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Synthesis of halal membrane capsule from water soluble chitosan by adding sodium lauryl ether sulphate

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Abstract. Membrane capsule has been synthesized from water soluble chitosan (WSC), agar, and Sodium Lauryl Ether Sulphate (SLES). The concentration of agar used 0.02% (v/v) and the concentration of SLES up to 0.16% (v/v). WSC has been synthesized from shrimp shells, shrimp shells through demineralization, deproteination and deacetylation. After going through 3 processes chitosan converted into WSC by depolymerisation process. Degree of deacetylation (DD) of WSC was calculated based on FTIR baseline method, the value of DD is 77.9%. Based on FTIR spectra showed that the obtained film capsule shell had vibration from its constituent molecules, i.e. chitosan, agar and SLES. Based on tensile test data, it showed that the increasing of SLES concentration would increase the elasticity of shell matrix. Swelling test data in water and 0.1 N HCl showed the greatest value in capsule film with SLES concentration 0.12% is 420.27% and the capsule can be degraded (broken) in 20th minutes. Capsule membrane with SLES concentration 0.12% has the largest degradation value in water and 0.1 N HCl that is 70.91% and 81.32%. Based on research data obtained that the membrane with SLES concentration 0.12% that most appropriate with commercial capsule.

Keywords: Capsule shell; Water-Soluble Chitosan, SLES; FTIR; tensile test; weight uniformity test; swelling; degradation.

1. Introduction

Capsule is one of the oldest forms of pharmaceutical that found by ancient Egyptians. There are two types of capsule, hard shell capsule and soft shell capsule. Hard shell capsules are commonly used, about 60 billion capsules are produced annually in industry pharmaceutical [1]. Hard capsules are in the first place in drug development, because of simpler production process. Capsules have more advantages than other oral, like as combination material according by need of patient and dose. The properties of hard shell are odourless and tasteless have the advantage of being easy to swallow and release in the appropriate time [2].

The capsule was first made by J.C Lehuby in 1846 [3]. The material used in the pharmaceutical industry is gelatine [4]. Based on data Gelatine manufactures of Europe 2005, the largest gelatine production is derived from pigskin by 44.5% (136,000 tons), second from cowhide by 27.6% (84,000 tons) and the remaining 1.3% (4,000 tons) comes from bones of fish and goats [5]. The data show that most of the gelatine obtained from cow and pig. This is a problem for people about halalness of food. Halal is important for muslims. Foods derived from pork are forbidden for consumption [6]. In addition to muslims, gelatine also cannot be consumed for Hindus and vegetarians. Gelatine also has the risk of viral contamination causes bovine spongiform encephalopathy (BSE), foot and mouth disease (FMD)



and swine influenza [7]. Therefore, gelatine substitution is needed as the base material of hard shell capsule.

Chitosan is natural product of chitin biopolymer, which is the second highest after cellulose. Chitin is commonly found in insect, crustacean and fungi. Chitosan has the characteristic of non-toxic, biodegradable and biocompatible. Chitosan also have many benefits in daily life such as heavy metal waste adsorbents and dyes, preservatives, anticancer and antibacterial [8]. Chitosan can be used as a gelatine replacement capsule shell, but has a longer disintegration time than gelatine. Thus, chitosan is suitable for drugs are broken out intestines. Based on research [8] reported capsules have anti-fungal properties, so it has added value compared to gelatine. The chitosan not soluble in water and slightly soluble in HCl. Therefore, in this study water soluble chitosan (WSC/ Water Soluble Chitosan) used as the basic material of shell membrane. The addition substances are agar as gelling agent and SLES (Sodium Lauryl Ether Sulphate) as filler for making capsule membrane.

2. Experimental

2.1. Material

The materials used in research were shrimp shell waste, NaOH 100%, NaOH 98%, HCl 37%, distilled water, ethanol 96% (SAP chemicals), ethanol absolute (Merck grade EMSURE), H₂O₂ 30% (Merck grade EMSURE), glacial acetic acid 100% (Merck grade EMSURE) and Agar (AA)

2.2. Chitin extraction

Shrimp shell waste was cleaned from the head and the meat, then the shell washed with soap water and soaked for ± 1 hour. Shrimp shell dried in the temperature 50°C for 12 hours. Dried shrimp shell mashed with blender and sieved [9].

