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PREPARATION OF 70% ETHANOL EXTRACT OF PUMPKIN NANOPARTICLES (SECHIA EDULLE) USING THE INONIC GELATION METHOD IN LOZENGES TABLET FORMULATION

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ABSTRACT

Introduction:*Chayote is a plant that is efficacious in lowering blood pressure. High blood pressure in hypertension can cause increased morbidity and mortality. These bad effects can be avoided by taking medication regularly. The aim of taking medication regularly is to ensure that drug levels in the blood remain within the therapeutic range. Apart from that, to achieve maximum drug levels in the blood is to make the drug into nanoparticles.* ***Objectives:*** *The aim of this research is to prepare nanoparticles of 70% ethanol extract of Chayote using the ionic gelation method.* ***Methods:****The research method is laboratory experimental. The research data is in the form of quantitative data, namely the size of the nanoparticles.* ***Results:*** *Nanoparticle preparation was carried out using a solution of 1% Chitosan and 0.5% Na TPP with a ratio of 2:1. The results of nanoparticle preparation were tested using Particle Size Analysis. The yield weight of the extract was also calculated at 27.73%. The measurement results of 70% Chayote ethanol extract nanoparticles were 556,5nm.* ***Conclusions:*** *the particle size meets the criteria for nanoparticles of 10-1000nm (556,5nm).*

Introduction

Chayote (*Sechium edule*) is a plant with a lot of nutritional content. This plant has been pre-clinically and clinically proven to lower blood pressure (Munawassalmiah et al., 2018; Pratiwi, 2018). The flavonoid content in chayote (*Sechium edule*) is thought to have activity in lowering blood pressure (Anas et al., 2021). The compounds phloridzin, naringenin, floretin, and epigenin are compounds that have anti-inflammatory and antioxidant (Rosado-Perez et al., 2019).

Uncontrolled blood pressure in hypertension patients can trigger complications and be the main cause of death. There is an increase in the number of hypertensive patients every year. The number of hypertension patients aged 30-79 years is 1.28 billion people. 2/3 of hypertension sufferers live in low and middle income countries (WHO, 2023). Data from Basic Health Research (Riskesdas) in 2018 stated that the number of hypertension sufferers in Indonesia was 34.1%.

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South Kalimantan occupies the highest position at 44.1% and the lowest in Papua at 22.2% (Indonesian Ministry of Health, 2018).

Effective hypertension therapy through blood pressure can prevent blood vessel damage, can reduce the incidence of morbidity and mortality (Dipiro et al, 2023). Research by Pratiwi (2018) on the antihypertensive effects of Swartz Chayote (*Sechium edule*) ethanol extract. in hypertensive rats induced by monosodium glutamate. Research result showed that chayote ethanol extract 132 mg/kg BW was able to reduce blood pressure (systolic and diastolic) in hypertensive rats. Another research by Munawassalmiah et al. (2018) regarding clinical observations of Chayote (*Sechium edule*) extract as an antihypertensive. The results of the study showed that Chayote (*Sechium edule*) Chayote extract dosage 100 mL/day for 7 days produced a significant difference in systolic (p=0.001) and diastolic (p=0.009).

Compliance with taking medication is one of the things that determines the success of hypertension therapy. Patients with hypertension will take medication continuously to be able to maintain drug levels within the therapeutic range (Dipiro et al., 2023). The use of drugs with natural ingredients also has other problems, namely low solubility and bioavailability. To increase solubility and bioavailability, efforts are made to size the active ingredients in the form of nanoparticles using the ionic gelation method (Hanutami and Budiman, 2014). The ionic gelation method uses a combination of polycations (eg chitosan) and polyanions (eg tripolyphosphate). The combination of these polymers produces nanoparticles with good stability. Research by Pakki et al. (2016) regarding the formulation of Dayak onion (Eleutherine americana (Aubl) Merr) extract nanoparticles with varying concentrations of chitosan-tripolyphosphate (TPP). 0.5% Chitosan Solution; 0.75%; and 1% combined with 1% TPP solution to produce nanoparticles with a size of 200-500nm.

Based on the above background, researchers want to prepare nanoparticles of 70% ethanol extract of Chayote (*Sechium edule*). The extraction method for Chayote (*Sechium edule*) uses maceration. The goal is that the flavonoid content is not damaged during the extraction process. The ionic gelation method was chosen using a ratio of 1% Chitosan solution and 0.5% Na-TPP with a concentration and volume ratio of 2:1.

Methods

Time and Place of Research : The time for research will be carried out from March to July 2024. The location for the research will be the Natural Materials Laboratory and the STIKES Banyuwangi Technology Laboratory. Determination of the particle size of Chayote (*Sechium edule*.) was carried out at Integrated Services Laboratory, Institute of Agricultural Technology, Brawijaya University.

