



## ANALYSIS OF THE EFFECT OF BODY LOSSION OF RATTUS NORVEGICUS ON GRANULATION FORMULATION OF CHITOSAN WASTE SHELL OF PLACUNA PLACENTA

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### ABSTRACT

Weight loss *Rattus norvegicus* wistar strain can be done by a variety of chemical and traditional medicine. One of the traditional medicines used for weight loss is to use *Placuna Placenta* chitosan. The purpose of this study was to determine the optimal concentration of *Placuna Placenta* granules as a weight loss agent for *Rattus norvegicus*. This study was an experimental study with 5 concentrations of 5%, 7.5% and 10% *Placuna Placenta* chitosan observed for 30 days. The positive control used weight loss drugs in the market, while the negative control used granule formulation without the addition of *Placuna Placenta* chitosan. The experimental research design posttest only control group design, treatment or intervention in the experimental group by comparing groups. Comparisons or differences were analyzed statistical tests using the ANOVA test. The optimal concentration of chitosan granule formulation of *Placuna Placenta* as a weight loss for *Rattus norvegicus* wistar strain was 10%.

**Keywords:** granule chitosan; weight loss of *rattus norvegicus* wistar

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## INTRODUCTION

*Placuna Placenta* is a shell that has a characteristic that is a thin shell that is yellowish brown sometimes brownish white (Ariyanti, Masruriati, et al. 2019). Content of dried *Placuna Placenta* has high nutritional value with water content of 16.47% -22.75%, ash content of 3.25% -4.15%, protein content of 63.89% -70.72%, fat content 2, 92% -4.96%, and carbohydrate content from 3.46% -4.72%. In addition, *Placuna Placenta* s contain chitosan which is a natural fiber when consumed by humans. The chitosan of the *Placuna Placenta* cannot be digested by the body. The mechanism of action in reducing body fat is by binding and inhibiting fat absorption. There are 3 fats that we know, namely Low Density Lipoprotein, High Density Lipoprotein and triglycerides. Triglycerides consist of vegetable oils and animal fats. Fats and oils are triglycerides which are composed of esters of glycerol and long-chain fatty acids as a result of the condensation of one glycerol molecule with three acid molecules fat (Hargono 2008) dan (Rismana E 2008).

Triglycerides are the main fat in human blood so they need to be limited. This is because triglycerides will accumulate in parts of the body such as the walls of blood vessels and the liver which increases the risk of diseases such as heart attacks and strokes. However, although limited triglyceride content is very important for humans because the body uses this fat as an energy source. In stabilizing kada fat, chitosan is needed that comes from natural resources (Hargono 2008). Chitosan is a polysaccharide that has an amine group so that chitosan has a strong positive partial charge. This positive charge causes chitosan to attract partially negatively charged molecules such as oils, fats and proteins. In this case, to make it easier to give it to *rattus norvegicus*, a granule preparation is made which will later be mixed in the husk. This is what causes Placuna Placenta chitosan to reduce cholesterol, uric acid, fat binding as well as body slimming. From this description, the researchers were interested in analyzing the weight loss effect of *rattus norvegicus* on the provision of Placuna Placenta chitosan waste granules (Ariyanti, Masruriati, et al. 2019) dan (Ariyanti Ariyanti, Eni Masruriati, Lina Lisiana Pasha 2021).

## **METHOD**

### **Tools and Materials**

The tools and materials used are glass, thermometer, mortar and stamper, pipette, analytical scale, pH meter, spatula, oven, porcelain dish, drying cupboard, Bunsen and tripod, Placuna Placuna Placenta waste, NaOH solution, HCl, NaOH solution. 50%, and Aquadest.

### **Sample Preparation**

Clean and dry the shells of the placenta in the sun for 8-12 hours. Drying can also be done using an oven with a temperature of 80 ° C for 24 hours. The shells of Placuna Placuna Placenta s are pounded in a mortar and pestle until they form a powder and then sieved using a mesh number 60 sieve to obtain an average powder of  $\pm 3$  mm (Ariyani, Farida 2008) and (Ariyanti Ariyanti, Eni Masruriati, Lina Lisiana Pasha 2021).

