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93550 Khairunnisa

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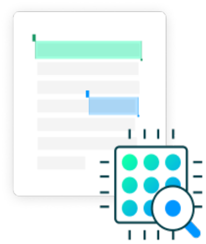

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The Characterization of Capsule Shell from Acid-Hydrolyzed Palm Oil Starch

Amalia Khairunnisa*, Pratika Viogenta, Nani Kartinah, Desy Aulia Azzahra

Department of Pharmacy, FMIPA, Lambung Mangkurat University
Jalan A. Yani Km. 36, Loktabat Selatan, Banjarbaru Selatan, Banjarbaru, Kalimantan Selatan, 70714, Indonesia

**Corresponding author: amalia.khairunnisa@ulm.ac.id*

Keywords:

Capsule Shell; Characteristics; Decolorization; HPMC; Modification by acid hydrolysis; Palm Oil starch.

ABSTRACT. Modification of palm trunk starch as film agent by acid hydrolysis can increase amylose content in starch so the product is stronger and more stable. This study aims to obtain the best modified starch concentration ratio with HPMC and determine the characteristics of starch modified capsule shell with HPMC. The method used in this study were modifying palm trunk starch with acetic buffer by using CH_3COONa and CH_3COOH, decolorizing starch with activated carbon, and manufacturing of capsule shell with variations in the concentration of modified stach and HPMC 1:1 (F1), 2:1 (F2), and 3:1 (F3). Organoleptic test results of all formulation produced firm and elastic capsule shells. The capsule size results were consistent with an average weight of F1 (0,10 grams); F2 (0,11 grams); and F3 (0,14 grams). The average results of disintegration test are F1 (10 minutes, 27,57 seconds); F2 (6 minutes 47,06 seconds); F3 (4 minutes 34,24 seconds). Tensile strength results are F1 (2,147 MPa); F2 (2,565 MPa); F3 (2,159 MPa). FTIR results show an absorbption peak at wave number 1560 cm^{-1}. SEM results show F2 capsule shell has the best morphology. The conclusion of this study are the capsule shell F2 (2:1) is the best formulation based on the results of organoleptic test, disintegration time, tensile strength and SEM and that the concentration of modified starch effecting the characteristics of the capsule shell.

INTRODUCTION

Palm Oil (*Elaeis guineensis*), a major commodity in Indonesia, provides many benefits, including starch found in the trunk (Bakewell-Stone, 2023; Pratama & Widodo, 2020). A single extraction from a 10 m long and 50 cm diameter trunk can yield up to 67% starch content (Cahyaningtyas *et al.*, 2019). In pharmaceuticals, this starch is used to produce capsule shells, typically made from gelatin or other polymers to create strong and elastic films (Watson & Cogan, 2019).

Research carried out by Azizah and Gracia (2023) shows the best capsule shell formulation using *E. guineensis* starch at 5% concentration with 5% of HPMC and 4% of glycerin (Azizah, 2023; Gracia, 2023). However, the disintegration test of 24 minutes did not meet the Indonesian Pharmacopoeia standard, which requires it to be 15 minutes or less. Research done by Mutia *et al* (2022) shows that modifying the *E. guineensis* starch using acid hydrolysis at pH 7 improves the starch content and may enhance the quality of the capsules (Azizah, 2023; Bhatt *et al.*, 2022; Mutia *et al.*, 2022).

Acid hydrolysis modification uses an acetate buffer, which is made up of sodium acetate and acetic acid (Villar *et al.*, 2017). This buffer solution is the simplest and most widely used method in both the food and non-food industries (Cahyaningtyas *et al.*, 2019). The mechanism of action is to break the glycosidic bonds in the amylopectin branching and increase the amylose content (Marseno *et al.*, 2022; Sjöö & Nilsson, 2018). This modification can reduce molecular weight, increase starch crystallinity, reduce viscosity, and increase solubility in warm water (Bambardekar, 2020; Kweon & Han, 2023). Considering these backgrounds, this study aimed to determine the best formulation and analysis the effect of modified *E.* starch concentration on capsule shell properties.

RESEARCH METHODS

Materials used in this study are oil palm (*Elaeis guineensis* Jacq.) trunk which was obtained from PT. Gawi Makmur, Sungai Danau, Satui, Tanah Bumbu, Kalimantan Selatan. Sodium acetate (CH_3COONa) p.a Merck was purchased in Nitrakimia (Yogyakarta), acetic acid glacial (CH_3COOH) p.a was purchased in Nurra Gemilang (Malang), HPMC K100, glycerine, and active carbon Iodine 100.