Chitin extraction from shrimp powder was done based on the research method [10]. Shrimp shell powder (40 mesh) weighed 50 g. The shrimp powder was immersed in 500 mL of HCl % (v/v) at room temperature for 24 h, then filtered and washed the shrimp shell with demineralized aqua to neutral pH. After that, shrimp shell immersed in 500 mL NaOH 10% (w/v) at 60°C then filtered and washed with demineralized aqua to neutral pH. Immersion in HCl 7% and NaOH 10% was repeated in 2 times. After the residue is neutral, it washed with 250 mL 96% ethanol and dried in oven at 50°C for 8-12 hours. The dried residue is called chitin and yield of chitin is calculated with equation 1

$$\text{Chitin (\%)} = \frac{\text{mass of chitin (g)}}{\text{mass of powder shrimp shell (g)}} \times 100\% \quad (1)$$

2.3. Synthesis of Water Soluble Chitosan (WSC)

Synthesis of water-soluble chitosan from chitin based on research method [10]. The chitin weighed as much as 10 g. The chitin is immersed with 20 mL NaOH 50% (w/v) at 60°C for 8 h. The residue was filtered and washed with hot demineralization aqua (60°C). The residue is dried at 50°C for 8-10 h, and the dried residue obtained is called chitosan.

The chitosan was weighed 1 g, then 20 mL of 2% acetic acid is added to the weighed chitosan. The chitosan and acetic acid mixture were stirred with hotplate magnetic stirrer at 40°C. When temperature reaches 40°C, 30% H₂O₂ is added as much as 1 mL and treated 4 h. After that, NaOH 10% added to obtain neutral pH. Then, the mixture filtered and washed with demineralization aqua. The product was added with absolute ethanol twice the volume of filtrate. The mixture is incubated in refrigerator for 24 h in a closed plastic wrap.

2.4. Determination of deacetylation degree

Determination of DD using FTIR spectrophotometric baseline method was proposed by baxter. Value of DD shown in equation 2

$$\text{DD (\%)} = 100 - \left(\frac{A_{1655}}{A_{3450}} \times \frac{100}{1.33} \right) \quad (2)$$

2.5. Synthesis of membrane capsule

Water-soluble chitosan (WSC) which has been incubated, then be filtered and heated at 50°C for 30 minutes to purified ethanol. Water-soluble chitosan (WSC) that has been purified from ethanol, stirred using a hotplate magnetic stirrer until homogeneous. Then a water-soluble chitosan (WSC), can prepared to agar solution. The mixture is stirred with a hotplate magnetic stirrer for ± 1 hour. Water soluble chitosan (WSC) with added SLES 0.04; 0.12;0.16% (v/v) of 2 mL, stirred for ± 30 min.

2.6. Characterization FTIR

The Characterization of FTIR is to identify functional groups of synthesis product. Characterization was carried out at wave numbers 400- 4000 cm^{-1} . Samples were mixed with KBr solids, then KBr samples and solids were homogenized with mortar and pestle. The homogenized sample are compressed by a hydraulic pump and moulded a thin pellet. Then, the pellet is placed in the pellet holder and inserted in the FTIR holder. Thus, the FTIR spectra are obtained in the form of curves between the wave numbers as the x-axis and the transmitters as the y-axis. Chitin FTIR spectra, chitosan, WSC and WSC-SLES were then compared with literature.