Tools and Materials : RE-2000E rotary evaporator, oven (B-ONE), analytical balance (Mettler Toledo), hotplate magnetic stirrer (BioBase), magnetic stirrer (Monotaro), Particle Size Analyzer (PSA) (Mastersizer MS3000), sieve mesh no. 40 (Retsch), blender (Cosmos), mortar and stamper, glassware (Pyrex®), and knives.

The materials used include Chayote (*Sechium edule*) purchased directly from farmers in Curah Leduk, Banyuanyar Village, Kalibaru District, Banyuwangi Regency; PVP K-30 (Sigma Aldrich); Chitosan (Sigma Aldrich); Sodium Tripolyphosphate (Brataco); Tween 80 (Brataco); Aquadest (Sigma Aldrich); and Ethanol 70% (Brataco

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Research Procedures :

Chayote Extraction : Selection of Chayote (*Sechium edule*) is an old fruit with a harvest time of 3 months 21 days. Siamese pumpkin (*Sechium edule*) weighing 15 kg, washed under running water and cut into 6-7 mm thick pieces without removing the skin. Then, it is dried in an oven at 55ºC until the water content is <10%. Next process is blended and sieved with mesh sieve no. 40. Maceration using 70% Ethanol in 750 g of dried simplicia for 3 days, stirring occasionally. Maceration was carried out 2 times. The maserate is mixed and left for 1 night. Solvent evaporation using a rotary evaporator at a temperature of 60ºC.

Process for Making Chayote (*Sechium edule*) Extract Nanoparticles : 1% Chitosan solution (80 mL) was added with Tween 80 (1 mL) and stirred using a magnetic stirrer at a speed of 1000 rpm for 10 minutes. Then 1000 mg of Chayote (*Sechium edule*) extract was added, stirred with a magnetic stirrer at 1000 rpm for 30 minutes. 40 mL of Na TPP 0.5% was added and stirred with a magnetic stirrer at 1250 rpm for 90 minutes. The mixture was left for 1x24 hours.

Data analysis : The research data is in the form of quantitative data which includes yield weight and nanoparticle size. The data is presented in table form as below:

Table 1 Data Analysis and Conclusion of Research Results

No.	Procedure	**Data** Analysis	Conclusion
1.	Making Chayote Extract	• Maceration • Extract yield = $\frac{weight\ of\ extract}{weight\ of\ simplisia}\ x\ 100\%$	Extract with a yield weight of >10%.
2.	Preparation of Chayote Extract Nanoparticles	*Particle Size Analysis* (PSA)	Particle size 10-1000nm

Results

The research results can be seen in the table below:

Table 2 Results of Maceration and Chayote Extract Nanoparticles

No.	Procedure	**Data** Analysis	Conclusion
1.	Making Chayote Extract	• Maceration • Extract yield = $\frac{750\ grams}{208{,}01\ grams}\ x\ 100\% = 27{,}73\%$	Extract with a yield weight of >10%.
2.	Preparation of Chayote Extract Nanoparticles	*Particle Size Analysis* (PSA)	Particles size 556,5nm (10-1000nm)

Discussion

The research began by making 70% ethanol extract of Chayote (*Sechium edule*) using the maceration method. 15 kg of Siamese pumpkin (*Sechium edule*) was sorted and washed with running water. Chopping is done to reduce the size so as to speed up drying. The results of drying Siam Pumpkin (*Sechium edule*) were obtained in the amount of 750

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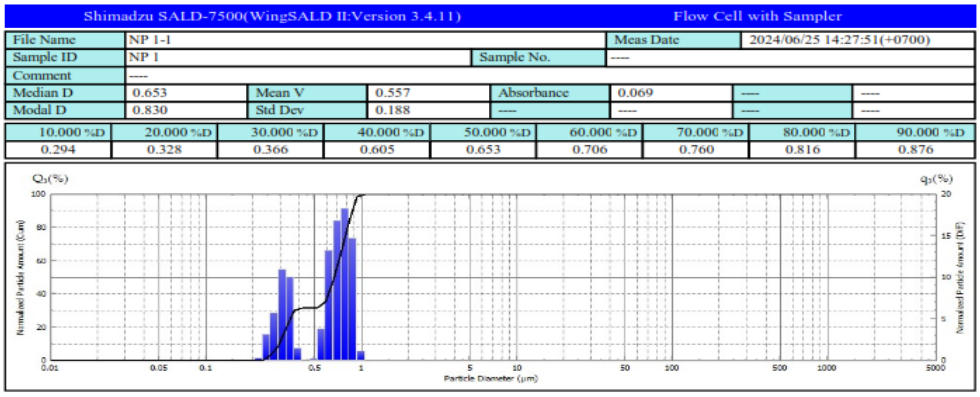
grams, blended, and sieved with No. 40 mesh sieve. Things that are done after harvesting, for example sorting, washing, chopping and drying, aim to ensure that the active compound content is guaranteed and the plant material is not damaged (Widiyastuti, 2020). This research produced dry simplicia with a weight reduction of 95% of the weight of wet simplicia.