Extraction of *E. guineensis* Starch

The *E. guineensis* used are 20-25 year old male trees, with 2 meters sections taken from the top of the trunk. The pith in the trunk was crushed into sawdust, soaked in water at a 1:2 (%b/v) ratio, filtered using cloth, and settled for 24 hours. Wet starch forms at the bottom, was washed with distilled water and dried in an oven at 50°C for 3 hours. The dried starch was grinded and sieved using a 50 mesh.

Modification of *E. guineensis* Starch by Acid Hydrolysis

The acid hydrolysis modification of *E. guineensis* starch is done by Cahyaningtyas *et al.* (2019) and Mutia *et al.* (2022) using acetate buffer at pH 7. The buffer solution was made by dissolving 44.52 grams of sodium acetate in 50 mL of aquadest, added with glacial acetic acid to reach pH 7. Aquadest was added to the solution until the volume reaches 1 L. *E. guineensis* starch was dissolved with the buffer solution at a 1:2 (%b/v) ratio, stirred and heated on a hotplate at 40°C until it thickened. The starch was dried at 50°C using an oven, grinded and sieved using a 50-mesh sieve (Cahyaningtyas *et al.*, 2019; Mutia *et al.*, 2022).

The Manufacture of Modified *E. guineensis* Starch Capsule Shell

The formulation used in this study is done by Azizah and Gracia (2023) then developed by varying the concentration of modified *E. guineensis* starch with HPMC by 1:1, 2:1, 3:1 and 4:0 which presented in Table 1. HPMC was dissolved in $\frac{1}{3}$ of the total formulation volume of aquadest at 70°C, then left for 30 minutes. The modified *E. guineensis* starch was dissolved in 10 mL of aquadest and decolorized by mixing it with activated carbon at 1:0.1 (b/b) ratio for 10 seconds and filtered using filter paper. The filtered starch was added to HPMC solution, then added the excess of aquadest, and glycerin. Stirred the mixture using a magnetic hotplate stirrer at 40°C and 200-400 rpm for 5 minutes or until the mixture thickens. Capsul moulding was done using a cylindrical pin dipped 3 times, with a 3-second interval between dips. The capsule was left for 10 minutes at room temperature, then placed in oven at 55°C for 3 hours. The fully dried capsule shells were carefully removed and stored in a desiccator (Azizah, 2023; Gracia, 2023; Rizal *et al.*, 2023).

Table 1. Formula of modified *E. guineensis* capsule shell

Materials	Function	Amount of Materials Used			
		F1	F2	F3	Control
Modified *E. guineensis* starch	Film coating	1.25 g	2,50 g	3,75 g	5,00 g
HPMC	Gelling Agent	1.25 g	1.25 g	1.25 g	-
Glycerin	Plasticizer	1 mL	1 mL	1 mL	1 mL
Aquadest	Solvent	ad 25 mL	ad 25 mL	ad 25 mL	ad 25 mL

Characterization Test of Capsule Shell

Characterization test includes organoleptic test, capsule size (length, diameter, thickness, volume, and weight), disintegration test, Fourier Transform Infrared (FTIR), and Scanning Electron Microscop (SEM).

RESULTS AND DISCUSSION

Modification of *E. guineensis* Starch by Acid Hydrolysis

The modified *E. guineensis* starch powder has a darker color compared to the starch powder before modification that can be seen at Figure 1. This color difference occurs due to the presence of mono and disaccharides in the starch undergoing heating. The modified *E. guineensis* starch in this study was heated twice, first at 40°C when mixed with acetic acid, and second at 50°C during the drying process. The heating process results in the occurrence of caramelization reactions, including changes in the size of the monosaccharide ring, breaking and re-forming of glycosidic bonds, dehydration, or the inclusion of double bonds, leading to a darker color for the modified starch (Gullón *et al.*, 2016; Kocadağlı & Gökmen, 2018; Wardana & Yulia, 2018).

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(a) (b)

Figure 1. *E. guineensis* starch powder (a) before and (b) after acid-hydrolyzed modification.

