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Addition of *Anadara granosa* shell chitosan in production bioplastics

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Abstract. *Bioplastic is a plastic composite material that can quickly degrade. The aims of the research are; to examine the optimal composition of bioplastics made from tapioca waste by adding blood clam shell chitosan and glycerol, to determine the quality of production bioplastics from tapioca waste with the addition of blood clam shell chitosan and glycerol, and to determine the chemical content of the clam shells. Raw material production of bioplastic was waste tapioca flour (%), chitosan (%), and glycerol (mL) of 65:35:5, 70:30:5, and 75:25:5. The sample was placed in a water bath and stirred at 80 ^{o}C for 15 minutes, then the bioplastics were pressed on aluminum foil. The bioplastics dried at 100 ^{o}C for 90 minutes and cooled at room temperature for 6 hours. The results of the tensile strength on PBA1, PAB2, PAB3 samples were 0.75 Mpa, 0.54 Mpa, and 0.34 Mpa, respectively. Elongation at break test value the PBA1 sample is 23.68%, the PBA2 sample is 15.33%, and the PBA3 sample is 12.12%. The test results of the sample do not meet the value of the quality standard SNI 7188.7:2016 for the bioplastic category. The optimal composition of bioplastic can be found in PBA2 sample using tapioca flour as raw material with chitosan and glycerol as much as 70%:30%:5mL with a tensile strength 0.54%, an elongation at break 15.33%, biodegradation of 43%, and has a bioplastic content with functional groups (C=C), (CO), (CH),(OH), (C=C), and $(CH_2)_n$ in the FTIR test results.*

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INTRODUCTION

The industries in Indonesia are developing quite modern. It is followed by population growth and the high interest of the community in meeting their daily needs using plastic. Along with the advancement of online-based technology, packing services using plastic materials that are cheap, lightweight, durable, and chemical resistant are growing rapidly (Agustin and Padmawijaya 2016). Based on worldwide data, more than 300 million tons of plastic were consumed in 2015, which resulted in 34 million tons of plastic waste worldwide, and 93% was thrown away on land and oceans (Arikan and Bilgen 2019). These plastic materials are generally not processed because it becomes the accumulation of plastic waste due to their high resistance to corrosion, water, and bacterial decomposition, so plastic waste is difficult to remove and will become a serious problem (Jabeen et al. 2015). Plastic waste that has been fragmented can still cause water contamination because it

undergoes many changes in form into particles known as microplastics, so they are easily swallowed by organisms (Khotimah et al. 2022). Currently, the circulating plastics, including synthetic polymers, are not easy to decompose in a short time. It tends to cause accumulation of waste and environmental damage problems such as flooding (Maneking et al. 2020), the release of toxic dioxin gas, a breeding ground for disease vectors, to the death of marine life. Plastic packaging contributed 57% of the total plastic waste generated in 2018 and accounted for most of the city's waste. In plastic waste generated in 2018, 20% was recycled, 29% of plastic waste was disposed of, and 31% of waste was disposed of improperly to leak (Shrestha et al. 2020).

On the other hand, Indonesia is a country with abundant marine life, such as shellfish. Blood clam shells are often found in waste from shellfish processing in restaurants that provide food made from marine life and around the Kenjeran beach. Meanwhile, the utilization of blood clam shells has not been optimal, what has been done is as a material for various crafts (Maneking et al. 2020). Blood clam shells contain chemical compounds such as calcium carbonate, chitin, calcium hydroxyapatite, and calcium phosphate. Chitin derived from blood clam shells can be processed into chitosan. Chitin content is found in various types of marine life, such as crustaceans (crabs, lobsters, shrimps) and mollusks (mussels, clam shells, squid, krill) (Fajri and Amri 2018). Chitosan is a polymer formed from glucosamine monomers with -(1-4) bonds. Chitosan can be formed when the acetyl group in chitin is substituted by hydrogen to become an amine group (Masindi and Herdyastuti 2017). Nur et al. (2019) stated that some chitin content could be found in marine biota that has shells like mollusk, such as the blood clams (*Anadara granosa*).