### **Manufacture of Placuna Placuna Placenta waste Chitosan**

Demineralization was carried out to remove minerals from the deproteinization results at a temperature of 60-70 ° C using a 70: 700 HCl solution (g powder / mL HCl) while stirring for 120 minutes then filtered to take the precipitate. The shells of Placuna Placuna Placenta s are washed with aquadest until the pH is neutral, then filtered and the precipitate is dried. The resulting demineralized powder is then removed from the protein group which is then carried out by the deacetylation process. Deacetylation is the removal of acetyl gums by dissolving the shells of Placuna Placuna Placenta s with NaOH 3% ratio (1: 5) then heated for 2 hours at 70°C - 80° then cooled and filtered to obtain Placuna Placuna Placenta solids. The solid is washed with water until the pH is neutral, then dried at 60 ° C for 36 hours (Faridah, Fathin 2012). The result of this process is a solid chitosan then dried in an oven at 60 ° C for 36 hours. The obtained Placuna Placuna Placenta chitosan shells were weighed and stored in a plastic bag at room temperature (Faridah, Fathin 2012). After obtaining the Placuna Placuna Placenta chitosan, it was then formulated into chitosan granules with several differences in concentration of 5%, 10%, 15%, positive control and negative control (Ariyanti Ariyanti, Eni Masruriati, Lina Lisiana Pasha 2021).

## Data analysis

In this study using ANOVA data analysis to analyze the weight loss effect of *Rattus norvegicus* on the provision of Placuna Placenta chitosan waste granule formulation at each concentration was significant or not significant, it was said to be significant if the significance value was  $<0.05$ .

## RESULTS

### Placuna Placenta chitosan weight

The weight of Placuna Placenta chitosan showed the results obtained after several times the isolation process of Placuna Placenta waste starting from the demineralization, deproteinase and deacetylase processes. Each process has a different level of difficulty and results. So that the chitosan produced varies. The weight of Placuna Placenta chitosan can be seen in table 1.

### Results of Analysis of the Effects of Weight Loss *Rattus norvegicus* on Placuna Placenta Chitosan Granules

Analysis of the weight loss effect of *Rattus norvegicus* when *Rattus norvegicus* was treated with Placuna Placenta chitosan granules is needed as a basic step in determining the next research process. The results of the analysis of the effects of weight loss *Rattus norvegicus* can be seen in Table 2.

Table 1.  
Weight of chitosan shells Placuna Placenta

Weight of sample (g)	Weight of deacetylation / weight of chitosan (g)
1000	567,67

Table 2.  
Results of the Weight Loss Effects of *Rattus norvegicus* Treated with Placuna Placenta Chitosan Granules

No	Group Treatment	Average weight loss (g)	Standart deviation
1	Concentration 5%	34	$\pm 0.002$
2	Concentration 10%	74	$\pm 0.021$
3	Concentration 15%	78	$\pm 0.011$
4	Kontrol Positif (orlistat)	60	$\pm 0.022$
5	Kontrol Negatif	5	$\pm 0.001$

### Results of Analysis of Anova Test on the Effect of Weight Loss *Rattus norvegicus* treated with Placuna Placenta chitosan granules.

The results of the anova analysis of the weight loss effect of *Rattus norvegicus* treated with Placuna Placenta chitosan granules can be seen in table 3. In table 3 the ANOVA analysis results show the significance of  $0.000 < 0.05$ , so it can be concluded that there are significant differences.

Table 3.  
Results Anova Analysis of the Weight Loss Effects of *Rattus norvegicus* Treated with Placuna Placenta Chitosan Granules

F	Signification	Information	Conclusion
78,23	0,000	Sig. $< 0,05$	Mean identic

Tabel 4.  
Results of Post Hoc Test Analysis of the Effect of Weight Loss *Rattus norvegicus*  
treated with Placuna Placenta chitosan granules

Concentration	concentration	Signification	Information	Conclusion
Concentration 5%	Concentration 10%	0,001	Sig. > 0,05	Mean identic
	Concentration 15%	0,002	Sig. < 0,05	Mean identic
	control positive	0,000	Sig. < 0,05	Mean identic
	control negative	0,000	Sig. < 0,05	Mean identic
Concentration 10%	Concentration 5%	0,003	Sig. > 0,05	Mean identic
	Concentration 15%	0,065	Sig. < 0,05	Mean not identic
	control positive	0,000	Sig. < 0,05	Mean identic
	control negative	0,000	Sig. < 0,05	Mean identic
Concentration 15%	Concentration 5%	0,001	Sig. < 0,05	Mean identic
	Concentration 10%	0,068	Sig. < 0,05	Mean not identic
	control positive	0,000	Sig. < 0,05	Mean identic
	control negative	0,000	Sig. < 0,05	Mean identic
control positive	Concentration 5%	0,000	Sig. < 0,05	Mean identic
	Concentration 10%	0,001	Sig. < 0,05	Mean identic
	Concentration 15%	0,000	Sig. < 0,05	Mean identic
	control negative	0,000	Sig. < 0,05	Mean identic
control negative	Concentration 5%	0,000	Sig. < 0,05	Mean identic
	Concentration 10%	0,001	Sig. < 0,05	Mean identic
	Concentration 15%	0,000	Sig. < 0,05	Mean identic
	control positive	0,000	Sig. < 0,05	Mean identic