Decolorization of Modified *E. guineensis* Starch

Decolorization results of the modified *E. guineensis* starch solution shown at **Error! Reference source not found.**. This process produces a solution that has better color with a light-yellow color, clear, and odorless when compared to the modified *E. guineensis* starch solution before decolorization which has a cloudy brownish color and characteristic smell of starch. Activated carbon used in the process adsorbs dyes and impurities through the pores of the adsorbent, causing the final product to lose its color. This process is carried out to modify the appearance of the product and enhancing the aesthetic value to increase consumer trust in the product (Hung *et al.*, 2023; Kan *et al.*, 2017) .

(a) (b)

Figure 2. *E. guineensis* starch (a) before and (b) after decolorization.

Characterization Test of Modified *E. guineensis* Capsule Shell

Capsul shell of modified *E. guineensis* starch formula 1, formula 2, and formula 3 are successfully obtained with good shape, firmness, and elasticity after passing the moulding and drying process with a slightly difference between the shape. The F1 capsule has an uneven oval shape, the F2 has an even oval shape, and the F3 also has an even shape but will become mushy after storing in a room temperature. The control formula generates a thin layer of capsule shell, making it difficult to separate from the moulding medium without causing damage. Capsule shells in this study can be seen at Figure 3.

The study examined the shape and properties of capsule shells manufactured by combining modified starch with HPMC. The control capsule shell made only from modified starch forms a thinner layer compared to the capsule shell made with the combination of modified starch and HPMC. It happens due to the ability of HPMC increasing viscosity of the capsule shell through hydrogen bonding, which subsequently traps water and produces a more compact outcome (Joshi, 2011; Yang *et al.*, 2022). Further research is required to optimize the manufacture procedures of this formula of capsule shells to adhere to industrial standards by paying attention to dissolution procedure and appropriate drying time and temperature (Prakoso *et al.*, 2023).

(a) (b) (c) (d)

Figure 3. Modified *E. guineensis* capsule shells (a) F1, (b) F2, (c) F3, and (d) control.

Organoleptic test

Organoleptic test uses to measure the shape, color, odor, and taste by relying on the five senses of the respondents to get accurate results (Dewi *et al.*, 2022). The results can be seen in Table 2 shows that increasing the concentration of modified *E. guineensis* starch did not affect the odor and taste of the capsule shell, but affected the color, shape, and time required for the formula to thicken. Increasing the concentration of the modified *E. guineensis* starch causes the cream color of the capsule shell fading and become more transparent. Capsule color clarity is likely due to the presence of acetyl groups from acid hydrolysis of *E. guineensis* starch modification, resulting in a higher clarity paste compared to unmodified *E. guineensis* starch (Sjöö & Nilsson, 2018; Teodoro *et al.*, 2015; Villar *et al.*, 2017).

The viscosity of capsule shell formulas (F1, F2, and F3) appeared visually identical during the moulding process but the duration for achieving that viscosity is varied. The time needed for the formulation to thicken decreased by increasing the concentration of modified *E. guineensis* starch at a constant temperature and stirring speed. The results suggest in this study that the modified starch contains more amylose compared to unmodified starch, leading to higher crystallinity and the formation of more intermolecular hydrogen bonds (Nisah, 2018). This contrasts with a previous study by Azizah (2023) on unmodified starch where viscosity depended on the composition of HPMC used and by increasing starch content resulted in a less viscous solution and a softer capsule shell (Azizah, 2023).

Table 2. The result of organoleptic test.

Formula	Color	Odor	Taste	Shape	Form of the mixture
F1 (1:1)	Cream color	Odorless	Tasteless	Oval, un-even shape	Thick
F2 (2:1)	Cream color	Odorless	Tasteless	Oval, even shape	Thick
F3 (3:1)	Cream color	Odorless	Tasteless	Oval, mushy in room temperature	Thick
 **Control** (4:0)	Cream color	Odorless	Tasteless	Oval, thin, soft	Not thick

Capsule size test

Capsule dimensions that can be seen in Table 3 were measured using a caliper to determine length, diameter, and thickness; and the capsule volume was estimated by filling the capsule to the upper meniscus limit with water using a measuring pipette. The results of all the capsules yielded results like the standard sizes as the capsule shells were adjustable during production.