Research conducted by (Muhammad et al. 2020) stated that with the addition of 2.5 grams of chitosan in the test sample has a tensile strength value of 4 Mpa. This shows that the more chitosan composition, the more hydrogen bonds are contained in the bioplastic so that it will be stronger and more difficult to break. Other research that has been conducted (Kelibay 2020) showed that the concentration of glycerol 30 mL has a significant effect on the rate of bioplastic degradation so that it can be completely decomposed for 18 days by the method of burial in soil media. Meanwhile, according to research by (Selpiana et al. 2016), the results were obtained with the addition of 5 grams of chitosan with a tensile strength of 132,175 Kgf/cm^{2,} while the addition of 3 mL of glycerol increased the optimum elongation of 11.95%.

Glycerol is an organic compound with the chemical formula $C_3H_8O_3$ which has biodegradable, hygroscopic, non-toxic, odorless, colorless (transparent) properties. Usually, glycerol is produced from various processes such as transesterification, saponification, fatty acid hydrolysis, and microbial fermentation (Panjaitan 2021). In the study by Septiosari et al. (2014), glycerol has an important role in the manufacture of bioplastics to produce hydrophobic plastics, increase elasticity, reduce water absorption, and decompose in nature. Tapioca flour is the result of cassava root extract, tapioca starch is used as raw material for the production of bioplastics, tapioca flour amylose content is the quite high range of 20 – 27% and amylopectin 83% (Indrianti et al. 2013). Starch is classified as a type of polysaccharide that is easily biodegradable, relatively cheap, and easy to obtain (Hidayat et al. 2020). According to (Astuti et al. 2019), the sample with the highest composition of cassava pulp flour experienced the fastest degradation. The aims of the research are; to research the optimal composition of bioplastic made from tapioca waste by adding blood clam shell chitosan and glycerol, to determine the quality of production bioplastics from tapioca waste with the addition of blood clam shell chitosan and glycerol, and to determine the chemical content of the clam shells. Bioplastic function of tapioca with the addition of blood clam shell chitosan and glycerol.

MATERIALS AND METHODS

Materials

The raw material of the research was blood clam shell waste obtained from the Kenjeran Beach area in the form of chitosan powder that had passed a 200 mesh sieve, tapioca waste, glycerol, 96% ethanol, 2% acetic acid, and equates. The tools used in this research were; a 20 cm tin mold, digital scale, electric stove, 250 mL measuring cup, and stirrer.

The Proses of Chitosan Production

The process of production of bioplastics is carried out in several stages, such as the production of chitosan from blood collar shells. The process of production of chitosan is as follows (Udyani 2017): 1) Separation of protein (deproteination), namely sifted shellfish powder (200 mesh) was put into a glass beaker and added 3.5% NaOH solution with a ratio between shellfish powder and NaOH solution of 1:10 (w/v). This stage lasts for 2 hours at a temperature of 65 °C with stirring. Furthermore, the solution was filtered with filter paper to obtain the residue. Then wash the residue with distilled water until the pH is neutral. The neutral residue was then oven-dried at 100 °C until dry. The result of drying is called Crude Chitin; 2) Separation of minerals (Demineralization) was carried out by inserting crude chitin into a glass beaker and adding 1 N HCl solution with a ratio between crude chitin and HCl solution of 1:10 (w/v).

This step was carried out using stirring for 30 minutes at room temperature. The results obtained were filtered with filter paper to obtain a residue. Next, wash the residue with distilled water until the pH is neutral, then oven at 100 °C until dry. The result obtained is called chitin powder; 3) Deacetylation (transformation of chitin into chitosan) is carried out by changing chitin into chitosan. Chitin powder was put into a three-neck flask, then 50% NaOH solution was added with the ratio between chitin powder and NaOH solution 1:10 (w/v). The solution was then refluxed at 100 °C for 5 hours with stirring. The results obtained were then filtered with filter paper to obtain the residue. The residue was washed with distilled water until the pH was neutral. The neutral residue was then oven-dried at 100 °C until dry.