## DISCUSSION

### Placuna Placuna Placenta chitosan weight

The weight of chitosan is the chitosan produced after going through several processes starting from demineralization, deproteination, and deacetylation. The results obtained in each process will decrease due to several things. The demineralization and deproteination processes are influenced by the solvent solution, the concentration of the solvent and the immersion time until a solid powder is obtained. Therefore, researchers must ensure the suitability of this matter when going through this process because the demineralization step is a determinant of the next process. The process that is no less important to get good quality chitosan is the deacetylation process. This is because if the chitin is deacetylated > 50% it is called chitosan. Deacetylated shellfish was determined by the degree of deacetylation as in the study (Hargono 2008), the best chitosan was obtained with the highest degree of deacetylation obtained by the deacetylation process using NaOH with a concentration of 50%. This is comparable with research (Ariyanti, Fajaryanti, et al. 2019) and (Ariyanti, Ariyanti, Masruriati et al. 2019) that NaOH solvent solution affects the levels of chitosan produced. According to (Sinaga, Luliana, and Fahrurroji 2015), deacetylation that occurs during the isolation stage of Placuna Placuna Placenta chitosan is a process of converting the acetyl group (-CH<sub>3</sub>COO-) in chitin to an amine group in chitosan with the addition of a high concentration of NaOH, namely 50%.

The reaction that occurs in the deacetylation process of Placuna Placuna Placenta chitin is an amide hydrolysis reaction of alpha 1,4-2-acet amide-2-deoxy-D-glucose. This process occurs with the release of the acetyl group from the chitin acetamide group with the OH ion concentration in the solution used. The OH concentration will be greater in a strong alkaline solution. Therefore, according to (Ariyanti, Masruriati, et al. 2019), the stronger a base is, the greater the OH<sup>-</sup> concentration in the solution, the greater the chitosan produced (Sartika, Alamsjah, and Sugijanto 2009).

### **Results of the Analysis of the Effects of Weight Loss Rattus norvegicus on the Placuna Placuna Placenta Chitosan Granules**

The weight loss effect of rattus norvegicus was due to the treatment of Placuna Placenta chitosan granules which were given via feed of these rats. The results were analyzed based on weight loss over 30 days. These results are based on table 2 showing a varied weight loss of rats in each treatment group. When compared with the positive control shows a good response so the results are not too far off. This is because the higher the levels of chitosan used, the more fat will cause weight loss of Rattus norvegicus. In a study conducted by ((Sartika, Alamsjah, and Sugijanto 2009) dan (Dewi, Dali Seniwati, and Muammar 2016), the absorption of triglycerides in goat fat was 3.36% within 60 minutes when treated with chitosan. So it can be interpreted that the chitosan mass affects the absorption of triglyceride fats. This is comparable to research (Hargono 2008) dan (Susanti, Happy Nursyam 2013), that the chitosan produced from the deacetylation process of 50% NaOH concentration will affect the results of the mass concentration of chitosan in the volume of fat so that it affects the absorption of total cholesterol. The results obtained in the (Hargono 2008), that the mass of 5 grams of chitosan in 50 ml of fat will affect the percentage of cholesterol absorption by 30.93% and 60 minutes of operation time shows the degree of cholesterol absorption is 45.46%. Therefore, the process of obtaining chitosan affects the results of fat absorption. The smaller the error occurs in the chitosan-making process, the maximum effect will be.

### **Results of Analysis of Anova Test on the Effect of Rattus norvegicus Weight Loss on Placuna Shell Chitosan Granules**

The results of the ANOVA test of the weight loss effect of rattus norvegicus on placenta chitosan granules showed that the mean (mean) weight loss for Rattus norvegicus for each chitosan concentration was different between groups, so the post hoc test was continued. This test is used to determine the weight loss of Rattus norvegicus with the concentration of chitosan which has a significant difference, indicated by a significance result of 0.000 less than 0.05. So to find out the difference between each concentration can be done with a further test, namely the post hoc test. Post hoc analysis of the scoring test for weight loss of rats at each chitosan concentration. The results of the post hoc test analysis in the significance column value > 0.05 means that there is an insignificant average difference, a value <0.05 means that there is a significant average difference. At a concentration of 15% with 10% there is no difference this is because although there is a small tp difference. So that if it is related to the optimal concentration between the two concentrations it can be said that the concentration of 10% is more optimal. This is because the smaller concentration can produce a weight loss effect that is almost the same as the 15% concentration group.

## CONCLUSION

Based on the results of a study conducted, the weight loss effect of *rattus norvegicus* on Placuna Placenta chitosan granules had a significant effect at a concentration of 10%.

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