The weight of each capsule was measured on an analytical balance. The overall weight of the capsule shells for all the 3 formulations was close to fall within the standard weight range for capsule shells, with a range of 0.096±10% grams than can be seen in Table 4 (Zilhadia *et al.*, 2022). Capsule shells in F3 weighted more when compared to F1 and F2. This results are due to variations in the added concentration of modified *E. guineensis* starch, leading to differences in the amount of dissolve starch in each formulation (Mutia *et al*., 2022). It is also possible that the increased weight of the F3 capsule shell is a result of the non-uniform distribution of the solution on the moulding surface, caused by the shorter viscosity time of the F3 formula compared to the other formulas. This variation in solution distribution could also be due to the manual and individual manufacturing process of capsule shell.

Table 3. The result of capsule size.

Parameters	Formula (Modified *E. guineensis* starch: HPMC)									Standard Capsule Size
	F1 (1:1)			F2 (2:1)			F3 (3:1)			
	I	II	III	I	II	III	I	II	III	
Cap Length (mm)	10.7	10.7	10.7	10.7	10.7	10.7	10.7	10.7	10.7	10.72
Body Length (mm)	18.4	18.4	18.4	18.4	18.4	18.4	18.4	18.4	18.4	18.44
Cap Diameter (mm)	7.6	7.6	7.6	7.6	7.6	7.6	7.6	7.6	7.6	7.65
Body Diameter (mm)	7.3	7.3	7.3	7.3	7.3	7.3	7.3	7.3	7.3	7.34
Cap Thickness (mm)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.107
Body Thickness (mm)	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.107
Volume (mL)	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.68

Table 4. The result of capsule weight.

Parameters	Formula (Modified *E. guineensis* starch: HPMC)									Standard Capsule Weight
	F1 (1:1)			F2 (2:1)			F3 (3:1)			
	I	II	III	I	II	III	I	II	III	
Weight (g)	0,11	0,11	0,10	0,11	0,11	0,11	0,15	0,14	0,15	0,096±10%
Mean±SD		0,10±0,005			0,11±0			0,14±0,005		

Disintegration test

The disintegration test determines how quickly a capsule breaks apart in human body fluids (Dewi *et al*., 2022). It shows how the mechanical breakdown of the capsule affects its surface area for a quick release of active ingredients (Floryanzia *et al.*, 2022). The test is performed using a disintegration tester with water as the media at a temperature of 37°C ± 1°C. The tester basket is filled with different formula of capsules and the vertical motion is applied until all the parts of the capsule disintegrate through the basket (Rizal *et al*., 2023). The test should take no more than 15 minutes according to the Farmakope Indonesia edition VI (RI, 2020).

The disintegration test for all formulations meets the requirements set by Farmakope Indonesia edition VI. Formula 1 had the longest disintegration time compared to formula 2 and formula 3, formula 3 showed the faster disintegration time compared to all formula, while formula 2 has disintegration time between the two formulations. The result can be seen at **.**

The disintegration time confirms the effectiveness of acid hydrolysis with an acetate buffer. Hydroxonium ions (H_3O^-) weaken the glycosidic bonds by attacking the high-energy oxygen atoms in the starch molecules, which allows water to bind and form new bonds. This destabilizes the hydroxyl groups, leading to the breakdown of amylopectin molecules into amylose molecules. As the degradation progresses, the amorphous part decreases, and new crystalline part is formed. This fragmentation also leads to an increase in the number of hydroxyl groups, making it easier for starch to dissolve in warm water. This combinations allow the product to have a firm physique while still dissolving easily in warm water (Bambardekar, 2020; Sjöö & Nilsson, 2018).

Table 5. The result of disintegration test.

Replication	Time (Minutes)		
	F1 (1:1)	F2 (2:1)	F3 (3:1)
1	10:20:23	06:41:17	04:15:25
2	10:27:20	06:48:24	04:57:41
3	10:36:07	06:51:38	04:30:05
Mean±SD	10:27:57±0,005	06:47:06±0,003	04:34:24±0,014

Tensile strength test

The tensile strength determines the ability of a sample to withstand a certain amount of applied force by maintaining the strength of the sample shortly before breaking (Tafa *et al.*, 2023). The test is perfomed using a set of equipment applied load to both ends of the sample until it was cut in half (Kweon & Han, 2023). The results of all capsule shells formulations shown at **Error! Reference source not found.** meet the minimum standard tensile strength value for packaging preparations in the form of edible films based on Japanese Industrial Standard (1997) of 0.392 MPa (Santoso & Atma, 2020). The tensile strength of capsule shell made of unmodified *E. guineensis* starch and HPMC with a ratio of 1:1 done by Gracia (2023) resulted in an average tensile strength of 0.976 MPa (Gracia, 2023). The tensile strength results of capsule shell made from modified *E. guineensis* starch shows a stronger and more elastic outcome due to the successful modification in starch (Mutia *et al*., 2022).