Experimental Design

The research was conducted experimentally at the Environmental Laboratory, Department of Environmental Engineering, Faculty of Engineering, Universitas PGRI Adi Buana Surabaya. This study consisted of 3 variables with the concentration of glycerol in each variable of 5 mL, ethanol 96% of 20 mL, acetic acid 2% of 30 mL, and distilled water of 30 mL. Comparison of the composition of the manufacture of bioplastics can be seen in Table 1.

Table 1 Comparison of bioplastic production composition

Sample code	Variation
PBA1	Tapioca waste:blood clam shell chitosan of 65%:35%
PBA2	Tapioca waste:blood clam shell chitosan of 70%:30%
PBA3	Tapioca waste:blood clam shell chitosan of 75%:25%
PBK	Tapioca waste, without blood clam shell chitosan

The next step was the production of bioplastic with tapioca waste composite with the addition of blood collar shell chitosan and glycerol into a basin according to the ratio of 65%:25%, 70%:30%, and 75%:25%. Each variable was added with 30 mL of distilled water and 20 mL of 96% ethanol and then in a mixer until well mixed. The polymerization process began by heating the bioplastic composite at a temperature of 80 °C and stirring for 10 minutes until a transparent biopolymer was formed. The homogeneous solution was then molded into a 20 cm baking pan. Dry (oven) the printed solution at a temperature of 100 °C for 2 hours and were cooled at room temperature for 6 hours.

The quality of the bioplastic would be tested using the tensile strength parameter (tensile strength) using the American Standard Testing and Material (ASTM) method or can be tested in a laboratory that has implemented ISO/IEC 17025. The standard value for tensile strength (tensile strength) was at least 13.7 MPa (139.74 N/mm^2), elongation at break using the ASTM method or can be tested in a laboratory that has implemented ISO/IEC 17025. The maximum quality standard value was 5%. FTIR used infrared wavelengths to determine chemical compounds contained in bioplastics provided that thermoplastics must meet the requirements for the heavy metal content of Cd < 0.5 ppm, Pb < 50 ppm, Hg < 0.5 ppm, Cr^{6+} < 50 ppm,

bioplastic and or a mixture of azo dyes and biodegradation of at least 30% according to SNI 7188.7:2016 category of easily biodegradable bioplastics.

RESULTS AND DISCUSSION

Tensile Strength and Elongation at Break Test Results in Bioplastics

From Table 2 it can be seen that the addition of blood clam shell chitosan affected the results of the tensile strength test on bioplastics consistently, where the addition of 35% chitosan in the PBA1 sample produced a value of 0.75 Mpa. While the PBA2 and PBA3 samples with the addition of 30% chitosan and 25% decreased with a value of 0.54 MPa and 0.34 MPa. The results of the calculation of the tensile strength test in the bioplastic process are presented in Figure 1.

Table 2 Test data result tensile strenght dan elongation at break

Sample code	Tensile strenght test	Elongation at break test
PBA1	0.75 Mpa	23.68%
PBA2	0.54 Mpa	15.33%
PBA3	0.34 Mpa	12.12%
Quality standard value*	minimum 13.7 Mpa	maximum 5%