2.7. Degradation Test

The degradation test is carried out by drying the membrane in the oven to obtain dry weight. Membranes cut to $\pm 1 \times 1$ cm were then immersed in water and HCl for 5 to 30 minutes, after being soaked the membrane was dried in an oven. The dried membrane is weighed, then the percentage of degradation is calculated using equation 3 [11]

$$\text{Degradation (\%)} = \frac{\text{Final mass-Intial mass}}{\text{Intial mass}} \times 100\% \quad (3)$$

2.8. Swelling degree test

The degree of swelling test is done by drying the membrane in the oven, so that the dry weight is obtained. The membrane is immersed in water and HCl to obtain wet weight. Wet weight and dry weight are included in equation 4 to obtain the degree of membrane swelling [12]

$$\text{Swelling degree} = \frac{\text{Wet mass-Dry mass}}{\text{Dry mass}} \times 100\% \quad (4)$$

2.9. Weight Uniformity Test

Capsule shell weight testing to determine the thickness of the capsule shell. The thicker the capsule shell will increase the weight of the capsule shell. Capsule shell weight is one of the standards to be met by the commercial capsule shell. Commercial standards of capsule shell weight are set at 69-83 mg/100 shells. Under Farmakope Indonesia terms, the difference in mass of each membrane to the average mass should not exceed 10% for an average mass of 120 mg and 7.5% for an average mass of 120 mg.

3. Result and Discussion

3.1. Water Soluble Chitosan (WSC) synthesis

Shrimp powder that through the process of demineralization and deproteination is called chitin. Chitin is converted into crude chitosan through the deacetylation stage. The method used in deacetylation uses alkaline solutions. Deacetylation is the process of converting an acetyl group ($-\text{NHCOCH}_3$) to an amine group ($-\text{NH}_2$) with the addition of a strong base such as NaOH. Based on research [13] reported that high NaOH concentrations can produce chitosan with a high deacetylation degree. Therefore, in this study using 50% NaOH with a temperature of 60°C for 24 h. To find out about the deacetylation process, determination of the deacetylation process (DD) using the FTIR baseline method which was added by Baxter. Based on these calculations obtained DD of 77.9%. The process of converting chitosan into

WSC through the depolymerisation process. Depolymerisation is the cutting of the polymer chain to be shorter. In this study, the depolymerisation process uses H_2O_2 .

3.2. Characterization FTIR of chitin, chitosan and WSC.

Chitin and chitosan spectra from this study have absorption with a pattern that is almost the same as the results of research conducted in the study [14]. Chitin spectra have specific peaks that show functional groups in chitin. In the 3446 cm^{-1} wave number which shows the stretch vibration of $-OH$. The peak of 2931 cm^{-1} shows the vibration of $C-H\ sp^3$. The wave number 1658 cm^{-1} shows the vibration of $C=O$ amide, while the vibrations of 1550 and 1381 cm^{-1} show the vibration of $N-H$ amide. The 1074 and 1028 cm^{-1} peaks show the vibration of the $C-O-C$ bond.

In contrast to chitin FTIR spectra, chitosan FTIR spectra did not have absorption of 1658 , 1550 , 1381 cm^{-1} which showed that there was a deacetylation process. There are several shifts in the absorption of wave numbers between chitosan and WSC, because the WSC is unstable and easily absorbs moisture in the air [10]. Images of chitin, chitosan and WSC FTIR spectra are shown in Fig. 1.

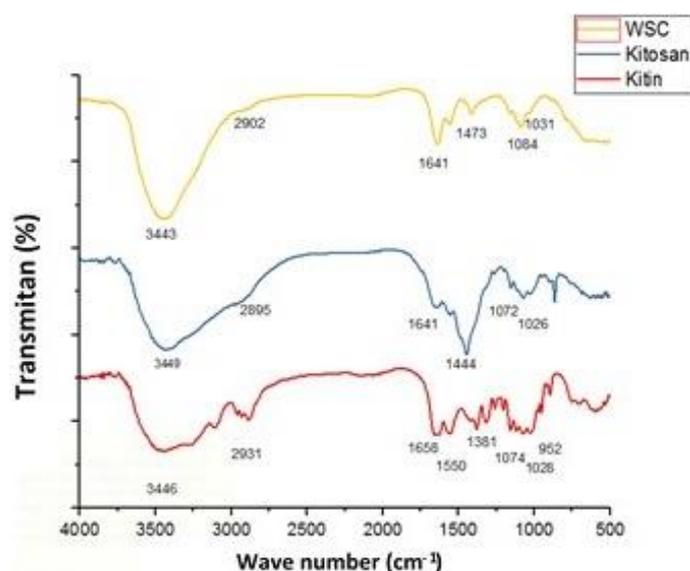


Figure 1. FTIR Spectra of chitin, chitosan and WSC.

