the sepal flower toward seaweed crude carrageenan were set at 1:3; 1:4; 1:5; 1:6 and 1:7 (w/w). The result of research obtained that the highest efficiency ratio is at 1:7 with percentage of 86.01%.

1. Introduction
Rosella (*Hibiscus sabdariffa* Linn) is one of the plants that can grow in all tropical and sub-tropical countries including Indonesia. Rosella plants have a red dye that many people use, especially in the food and cosmetics industry. Some examples of types of foods that use rosella petal extract as a food product coloring agent are jelly, sauce, tea, syrup, jam, pudding, and sweets while some examples of types of cosmetics use rosella petal extract, namely lipstick and lip smear preparation [1].

Rosella flower petals contain active ingredients in the form of flavonoids, phenols or polyphenols, citric acid, ascorbic acid, tartaric acid, malic acid, calcium, protein, beta carotene, saponins, tannins, antioxidants such as gossyipeptin, anthocyanin, glucose hibiscin [2]. The important content found in the rosella petals is the anthocyanin pigment that forms flavonoids which act as antioxidants. Based on research results [3] rosella flower petals have the highest total anthocyanin of 128.76 mg / 100g.

Anthocyanin is a red pigment; its stability is influenced by pH and temperature. Anthocyanins are more stable in acidic or low pH [4]. Based on the results of a study [5], the anthocyanin compound of rosella petal extract is stable at pH <3 and the temperature <60°C. Efforts to improve stability can be done by coating or microencapsulation methods. One of the coating materials used is carrageenan. Carrageenan is a natural additive which is widely used in various industries, especially food industry and cosmetics. Coarse Carrageenan or Semi Refined Carrageenan (SRC) is one of the carrageenan products with a lower purity level compared to pure carrageenan or Refined Carrageenan (RC). SRC contains a small amount of cellulose which settles along with carrageenan. This product is produced by heating process using alkali solution of Potassium Hydroxide (KOH) [6]. Based on the result of research...
[7], SRC seaweed production get the highest yield of 33.83% at 8% KOH concentration. SRC is easy to hydrolysed at acidic pH while at SRC alkaline pH it is difficult to hydrolysed, but stable in gel form [8]. This property is related to its ability as a coating material in the manufacture of microcapsules.

According to [9], microencapsulation is a coating technique of filler material with a thin layer of coating material. Selection of coating material is important because it affects the stability of the emulsion before drying. In addition to the coating factor that influences the microencapsulation is the ratio of the core material to the coating. The method of making microcapsules can be done by conserving phase separation, dry spray, frozen spray, evaporation emulsification system, air suspension, centrifugal multi-hole process, coating in the pan and polymerization [10].

The effect of coating ratio extract of purple-maltodextrin eggplant skin on the efficiency of anthocyanin pigment encapsulation of purple eggplant skin produced the highest efficiency value of 63.85% at a ratio of 1: 5 (b / b) [11]. While the research of [7] which examines the effect of ethanol extract coated with maltodextrin ratio (1: 1); (1: 3); (1: 4); (1: 5) obtained anthocyanin total coated maltodextrin at a ratio of 1: 5 (v/b) that is equal to 309.32 mg / 100g at freezing temperature (-20 °C) for 24 hours and decreased after storage for eight weeks to 212.75 mg / 100g.

Based on the description, in the study will be carried out an analysis of the efficiency of ethanol extract microcapsules coated rosella petals in coarse carrageenan by emulsification using tween 80.

2. Methods

2.1. Materials
The materials used in this study are rosella flower petals (Hibiscus sabdariffa Linn), seaweed (Eucheuma cottonii), 96% ethanol, 8% KOH, filter paper and aquadest.

2.2. Instrumentation
The equipment used in this study are blender, filter cloth, 2 liter Erlenmeyer, 100 ml Erlenmeyer, rotary vacuum evaporator, analytical balance, glass plate, iron gutter, freeze dryer, oven, baking pan, vial bottle, magnetic stirrer, 50 mesh sieve and 250 ml beaker, 500 ml beaker and Scanning Electron Microscopy (SEM) at the Bandung Institute of Technology's Nano science and Nano Centre Laboratory.