Source: *SNI 7188.7:2016

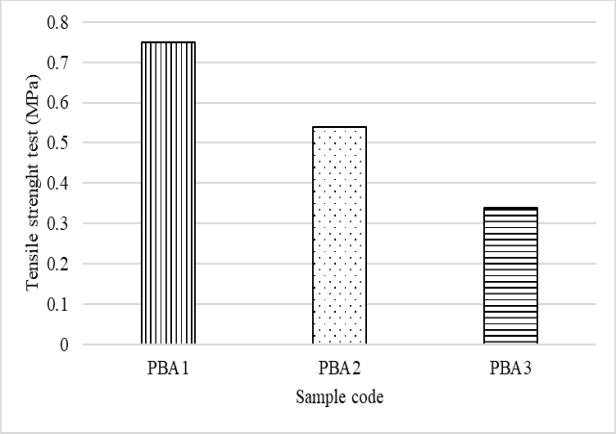

Figure 1 Tensile strength test results

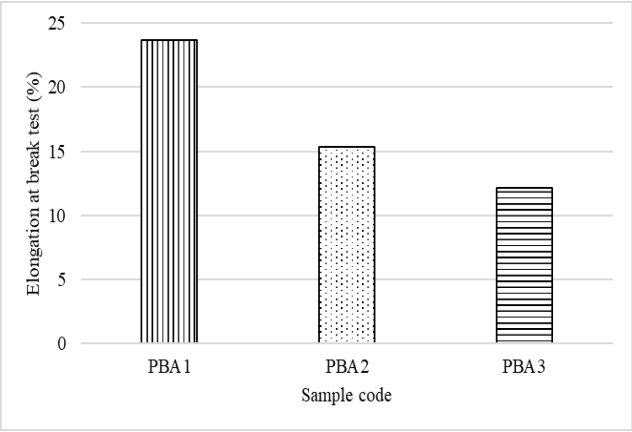

Figure 2 Elongation at break test results

The results of the calculation of the tensile strength test in the bioplastic process are presented in Figure 2. Based on the results of the analysis of the tensile strength test data and elongation at break, it is known that the use of organic raw materials, such as tapioca flour and blood collar shell chitosan as well as the addition of glycerol, have an effect on the tensile strength test results and elongation test results at the break on bioplastic samples. The results of the tensile strength test analysis that were closest to the minimum value of 13.7 MPa according to SNI 7188.7: 2016 are PBA1 samples with a value of 0.75 MPa with tapioca flour and blood clam shell chitosan as raw materials as much as 65%:35%. The research conducted by (Melani et al. 2017) explained that using 4% clay (chitosan) filler added with 25% glycerol by weight of starch (2.5 g) obtained a tensile strength value of 76.575962 MPa. While in this research, the process of only 5% glycerol with chitosan 35%, 30%, and 25% of the total weight of 20 g was used. Thus, tensile strength test results in this research does not meet SNI 7188.7:2016 quality standards. This is following the research (Hartatik et al. 2014) that the more levels of chitosan added, the lower the tensile strength value of bioplastics so that they were prone to breaking.

Elongation at Break

In the results of the analysis of the elongation test at the break, the lowest value was taken, namely the PBA3 sample with a value of 12.12% with a comparison of the composition of tapioca flour, chitosan, and glycerol, namely 75%:25%:5 mL because the value of the bioplastic quality standard SNI 7188.7:2016 was 5%. Based on Kurniawati et al. (2022) the addition of 25% glycerol has a tensile strength value that almost meets the standard of SNI 7188.7:2016, which is 7.12 Mpa. Based on the research by (Nafiyanto 2019), there is a hydrogen interaction between chitosan-glycerol-starch, where the addition of glycerol can produce a more elastic bioplastic film so that the elongation at break value increases but the tensile strength decreases. Glycerol which functions as a plasticizer, was located between the biopolymer chains. Thus, the distance between chitosan and starch increased. This made the hydrogen bonds between chitosan-starch reduced and replaced by hydrogen interactions between chitosan-glycerol and glycerol-starch. Therefore, the bioplastic will be more elastic so that elongation tends to increase even if it is pulled with small pressure. The composition of the bioplastic manufacture from this study that was optimally close to the minimum value of tensile strength of 13.7 MPa and elongation at break of 5% was PBA2 samples of 0.54 MPa and 15.33% with a ratio of tapioca flour and chitosan of 70%:30%.