These results show by increasing the concentration of modified *E. guineensis* starch in the capsule shell formulations can increase its tensile strength. It happens due to stronger hydrogen bonding between hydroxyl groups of modified starch and HPMC with increased concentrations, leading to greater strength and elasticity. However, at specific concentrations this interaction weakens, resulting in a decrease in tensile strength as observed in the case of F3 capsule shell. This may be attributed to the optimal ratio of modified starch and HPMC being reached at a concentration of 2:1, beyond which additional modified starch has no further impact on the structure and ingredients compatibility. Modified starch in F3 capsule shell formulation weakened the interaction between the molecular matrices, resulting in reduced tensile strength and heightened water absorbability (Frangopoulos *et al*., 2023; Mutia *et al*., 2022).

Table 6. The result of tensile strength test.

Tensile Strength	Formula (Modified *E. guineensis* starch : HPMC)		
	F1 (1:1)	**F2 (2:1)**	**F2 (2:1)**
kgf/cm^2	21,898	26,161	22,025
MPa	2,147	2,565	2,159

Fourier Transformed Infra Red (FTIR)

Fourier Transformed Infra Red (FTIR) is used to identify the functional groups present in molecules by analyzing their interactions within specific fingerprint regions. The FTIR results of modified *E. guineensis* starch shown at **Error! Reference source not found.**. A study conducted by Gracia (2023) found similarities in the shape and functional groups of modified to unmodified *E. guineensis* starch. However, a significant difference was observed in the absorption peak at 1556.15 cm^{-1}, indicating the presence of R-COO groups in the modified starch. This finding aligns with the previous research by Mutia *et al* (2022) and Sakeer *et al* (2017), which also identified peaks with wavenumber 1560 and 1556 cm^{-1} respectively (Gracia, 2023; Mutia *et al*., 2022; Sakeer *et al*., 2017). These peaks indicate the presence of acetate groups with a weak and sharp spectra. The literature indicates that the acetate group appears in the wavenumber range of 1550-1620 cm^{-1} for asymmetric COO stretching group and 1300-1420 cm^{-1} for the symmetric COO stretching group, while the carboxylic acid group appears in the 1700-1725 cm^{-1} range for the C=O stretching group (Ibrahim *et al*., 2005; Nandiyanto *et al*., 2023).

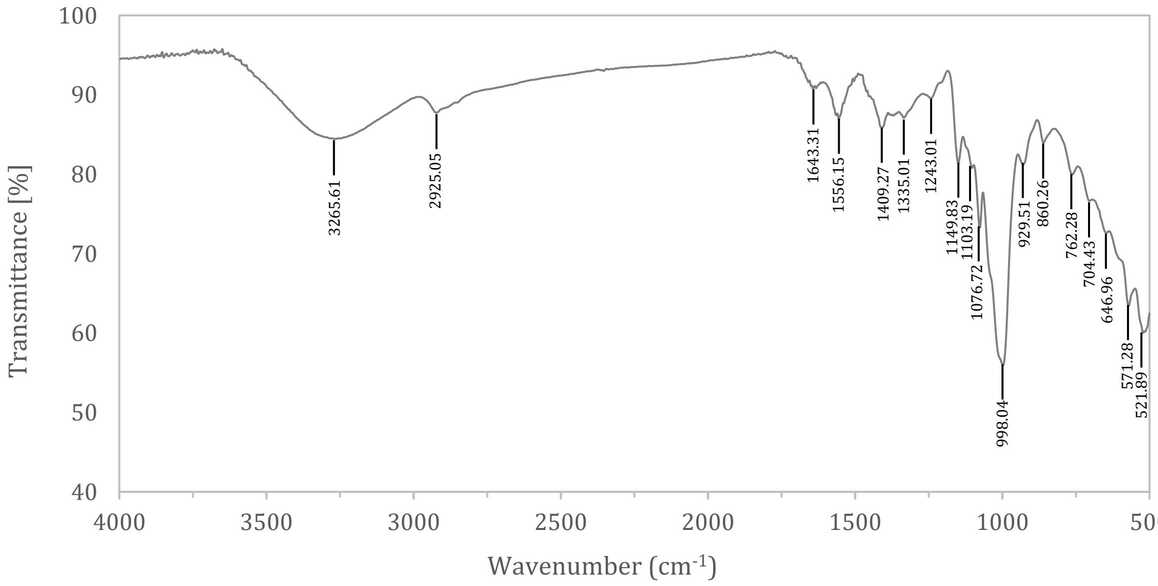


Figure 4. FTIR spectra of modified *E. guineensis* starch.