3.3. Synthesis and Characterization of membrane capsule

The process of making capsule shells is carried out on the basis of WSC, this research combines 6.5% WSC (w/v) with agar and Sodium Lauryl Ether Sulphate (SLES). The addition to function as a gelling agent and added agar concentration is 0.02% (v/v). The SLES addition function is as a wetting agent on the capsule shell membrane, with a variation of SLES concentration of 0; 0.04; 0.12; 0.16% (v/v). The bond between chitosan and SLES is shown in Fig. 2

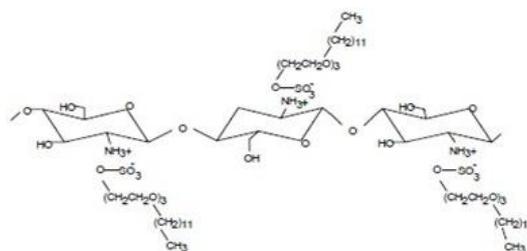


Figure 2. Structure of Chitosan-SLES [15].

FTIR spectra of WSC-agar membranes and WSC-agar-SLES membranes generally have the same absorption as the WSC FTIR spectra shown in Fig. 3. In the WSC-Agar membrane there is an absorption of wave numbers that is 3448 cm^{-1} which shows the vibration of -OH. At the peak of 2931 cm^{-1} is the vibration of C-H sp^3 , the wave number 1629 cm^{-1} is the vibration of primary amine, the absorption at wave number 1545 is the vibration of N-H amide and this indicates the WSC reaction with agar. In WSC-Agar-SLES FTIR spectra there is a new absorption, that is at wave number 1254 cm^{-1} which is the vibration of S = O and 1087 cm^{-1} is the vibration of $-\text{SO}_3$ [15]. This indicates a reaction between SLES and WSC

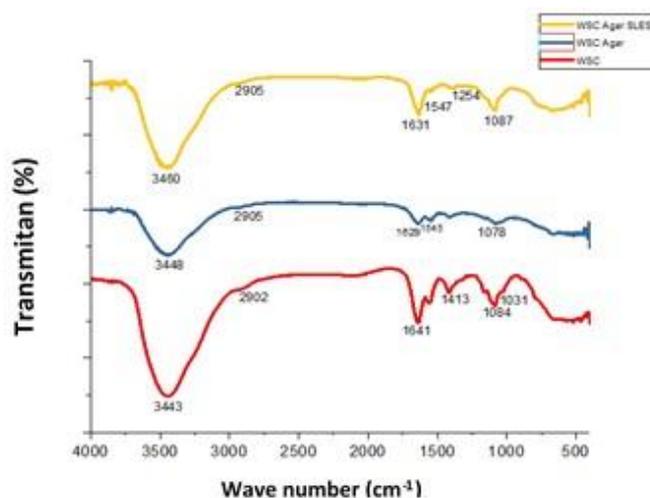


Figure 3. Spectra of WSC, WSC-Agar, WSC-Agar-SLES.

3.4. Swelling Test

Swelling degree test is used to determine the effect of SLES composition on percent of water absorbed by capsule. The swelling degree test was performed by immersing the capsule shell membrane in 50 mL water and 50 mL of HCl 0.1 M. The reason for the use of water and HCl media is because it adjusts the state of the human body which is mostly water [16] and HCl 0.1 M adjusts gastric state which has a pH of 1- 1.5 [11].