Data Analysis and Discussion of Biodegradation Test Results on Bioplastics

From the biodegradation test process that has been carried out at the Environmental Engineering Laboratory of PGRI Adi Buana Surabaya, the results of bioplastic testing were obtained. From Table 3 and Table 4 the results of the biodegradation test were quite significant. the PBA1 sample obtained a value of 55%. There was a decrease in the PBA2 sample with a value of 43%, but there was a fairly high increase in the PBA3 sample of 72%. The results of the biodegradation test on bioplastics are presented in Figure 3.

Table 3 Biodegradation data results in bioplastics

	PBA1	**PBA2**	**PBA3**	**PBK**
Initial weight	0.99 g	0.66 g	0.80 g	0.87 g
Final weight	0.44 g	0.37 g	0.22 g	0.79g

Source: The results of the biodegradation test refer to SNI 7188.7:2016

Table 4 Calculation results of biodegradation test on bioplastics

Biodegradation	
PBA1	55%
PBA2	43%
PBA3	72%
PBK	10%
Quality standard value	*minimal 30%

Source: Calculation results refer to SNI 7188.7:2016

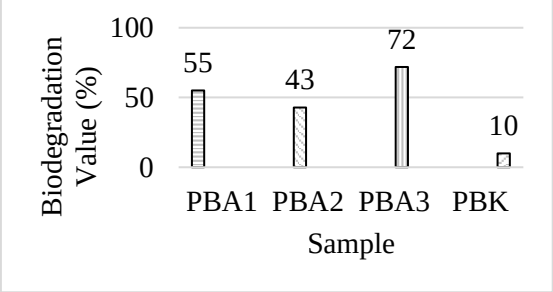

Figure 3 Biodegradation test results

The results of the biodegradation analysis on bioplastics stated that the PBA1 sample with the composition of tapioca flour added with chitosan and glycerol was 65%:35%:5 mL. PBA2 sample with the composition of tapioca flour added with chitosan and glycerol was 70%:30%:5 mL. PBA3 samples with the composition of tapioca flour added with chitosan and glycerol were 75%:25%:5 mL. They could be degraded well for 7 days in nature using soil media with the value of each sample that meets the bioplastic quality standard SNI 7188.7:2016 with a minimum value of 30%. While the CPB sample as a control variable with the composition of tapioca flour without the addition of chitosan and glycerol showed a biodegradation value of 10%. This was in line with research (Hasanah and Mahyudin 2022) that it is necessary to add a plasticizer such as glycerol because it has hygroscopic properties that can absorb water easily, and decomposes easily in nature. The best results of the biodegradation test from this research were found in the PBA3 sample with the composition of tapioca flour added with chitosan and glycerol as much as 75%:25%:5 mL which had a high-water absorption ability because, in only 7 days, it obtained 72% yield. The value of biodegradation of bioplastics from banana peels was in the range of 46.00 – 59.40% in 8 days. The highest biodegradation using soil media in 2 days with a value of 50% (Widyastuti et al. 2021).

Based on Widiatmono et al. (2021), Increasing levels of chitosan will affect the growth of microorganisms that function to break down bioplastics. Thus, the acceleration of degradation in bioplastics will slow down. This statement was in line with the results of the biodegradation test on the PBA1 sample with the composition of tapioca flour added with chitosan and glycerol of 65%:35%:5 mL. It had an absorption capacity of 55%. Meanwhile, for the PBA2 sample with the composition of tapioca flour added with chitosan and glycerol of 70%:30%:5 mL, the biodegradation test results were 43%. It can be assumed that the influence of heavy rain that falls directly on the soil where the bioplastic is buried will affect the percentage of biodegradability.

Data Analysis and Discussion of FTIR Test Results

The FTIR test process that has been carried out in the pharmaceutical laboratory of Universitas Airlangga is shown in Figures 4, 5, 6, and 7. The results of bioplastic testing that referred to the quality standard value of SNI 7188.7:2016 for the bioplastic category are presented in Table 5.