The FTIR results of modified *E. guineensis* capsule shells shown at **Error! Reference source not found.**. The spectra obtained are similar in shape and functional groups to the unmodified starch capsule shell by Gracia (2023) (Gracia, 2023). Significant differences were also noticed in the peaks formed as a marker of the presence of acetate groups as found at wavenumber 1565.93 cm^{-1}, 1564.80 cm^{-1}, and 1564.49 cm^{-1} for F1, F2, and F3 respectively (Mutia *et al*., 2022).

Acetate groups present in the modified starch after the successful acid hydrolysis using acetate buffer solution. The process involves the hydronium ions breaking the glycosidic bonds in starch molecules and attracting water molecules, leading to the fragmentation of the molecules into smaller fragments. The reactive hydroxyl groups in starch are substituted by acetate groups through an addition-elimination mechanism (Haq *et al.*, 2020; Kusumaningsih *et al.*, 2023; Subroto *et al.*, 2023).

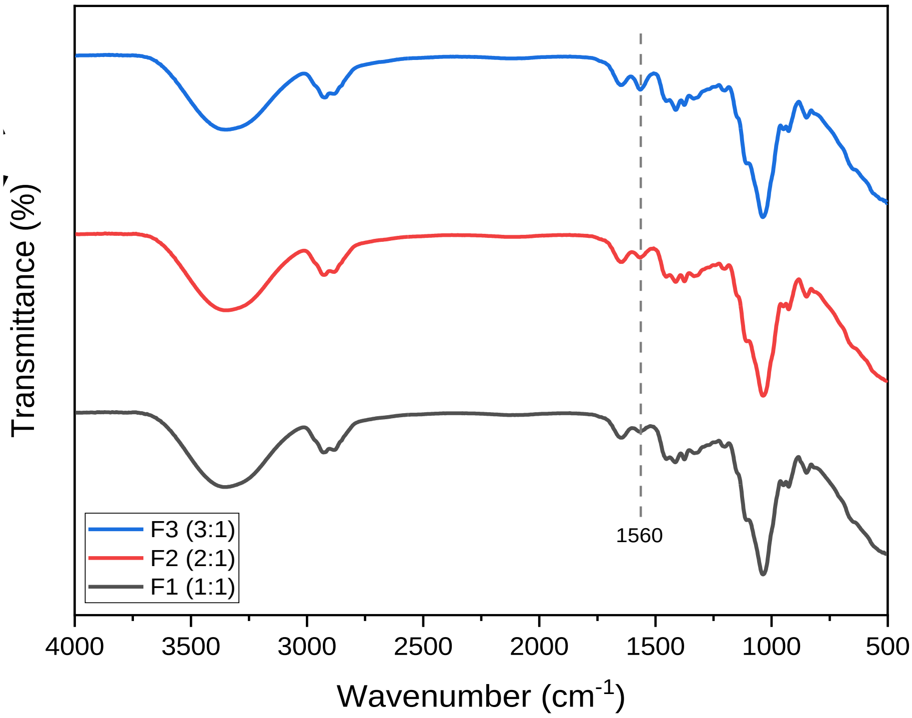


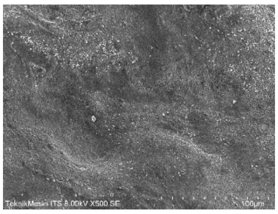
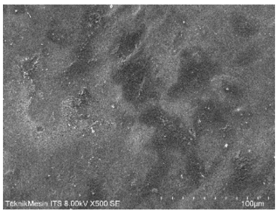
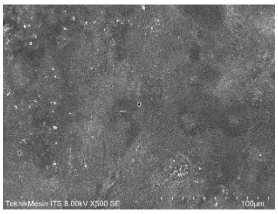
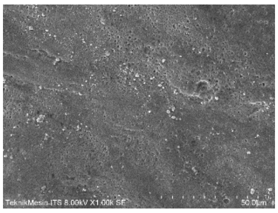
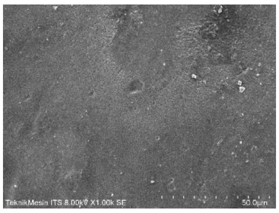
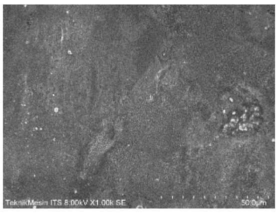
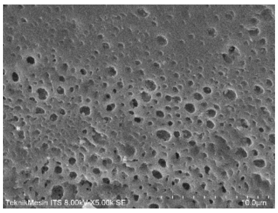
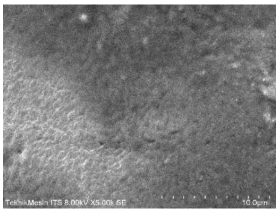
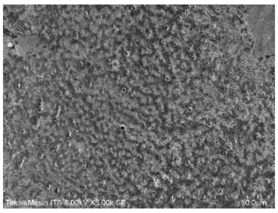
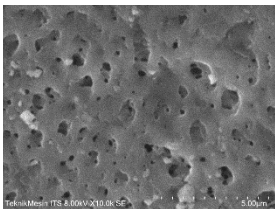
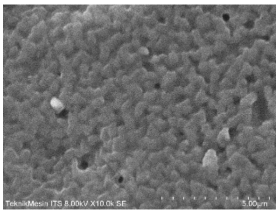
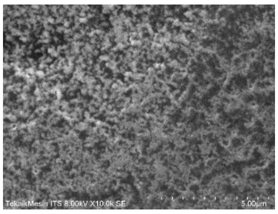
Figure 5. FTIR spectra of modified *E. guineensis* capsule shells.

Scanning Electron Microscope (SEM)

Scanning Electron Microscope (SEM) was conducted to observe the effect of modified *E. guineensis* starch on the surface morphology of the capsule shell microscopically. The results of morphological observations shown at Table 7 show that the F1 capsule shell has many pores and is larger when compared to other formulation capsule shells, observations on the F2 capsule shell is seen to have the least pores with a bumpy surface, while the morphology on the F3 capsule shell is seen to have an irregular surface and smaller pores compared to other capsule shells.