3.4.1. Water as a Swelling Test Media. The result of swelling degree test using water media, shown in Fig. 4. Based on Fig. 4 at 10 minutes swelling, the greater the SLES concentration, the higher the degree of swelling. This shows that the higher the SLES concentration, the greater the ability to adsorb water [17]. The highest swelling degree value at 10 minutes was 243.30% with a concentration of 0.16%. At 20 minutes swelling time, the highest swelling value was 420.27% at a concentration of 0.12% and a decrease in the degree of swelling at a concentration of 0.16% was 301.18%. At 30 minutes swelling time, the highest swelling value was 366.24% at a concentration of 0.12% and there was a decrease in the concentration of 0.16% at 234.88%. The cause of the decrease, in the degree of swelling at the concentration of SLES 0.16% is the saturated state of the SLES which causes a decrease in the ability to adsorb water. Based on Fig. 4, the concentration of 0.12% is the optimum concentration on the degree of swelling in the water medium

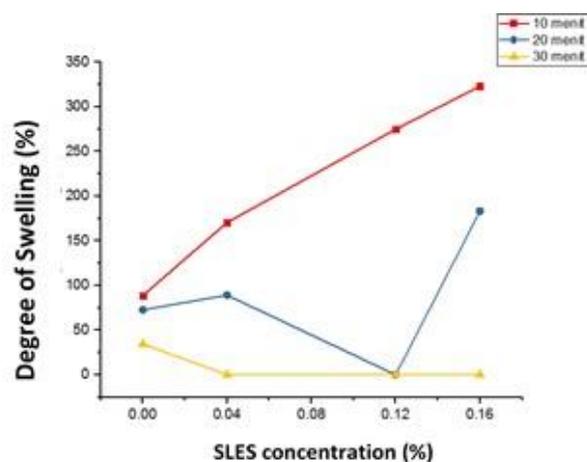


Figure 4. Swelling Degree of membrane shell in water medium

3.4.2. HCl as a media for Swelling Test. The result of swelling degree test using HCl media, shown in Fig. 5. Based on Fig. 5 with a 10 minutes swelling time, the higher the concentration of swelling increases, the highest degree of swelling is 323% at a concentration of 0.16%. At swelling time 20 minutes the highest level of swelling was 0.16% at 183.32% but at 0.12% at 20 minutes the membrane broke. This is because at 20 minutes HCl solution is able to break the bonds of the capsule shell membrane. At 30 minutes the capsule shell membranes of 0.04 and 0.16% concentrations broke, it also indicated that 30 min time was the crushed time of the capsule shell membrane.

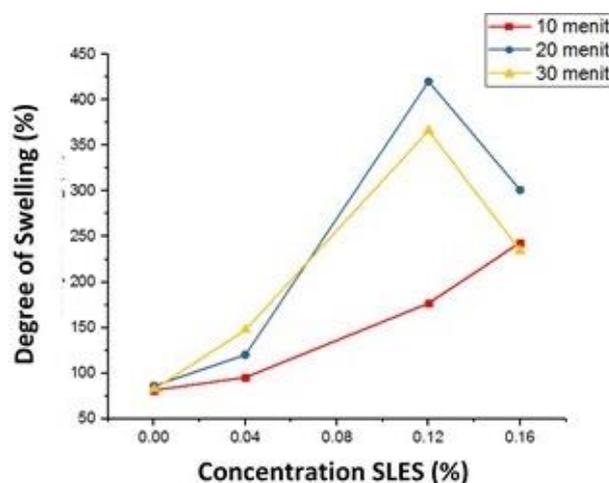


Figure 5. Swelling Degree of membrane capsule in HCl medium

3.5. Tensile Test

Another characterization carried out in addition to the swelling degree test is the tensile test. This is done to determine the resistance of the membrane when pulled in a certain style [13]. Tensile test results on capsule shell membranes with variations in SLES concentration are shown in Table 1

Table 1. Data of tensile test

SLES Concentration (%)	Stress* (N/m ²)	Strain*	Modulus Young* (N/m ²)
0.00	48,420.33	0.03425	1432847.7
0.04	2,451.663	0.13975	16368.033
0.12	3,677.494	0.158	23331.411
0.16	2,451.663	0.295	8300.598

Based on Table 1, the higher the concentration of SLES also increases the value of the strain. The value of the strain shows the elasticity value of a shell [12]. This is because the plasticizer will occupy intermolecular space in the polymer chain [16]. Therefore, the addition of SLES functions as a plasticizer and the addition in higher concentrations causes the polymer chain to become more saturated and cause the membrane to become more plastic. The results of the tensile test other than strain are stress and modulus young. Stress is the maximum tensile force that a membrane can hold. This parameter describes the maximum force that occurs in the membrane during measurements [18].