The results of this FTIR test were in the form of peak data that showed the chemical compounds for the production of bioplastics in a certain absorption range. The PBA1 with the composition included tapioca flour with the addition of chitosan and glycerol of 65%:35%:5 mL showed a wave absorption between 699.52 – 3,737.62. It indicated the presence of Alkene compounds (C=C) which are hydrocarbon groups that have double bonds. belonging to unsaturated hydrocarbons, carboxyl (C-O) is an organic compound derived from alkanes with the functional group COOH and the general formula CnH_2nO_2, simple aromatic compounds, esters, alkanes (C-H) are part of aliphatic hydrocarbon compounds which were classified as saturated hydrocarbon compounds where all bonds in single carbon atom, alkane can dissolve easily in organic solvents, and a hydroxyl group (O-H). For samples, PBA2 and PBA3 with the composition consisting of tapioca flour with the addition of chitosan and glycerol, were 70%:30%:5 mL and 75%:25%:5 mL. The wave absorption was between 643.92 – 3,898.63 and 644.35 – 3,951.22. It showed the presence of different compounds from the PBA1 sample, namely Hydrocarbon compounds $(CH2)_n$. While the CPB sample with 20 g tapioca flour composition as the control variable showed the least wave absorption, namely 648.77 – 3,305.47, the compounds contained were the same as PBA1, PBA2, and PBA3 samples.

Based on the research by (Harsojuwono et al. 2016), Bioplastics made from starch polymers were estimated to contain hydroxyl functional groups (O-H) bonded to hydrogen, alkanes (C-H), aldehydes (C-H), carboxylic acid hydrogen bonds, alkynes (C≡C), esters, simple aromatic compounds, carboxyl (C-O), alkenes (C=C) and Hydrocarbons $(CH_2)_n$. Based on research by (Widiatmono et al. 2021), the chemical bonding group between chitosan and glycerol is characterized by the presence of O-H groups from glycerol and N-H groups from chitosan. Yet, in this research, the results obtained were known that the bioplastic solution was not completely homogeneous. This was indicated by the absence of visible N-H functional groups. The results of

the FTIR test in the manufacture of bioplastics in this research had met the standard values for bioplastics. In samples PBA1 to PBA3 with compositions containing tapioca flour and glycerol with added chitosan of 35%, 30%, and 25% containing functional groups (C=C), (C-O), (C-H),(O-H), (C≡C), and $(CH_2)_n$. and no indication of heavy metal content, Cd < 0.5 ppm, Pb < 50 ppm, Hg < 0.5 ppm, Cr^{6+} < 50 ppm, and did not contain azo dyes.