The large pores found in the F1 shell can be caused by the ratio of materials used not being able to cover all parts of the capsule shell. This can occur because the formula used has not thickened completely. The result of the F2 capsule shell has the best morphology that is closely resembles the polyamide capsule shell from the Essawy & Tauer (2010) research, with a bumpy surface. This similarity is caused by the ratio of modified starch and HPMC are able to bind each other perfectly, which reduces the number of pores and ensures complete coverage of the capsule shell surface. The irregular surface and smaller pores in F3 can be caused by the difference in shell thickness which is able to close the pores gradually in some parts, resulting in an irregular shape and fewer visible pores (Essawy & Tauer, 2010).

Table 7. The result of SEM on capsule shell formulations.

Magnificient	Morphology of Capsule Shell		
	F1 (1:1)	**F2 (2:1)**	**F3 (3:1)**
500x			
1000x			
5000x			
10,000x			

CONCLUSION

The best concentration of modified *E. guineensis* starch and HPMC with a ratio of 2:1 based on its disintegration time in average of 6 minutes and 47.06 seconds, tensile strength 2.565 MPa, and has the best morphological surface of capsule shell compared to the other formulations. There is an effect of modified starch concentration on organoleptical characteristics, capsule shell weight, disintergration time, tensile strength, FTIR, and SEM, however there is no effect on the capsule size. Suggestions for future research are to use pin bar in manufacturing process to get more uniform results and further research is needed to modify the method used to make capsule shells using only modified *E. guineensis* starch.

SUPPLEMENTARY INFORMATION

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CONFLICT OF INTEREST

There is no conflict of interest in this article.

AUTHOR CONTRIBUTION

Amalia Khairunnisa (AK) : Characterization (tensile strength, FTIR and SEM) ; Nani Kartinah (NK) :Formulation, decolorization ; Pratia Viogenta (PV): Preparation sample, extraction ; Dessy Aulia (DA) : modification palm starch

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