Based on Table 3, the concentration of SLES 0% has the highest stress value of 48,420.33 Pa, then a fairly high decrease in the concentration of 0.04; 0.12 and 0.16%. In 2018, Rachmawati [19] reported that high stress values exhibit rigid shell properties, this is because the inter-polymer chains of the constituents are tightly bound together. As for the difference in value of stress SLES 0% with SLES 0.04; 0.12; 0.16% is 45,968,697; 44,742.84; 45,968.7 Pa. This decrease in stress value is due to the presence of plasticizers that occupy space between molecules in the polymer chain, so that it can reduce the energy needed to carry out the movement and cause the tensile strength to decrease. Plasticizer will act as a lubricant causing the value of the tensile strength to decrease [16].

When compared between SLES 0.04 concentrations; 0.12; 0.16%, the highest stress value at the concentration of SLES 0.12% which is 3,677.49 Pa. There was an increase in stress value of 1,225.86 Pa from a concentration of 0.04% and a decrease in 0.16% occurred at 1,225.86 Pa. This shows that the capsule shell membrane with a concentration of 0.12% SLES has the best resistance force.

Based on Table 3.1, the biggest modulus young value is at 0% concentration, which is 1,432,847.7 Pa and there is a significant decrease at 0.04% concentration; 0.12% and 0.16%. At a concentration of 0.12% there was an increase of 6,963.37 Pa compared to 0.04% and decreased again to 15,030,813 at a concentration of 0.16%. Modulus young values are influenced by stress and strain values. This is consistent with Table 4.3, with the addition of SLES decreasing the modulus young value, which is due to the decrease in stress value and the increase in the value of the strain. In Rachmawati [19] reported that with the addition of plasticizers, the material became more plastic and reduced the value of the capacity degree. Therefore, based on the degree of swelling test and data of tensile test showed that the capsule shell membrane with SLES 0.12% concentration had the best physical resistance

3.6. Degradation Test

The degradation test is a physical test of the capsule shell membrane in addition to the degree of swelling test and tensile test. This degradation test aims to find out the mass lost in a certain time. The time range used in the degradation test is 10 - 30 minutes, this is because the disintegration time demanded by Farmakope is generally 15 minutes or 30 minutes [4]. In this study using the method from [11], using two media, namely water that adjusts the body state which is partially water and the condition in the mouth and 0.1 M HCl which adjusts the stomach state with a temperature of $\pm 37 - 38^{\circ}\text{C}$ according to human temperature. The degradation test results on water and HCl media are shown in Fig. 6 and 7

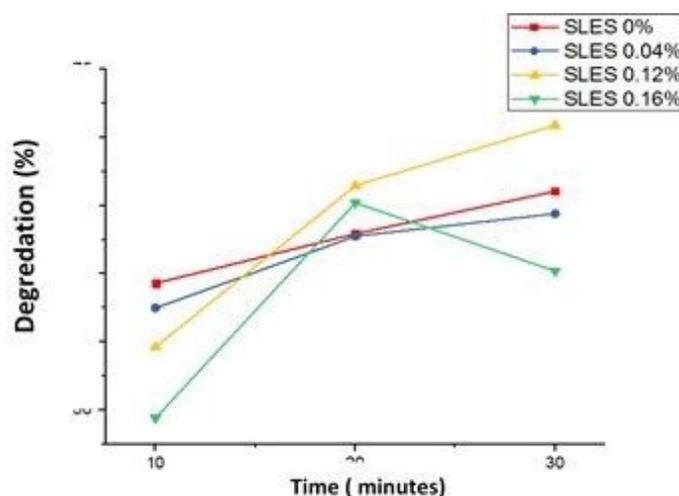


Figure 6. Degradation of membrane shell in medium water.