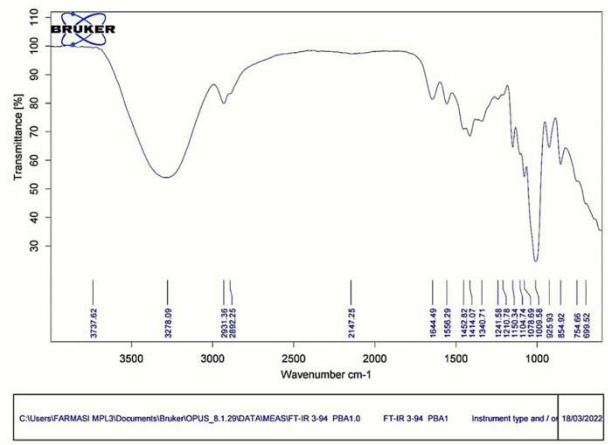

Figure 4 FTIR in PBA1

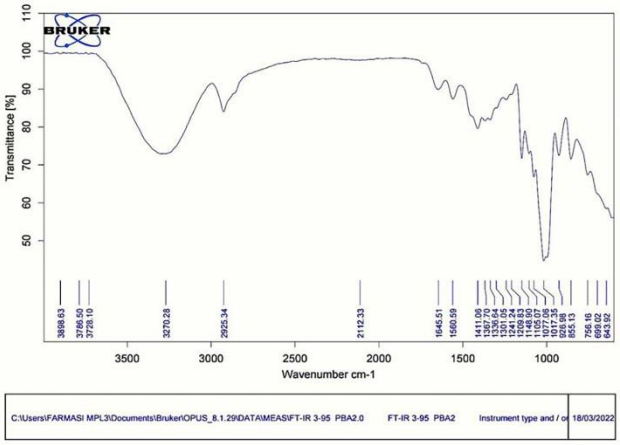

Figure 5 FTIR in PBA2

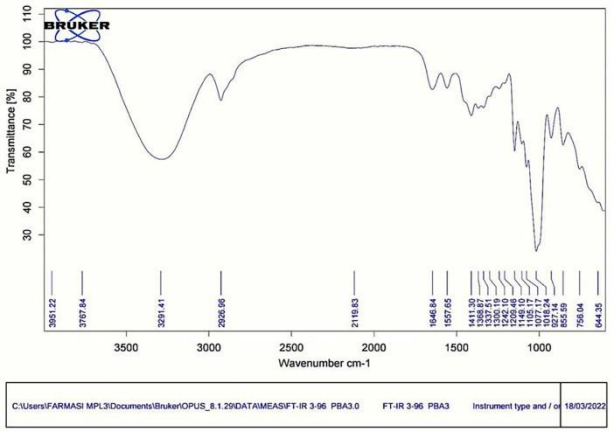

Figure 6 FTIR in PBA3

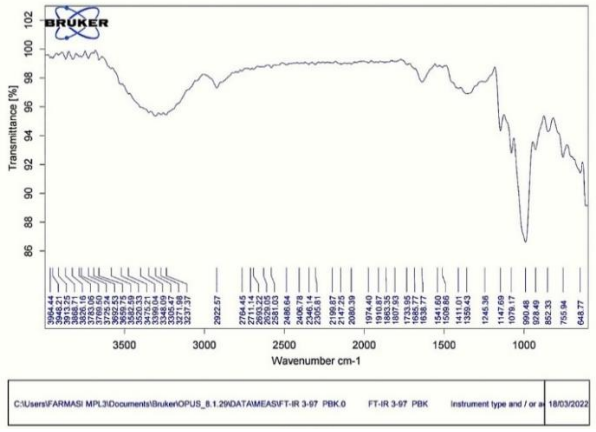

Figure 7 FTIR in PBK

Table 5 FTIR test results on bioplastics

Sample	Peak	Compound	Sample	Peak	Compound
PAB1	699.52	Alkena (C=C)	PBA2	643.92	Hidrokarbon $(CH_2)_n$
	754.66	Alkena (C=C)		699.02	Alkena (C=C)
	854.92	Alkena (C=C)		756.16	Alkena (C=C)
	925.93	Alkena (C=C)		855.13	Alkena (C=C)
	1,009.58	Carboxyl (C-O)		926.98	Alkena (C=C)
	1,078.69	Carboxyl (C-O)		1,017.35	Carboxyl (C-O)
	1,104.74	Carboxyl (C-O)		1,077.06	Carboxyl (C-O)
	1,150.34	Carboxyl (C-O)		1,105.07	Carboxyl (C-O)
	1,210.78	Carboxyl (C-O)		1,148.90	Carboxyl (C-O)
	1,241.58	Carboxyl (C-O)		1,209.83	Carboxyl (C-O)
	1,340.71	Simple aromatic compounds		1,241.24	Carboxyl (C-O)
	1,414.07	Simple aromatic compounds		1,301.05	Simple aromatic compounds