The next process was carried out by maceration and a thick extract of 208.01 grams was obtained. The yield calculation results are known to be 27.73%. Based on the Herbal Pharmacopoeia II edition, the yield weight requirement is not less than 10%. The results of this research are in accordance with the yield weight requirements listed in the Herbal Pharmacopoeia edition II (Ministry of Health of the Republic of Indonesia, 2017). The high yield weight in this research indicates that the extract produced is greater. The results can be seen in the image below:

Figure 1. Condensed Chayote Extract (*Sechium edule*)

The extract that has been obtained is then made into nanoparticles. The results of nanoparticle measurements can be seen in the image below:



Shimadzu SALD-7500(WingSALD II:Version 3.4.11)					Flow Cell with Sampler			
File Name	NP 1-1				Meas Date	2024/06/25 14:27:51(+0700)		
Sample ID	NP 1			Sample No.	----			
Comment	----							
Median D	0.653	Mean V	0.557	Absorbance	0.069	----	----	
Modal D	0.830	Std Dev	0.188	----	----	----	----	

10.000 %D	20.000 %D	30.000 %D	40.000 %D	50.000 %D	60.000 %D	70.000 %D	80.000 %D	90.000 %D
0.294	0.328	0.366	0.605	0.653	0.706	0.760	0.816	0.876

Figure 3. Nanoparticle Measurement Results

Nanoparticle measurements were carried out using a Particle Size Analyzer (PSA). The nanoparticle size obtained was 0.5565μm (556.5nm). The resulting particles meet the criteria for nanoparticles, namely 10-1000nm (Tiyaboonchai, 2003; Aloys et al., 2016). The ionic gelation method used in this research with a ratio of 1% Chitosan and 0.5% Na-TPP can produce particle sizes that meet the standart. The measurement results can be seen in table 2 as follows:

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Table 2. Particle Diameter Measurement Results

Sample Code	Median (µm)	Mean (µm)	Modul (µm)	Absorbansi	Std Dev
NP1	0,652	0,556	0,830	0,070	0,188
	0,653	0,557	0,830	0,069	0,188

The formation of nanoparticles is influenced by the ratio of extract, chitosan, and NaTPP; comparison of chitosan and NaTPP; amount of chitosan and extract; as well as the pH of chitosan (Taurina et al, 2013; Choiri et al, 2016). The amount of Chayote (*Sechium edule*) extract used in making nanoparticles was 1 gram with 80 mL of 1% chitosan and 40 mL of 0.5% NaTPP. A large amount of extract will cause the particle size to become large. This is the same as research by Prihantini et al. (2021). The 300 mg extract has a particle size of 612.8nm compared to the 100 mg extract with a particle size of 444.5nm.

The ratio of chitosan and NaTPP used in this research is 2:1. Another study using a similar comparison produced a nanoparticle size of 419.18 nm (Pakki et al., 2016). Research by Fitri et al. (2021) used a chitosan-NaTPP ratio of 2:1 with a smaller volume to produce a particle size of 284.1 nm. This difference is caused by the volume comparison of chitosan-NaTPP and the preparation. To obtain a small nanoparticle size, the chitosan concentration can be reduced.

Samples that have been prepared using the ionic gelation method are then measured for their particles. However, the samples were not immediately tested because they were waiting in line. During storage, samples were placed at room temperature. Storage can affect the size of the nanoparticles. This is similar to research conducted by Handani et al. (2016). The results of this research showed that 0.6% chitosan stored for 30 days increased from 600nm to 700nm. During the storage process, the chitosan solution can experience growth. The growth of chitosan nanoparticles consists of three processes, namely diffusion, adsorption, and a combination of adsorption and diffusion (Wen et al., 2014). If the initial nanoparticle size is < 500nm then the growth process that occurs is adsorption, whereas if the particle size is > 500nm then a diffusion process occurs. Temperature does not significantly influence changes in nanoparticle size (Handani et al., 2016).

pH measurements were carried out after making the nanoparticles. The results of pH measurements obtained pH 4. The chitosan used in the research had a concentration of 1% with a volume of 80 mL. The greater the concentration of the chitosan solution, the lower the resulting pH. These results are similar to research conducted by Wijaya (2013). A 0.1% chitosan solution produces a pH of 4.412 compared to a 0.3% concentration with a pH of 4.030. The pH range is 3.5-5.5, the size of the nanoparticles produced is small. Increasing pH will cause the size of the nanoparticles to become smaller. A decrease in pH will cause an increase in protonated amino groups so that more phosphate groups are needed to form ionic bonds (Liu and Changyou, 2009).

Conclusion

Chayote (*Sechium edule*) which was macerated with 70% ethanol solvent produced a weight yield of 27.73%. The yield weight meets the requirements in the Herbal Pharmacopoeia II edition. Preparation of nanoparticles in 70% ethanol extract of Chayote using the ionic gelation

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method using 1% Chitosan and Na solution. TPP 0.5% ratio 2:1 produces a particle size of 556.5nm. The particle size meets the nanoparticle criteria (10-1000nm).

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