2.3. Procedure

2.3.1. Rosella Flower Petals Extraction. Rosella flower petal extraction was done using maceration method as follows: as much as 200 grams of rosella petal flour were put into 2 liter Erlenmeyer, then 2 liters of 96% ethanol were added and stored for 3 × 24 hours. The mixture is filtered, the filtrate is vacuum-concentrated using a rotary evaporator to obtain concentrated extract, then stored in cold temperature. Extract yield is calculated using the following equation:

\[
\% \text{ Yield} = \frac{\text{weight of thick extract (g)}}{\text{weight of dried rosella petal flour (g)}} \times 100\% \quad (1)
\]

2.3.2. Seaweed Extraction. Dry red seaweed is weighed as much as 25 g and put into a 500 mL beaker. Then added 250 mL of 8% KOH solution and heated at 50°C for 120 minutes. After that it is filtered and dried in an oven at 60°C for 3 hours, after drying it is smooth using a blender. The rendemen is determined by using the following equation:

\[
\% \text{ Yield} = \frac{\text{weight of coarse carrageenan (g)}}{\text{weight of dried seaweed (g)}} \times 100\% \quad (2)
\]

2.3.3. Microencapsulation. Microcapsules of ethanol extract of rosella flower petals coated with coarse carrageenan are made in a ratio of 1: 3; 1: 4; 1: 5 1: 6; 1: 7 on the basis of (b / b) by mixing the two ingredients in a beaker, stirring using a magnetic stirrer for 1 hour and adding 3 drops of tween 80 and
then storing it in frozen storage $3 \times 24$ hours. The mixture is dried using freezer drying. Each microcapsule was analysed for coating efficiency.

2.3.4. Microcapsule Efficiency. A total of 0.5 grams of microcapsules were crushed and extracted with 10 mL of 96% ethanol, then filtered and the filtrate concentrated using a rotary evaporator, concentrated extract obtained then in the oven and weighed, then microcapsule efficiency was calculated by the equation:

$$\text{% Efficiency} = \frac{(\text{Mass extract of microcapsules (g)} - \text{Control mass} \times \frac{n}{b})}{\text{Mass of earlypetal roselula flower extract (g)}} \times 100\% \quad (3)$$

Description: $a = \text{Total microcapsule mass (g)}$
$b = \text{Mass of microcapsules analysed (g)}$

2.3.5. Determination of Microcapsule Morphology. The surface morphology of microcapsules which have the highest efficiency value was analysed using a SEM tool with a magnification of 1000 X.

3. Results and Discussion

3.1. The yield of Rosella Flower Petal of Ethanol Extract
Extraction method used is maceration method. The results showed that the yield of ethanol extract was 32.07% in 200 grams of roselula flower petals.

Extraction of rosella petals by maceration method at room temperature of 25°C for 24 hours showed that the yield obtained was 17.7% in 100 grams of rosella flower petals, so that in this study produce the highest yield compared. The cooking time used is $3 \times 24$ hours, the extraction time is longer than the yield and produced is greater because the contact time is fulfilled between the solvent to the interact the extracted substance [12].

3.2. Rough carrageenan yields from seaweed
Coarse carrageenan extract is the product of seaweed through a heating process using potassium hydroxide (KOH) solution. The results showed that the crude carrageenan content of seaweed was 62% can be seen in Annex 4. The resulting crude carrageenan content can be influenced by KOH concentration, cooking time, temperature, and seaweed particle size.

Produced SRC yield of 33.83% at 20 grams of 100°C temperature for 60 minutes found in the use of 8% potassium hydroxide concentration, so that in this study produces the highest yield due to the longer extraction time used, the longer the cooking time, the higher the yield produced [13].

3.3. Microcapsule Efficiency
Calculation of microencapsulation efficiency is used to determine the success rate of the microencapsulation process. The efficiency referred to in this study is the ratio between the extract of rosella petals and coarse carrageenan coating.
Figure 1. Effect of Microcapsule Ratio Ethanol Extract - Coarse Carrageenan on Coating Efficiency.