Based on Fig. 6 the value of the percentage of degradation increased with the length of time, except at a concentration of 0.16% there was a decrease in the 30th minute. At the concentration of SLES 0% the greatest percentage degradation value at 30 minutes was 66.05%, concentration of 0.04% at 64.41% at 30 minutes, at a concentration of 0.12% at 70.91% at 30 minutes and 0.16% at 65.2% at minute 20. The greatest percentage of degradation was at a concentration of 0.12% and decreased at a concentration of 0.16%, this shows the optimum concentration of SLES addition at 0.12% and saturation occurs on the capsule shell membrane so that its ability decreases. The higher the SLES concentration causes the more difficult to dissolve in water [24]. In the degradation process in the water medium, no broken capsule shell membrane was found. This shows that water is unable to break down the chemical bonds between the constituent molecules of the capsule shell membrane.

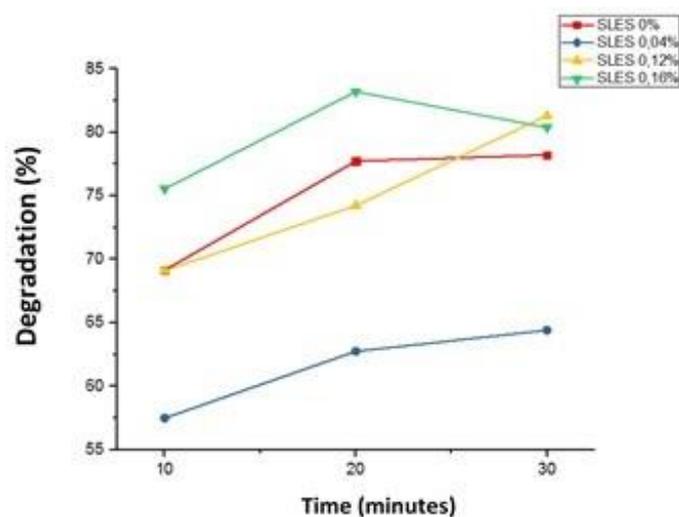


Figure 7. Degradation of membrane shell in HCl medium.

According to Fig. 7, the longer degradation time of degradation of each degradation of concentration variation also increases, except at 0.16% concentration decreasing at minute 30. It has the same pattern with degradation in water medium. At concentration 0% biggest percentage is 78.22 at minute 30, at concentration 0.04% is 64.41 in minute 30, at concentration 0,12% is 81.32 at minute 30, at concentration 0.16% is 83.21 in the 20th minute. When compared between Fig. 6 and 7, the percentage

of degradation in HCl media is higher than that in water media and HCl is capable of causing the shell of the caulpritt shell membrane. This is because HCl has destructive properties in certain materials such as metals, minerals and biopolymers [19].

Based on the degradation test data, the capsule shell membrane with SLES concentration 0.12% was able to break at minute 20, while the concentration of 0.04 and 0,16% broke at minute 30. Thus, membrane with SLES concentration 0,12% had ability the best degradation. Therefore, based on the swelling degree test, tensile test and degradation test showed that the capsule shell membrane with SLES concentration of 0.12% is the best capsule shell formulation.

3.7. Weight Uniformity test

Based on Table 2, capsule membrane has fulfilled the requirements of uniformity of weight, where the percentage of mass difference is average with uniformity of weight below 10%. At a concentration of 0% SLES had a percentage difference of 1.99%, at a concentration of SLES 0.04% at 2.24%, SLES concentration of 0.12% at 2.71% and a concentration of 0.16% at 2.138%. The highest percentage of weight uniformity at a concentration of 0.12%.

Table 2. Weight Uniformity membrane shell in variation concentration SLES

No	SLES (%)	Average Weight of membrane (gram)	Difference (%)
1	0.00	0.0223	1.990
2	0.04	0.0125	2.240
3	0.12	0.0128	2.710
4	0.16	0.0123	2.318

4. Conclusion

Water-soluble chitosan (WSC) can be used as the main ingredient in capsule making with the addition of agar as a gelling agent and variations in SLES concentration. Based on tensile test show that SLES as filler also plasticizer. Data degree of membrane swelling with a concentration of 0.12%, is the best concentration with the degree of swelling in the medium of water is 420.27% and breaks at 20 minutes in 0.1 N HCl media.

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