Sample	Peak	Compound	Sample	Peak	Compound
	1,452.82	Simple aromatic compounds		1,336.64	Simple aromatic compounds
	1,556.29	Simple aromatic compounds		1,367.70	Simple aromatic compounds
	1,644.49	Simple aromatic compounds		1,411.06	Simple aromatic compounds
	2,147.25	Ester		1,560.59	Simple aromatic compounds
	2,829.25	Aldehida (C-H)		1,645.51	Simple aromatic compounds
	2,931.36	Alkana (C-H)		2,112.33	Alkuna (C=C)
	3,278.09	Gugus hydroxyl (O-H)		2,925.34	Alkana (C-H)
	3,737.62	Gugus hydroxyl (O-H)		3,270.28	Gugus hydroxyl (O-H)
				3,728.10	Gugus hydroxyl (O-H)
				3,786.50	Gugus hydroxyl (O-H)
				3,898.63	Gugus hydroxyl (O-H)
PAB3	644.35	Hidrokarbon $(CH_2)_n$	PBK	648.77	Hidrokarbon $(CH_2)_n$
	756.04	Alkena (C=C)		755.94	Alkena (C=C)
	855.59	Alkena (C=C)		990.48	Alkena (C=C)
	927.14	Alkena (C=C)		1,359.43	Simple aromatic compounds
	1,018.24	Carboxyl (C-O)		1,638.77	Simple aromatic compounds
	1,077.17	Carboxyl (C-O)		2,922.57	Alkana (C-H)
	1,105.17	Carboxyl (C-O)		3,305.47	Gugus hydroxyl (O-H)
	1,149.10	Carboxyl (C-O)			
	1,209.46	Carboxyl (C-O)			
	1,242.10	Carboxyl (C-O)			
	1,300.19	Carboxyl (C-O)			
	1,337.51	Simple aromatic compounds			
	1,368.87	Simple aromatic compounds			
	1,411.30	Simple aromatic compounds			
	1,557.65	Simple aromatic compounds			
	1,646.84	Simple aromatic compounds			
	2,119.83	Ester			
	2,926.96	Alkana (C-H)			
	3,291.41	Gugus hydroxyl (O-H)			
	3,767.84	Gugus hydroxyl (O-H)			
	3,951.22	Gugus hydroxyl (O-H)			

Product Bioplastic

Production of bioplastics from *Anadara granosa* shell chitosan are shown in Figure 8, 9, 10, and 11. The bioplastic produced was in the form of sheets. The bioplastic in samples PBA1, PBA2, and PBA3 had was smooth surface and there were small bubbles. The bioplastic in samples PBK had was rough surface, slighty stiff, and there were small bubbles.

Figure 8 Bioplastic product in PBA1

Figure 9 Bioplastic product in PBA2

Figure 10 Bioplastic product in PBA3

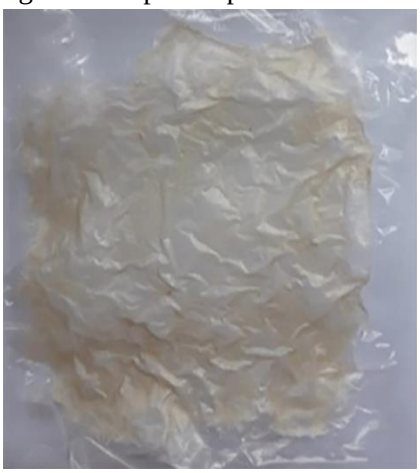

Figure 11 Bioplastic product in PBK

CONCLUSION

From the results of research and discussion, it can be concluded that in PBA2 sample is the optimal composition of bioplastics using tapioca flour as raw material with added chitosan and glycerol of 70%:30%:5 mL with a biodegradation test value of 43%. A bioplastic derived from tapioca flour with the addition of chitosan and glycerol has a tensile strength test value of 0.75 Mpa PBA1 sample, 0.54 Mpa PBA2 sample, and 0.34 Mpa PBA3 sample. The elongation at the break test value of the PBA1 sample is 23.68%. The PBA2 sample is 15.33%, and the PBA3 sample is 12.12%. The test results of these samples did not meet the quality criteria of SNI 7188.7:2016 for the bioplastic category. The chemical content of bioplastics from tapioca flour with the addition of chitosan and glycerol has functional groups (C=C), (C-O), (C-H), (O-H), (C≡C), and $(CH_2)n$ in the FTIR test results.

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