

PREPARATION AND CHARACTERIZATION OF CHITOSAN FROM GOLDFISH SCALES FOR ANTI-DANDRUFF SHAMPOO

Eldi Firmansyah¹, Rini Hardiyanti^{1,2,3,✉}, Artha Klara Samosir¹, Minanda Payungta Beru Sitepu¹ and Nurmiahayati Boru Siagian¹

¹Department of Chemistry, Faculty of Mathematics and Natural Sciences, Universitas Sumatera Utara, Medan, North Sumatra 20155, Indonesia

²Pusat Unggulan Inovasi (PUI) Kitosan dan Material Maju, Universitas Sumatera Utara, Medan, North Sumatra 20155, Indonesia

³Cellulosic and Functional Materials Research Centre, Universitas Sumatera Utara, Medan, North Sumatra 20155, Indonesia

✉Corresponding Author: rinihardiyanti@usu.ac.id

ABSTRACT

The present study investigated the preparation of anti-dandruff shampoo formulated with goldfish scale containing chitosan as a bioactive compound of the product. The resulting chitosan is analyzed using FT-IR (Fourier Transform Infrared) spectroscopy to examine the functional groups before being transformed into nano chitosan by an ionic gelation process. Physicochemical properties of the product were evaluated including organoleptic test, pH, foam stability, viscosity, and antifungal activity. The results demonstrated that the formulated shampoo (3 g of chitosan) displayed a high inhibition of *Candida albicans* (27.30 ± 0.98 mm) with acceptable physical properties for commercial products.

Keywords: Chitosan, Goldfish scales, Shampoo, Anti-dandruff.

RASĀYAN J. Chem., Vol. 15, No.2, 2022

INTRODUCTION

Goldfish (*Cyprinus carpio* L.) is one of the most consumed fish in the world including Indonesia with a productivity of 332,206 tons and 375,200 between 2011 and 2012.¹ Remaining than the meat, the other parts of the fish are referred to as by-products. On average, 40-50% of edible fish portions are consumed. As a result, the remnants are not consumed. Such issues refer to the waste products that emerge as a consequence of such fish processing. Normally, such wastes are just thrown away without being processed. In the social and healthcare society environments, such little amounts of waste processing have become a critical concern. Unpleasant scents might result from the persistent accumulation of waste and may disturb the urban society.² Kara *et al.*, (2019)³, claimed that goldfish scale wastes contain elements like chitin (except other metallic ions) and chitosan which can be processed further. Chitosan is a biodegradable⁴, non-toxic polymer that can be used as a promising polymer material.⁵ Chitosan exhibits antibacterial and antifungal properties, constituting it an antimicrobial agent. It comprises both lysozyme enzyme and the amino polysaccharide group, which can inhibit microorganism growth.^{4,6} In addition, the free amino acid group in chitosan also prevents the growth of bacteria and molds (fungi). The bioactivity is the potential to be incorporated into a commercial product, one of which is anti-dandruff shampoo.^{7,8} Nweze (2001)⁹ stated that the scalp-repair processes may deteriorate leading to dirt accumulation in the hair which triggers the initiation of infection of the human scalp. Widowati (2016)¹⁰ stated that Chitosan has fungistatic properties, which indicates it can prevent yeast growth in dandruff. Previous studies have documented that chitosan-based shampoo at concentrations of 0.15% and 0.0075% exhibits fungistatic properties on scalp yeast infection. The intensity of dandruff was reduced by 94.81% and 77.27% in Proband I and Proband II, respectively. Hairfall intensity decreased by 91.52% and 88.25% in Proband I and Proband III, respectively. The Minimum Inhibitory Concentration (MIC) of chitosan shampoo is lower than that of chitosan, thus chitosan shampoo also aids in reducing the incidence of dandruff and hair fall.

EXPERIMENTAL

Material

The goldfish scales are obtained in the water areas of Medan, North Sumatra, Indonesia, in July 2021.

Sample Preparation

The scales were washed thoroughly and dried in an oven until constant weight. The dried scales were deproteinized using 5% NaOH in the ratio of 1:10 (m/v) and stirred for 2 hr at 100°C. The suspension was filtered and neutralized of its pH then dried in an oven until constant weight. The dried materials were demineralized using 1N HCl in the ratio of 1:10 (m/v) at 25°C for 1 hr. The solution was filtered, pH neutralized and dried in an oven at 50°C until constant weight. The dried materials or chitin can be stored for further experimentation.

Isolation of Chitosan

Chitin products from goldfish scales were deacetylated using 50% NaOH in the ratio of 1:10 (m/v) and stirred for 1 hr at 50°C. The suspension was filtered then the residue was neutralized of its pH and dried in an oven at 50°C until constant weight. The resulting material or chitosan was confirmed for its functional group using FT-IR analysis.¹¹

Synthesis of Nanochitosan

Chitosan products from goldfish scales were transformed into chitosan nanoparticles or nano chitosan with the ionic gelation method. Chitosan products were dissolved in 1% acetic acid, homogenized for 1 hr at 25°C and sprayed with 0.1% of tween 80. Fifty mL of chitosan solution was mixed with 0.1% tripolyphosphate (TPP) in the ratio of 5:1 and stirred until the suspension of nanoparticles is formed for 1 hr. The resulting materials of nano chitosan products were validated using a particle size analyzer (PSA) to determine its dimension scale and distribution in the product.