Figure 1. shows that the efficiency of ethanol extract microcapsules coated with coarse carrageenan produced by seaweed each ratio 1:3; 1:4; 1:5; 1:6 and 1:7 are 5.23%; 34.05%; 63.59%; 74.83% and 86.01% respectively. This shows that the highest efficiency is obtained at a ratio of 1:7 on the basis of (b/b) due to the more coating material, the higher efficiency. According to [14] states that one of the factors that cause high efficiency of microcapsules is the comparison between coating material and core material. Emulsifying concentration is also a factor that influences the efficiency of microcapsules. The viability test of lactobacillus casein encapsulation using a matrix of kappa carrageenan produced the highest efficiency value at a concentration of 2% is 60.49% compared to the other two concentrations (1% and 1.75%) [15], so the results of this study are higher than those of them.

Coating material used must have high solubility and emulsifying ability, and must be able to form a film layer, and produce a high concentration solution. In addition, coating material must be able to protect the active ingredients from oxidation, heat, light and moisture. Coarse carrageenan coating material has high solubility and emulsifying ability so that the active ingredient of rosella flower petals can be coated [16].

Based on variance data can be seen, the value of microcapsule efficiency of rosella petal extracts coated with coarse carrageenan with various ratios has a significant value <α (0.05), then the ratio treatment has a significant effect on coating efficiency, followed by Duncan test. Duncan's further test results can be seen in the ratio 1:3; 1:4; 1:5; 1:6 and 1:7 ratios were significantly different from the efficiency value at the use of a 1:7 ratio which was expressed as the best result (86.01%).

3.4. Microcapsule Morphology

Morphological observations of microcapsules of rosella flower petals extract with coarse carrageenan coating were carried out using SEM (Scanning Electron Microscope). The observation results of microcapsule morphology can be seen in Figure 2 as follows:
Based on the analysis using SEM (Scanning Electron Microscope), the morphology of coarse carrageenan microcapsules and coarse-ethanol-carrageenan extract microcapsules with 1000 x magnification showed aggregate form. Aggregate is a coagulation between particles with one another, that an increase in the number of semi refined carrageenan will result in an increase in the amount of aggregate formed due to the double helix [17]. The more double helix will produce a large amount of aggregate in the form of very strong meshes, resulting in the structure of the gel becoming harder and harder.

Microcapsules have various forms including spherical spheres and irregular shapes. Microcapsules which have both spherical and irregular shapes can be classified into 3 types, namely mononuclear, polynuclear and matrix. Pure ethanol-carrageenan extract microcapsules 1000 times the magnification seen throughout the surface of the microcapsules which are irregular with the matrix type. The matrix type is a type of microcapsule containing the core material which is evenly dispersed in the coating material. Mononuclear is a microcapsule that contains core material which is surrounded by coating material, polynuclear is a microcapsule containing several core materials in its coating material, while the matrix is a microcapsule containing core material that is evenly dispersed in coating material [18]. The morphology of coarse carrageenan microcapsules and coarse ethanol-carrageenan extract microcapsules with 1000 x magnification cannot be expressed in 3 types of microcapsules due to aggregation.

The number of coatings used in microencapsulation can also affect the particle size produced. The more the amount of coating material compared to the core material, the larger the particle size will be because there is an increase in the thickness of the microcapsule wall formed. The microcapsule particle size produced from the ratio of core and 1: 1 coating materials was smaller at 71.10μm compared to the ratio of core and 1: 2 coating materials, namely 81.91μm. In addition, the particle size can be affected by the stirring speed used in making microcapsules [19]. The greater the stirring speed used, the smaller the microcapsule particle size produced, and vice versa. In this research the results of particle calculation, that the average value of pure ethanol-carrageenan extract microcapsules is 14.91 μm. Overall the results obtained have met the requirements for microcapsule particle size between 1-1000μm [20].

**4. Conclusion**

Based on the results of the research that has been done it can be concluded that the ethanol extract of rosella petals (*Hibiscus sabdariffa* Linn) can be coated with coarse carrageenan to produce microcapsules with a particle size of 14.91μm and the highest coating efficiency at a ratio of 1:7 is 86.01%.

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