Formulation of Nanochitosan into Anti-dandruff Shampoo

The formulation of anti-dandruff shampoo is given in Table-1 as follows:

Table-1: Chemical Ingredients in the Formulation of Anti-Dandruff Shampoo

Ingredients	Formula (%)			Function
	F1	F2	F3	
Nanochitosan	1	3	5	Active agent
HPMC	0.9	0.9	0.9	Thickener
SLS	9	9	9	Detergent
TEA	1	1	1	pH stabilizer
Propylene glycol	15	15	15	Humectant
Methylparaben	0.18	0.18	0.18	Preservative
Na ₂ EDTA	0.1	0.1	0.1	Chelating agent
Propylparaben	0.02	0.02	0.02	Preservative
Green tea oil	0.5	0.5	0.5	Deodorizer
Distilled water	Ad 100	Ad 100	Ad 100	Dissolving agent

Note: HPMC = Hydroxypropyl methyl cellulose, SLS = Sodium lauryl sulfate, TEA = Triethanolamine

HPMC, Na₂EDTA, and SLS were dissolved separately in distilled water until homogenized. Methylparaben and propylparaben were mixed thoroughly in propylene glycol. Each chemical was then mixed and stirred gradually before addition with nano chitosan, TEA, and green tea oil until homogenized. The products were added with 100 mL of distilled water until an adequate volume was obtained.

Sensory Evaluation

The organoleptic test was performed by manually observing the forms, colors, and odors of the shampoo.

Determination of pH

One gr of shampoo products was dissolved in 10 mL of distilled water. The pH value was recorded using a digital pH meter.

Determination of Viscosity

An adequate volume of shampoo products was filled in the Ostwald viscometer and sucked using a pipette until reaching the threshold lines. The flow times were recorded to determine the viscosity of shampoo products.

Foam Stability Test

Approximately 0.1 gr of shampoo products were dissolved in 10 mL of distilled water in a reaction tube. The mixture was vortexed for 20 secs until foam is formed. The foam stability as indicated by the foam height was recorded.

Anti-Dandruff Activity

The antifungal activity of shampoo products was assayed using the good diffusion method. Briefly, 100 μ L of *Candida albicans* suspension was inoculated into Mueller Hinton Agar (MHA) medium until solidified. Wells were made using a punching hole at the appropriate distance and filled with 10 μ L of shampoo products of each formulation (F1, F2, F3). The plates were incubated at 37°C for 18–24 hr. The lack of fungal growth as indicated by the clear zone around wells was recorded using a digital caliper (mm).

RESULTS AND DISCUSSION

Chitosan can be extracted from a variety of biological sources, including fish scales. The chitin portion in the fish scales can be converted into chitosan under chemical processes. The presence of chitin was discovered in some of the previously studied fish scales. Milkfish scales, red snapper scales, and gourami scales, for example, contain about 0.4% and 3.7% of chitin.

Chitosan Characterization

The chemical group of chitosan from goldfish scales using FTIR analysis is given in Fig.-1:

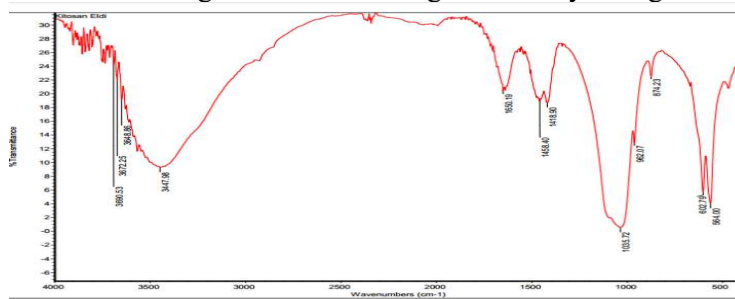


Fig.-1: The Graphic FTIR Result on the Goldfish Scales' Chitosan

The chitosan material showed an absorption peak at 3447.98 cm^{-1} and 1650.19 cm^{-1} indicating the presence of –OH and –NH stretch, respectively while at 1035.72 cm^{-1} showed the presence of the C-O-C functional group.

Nanochitosan

The size of the resulting nano chitosan was analyzed using a particle size analyzer (PSA) as given in Fig.-2.

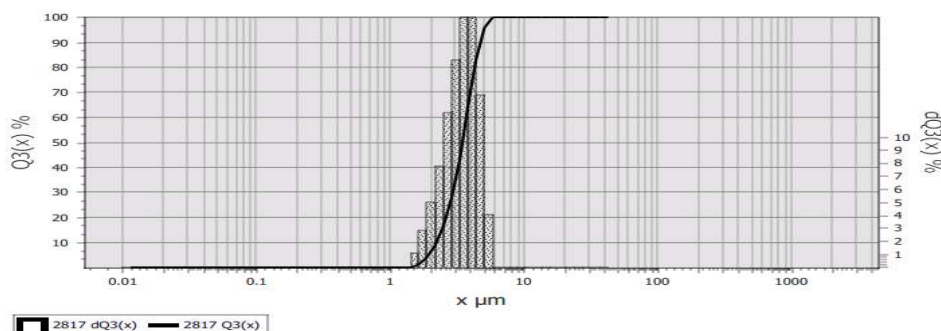


Fig.-2: The PSA Analysis Graphic

The nano chitosan was distributed from 10 nm until 42300 nm in the sample with an average value of 3450 nm. Based on the results, it can be seen that the particle size was not evenly distributed indicating the suboptimal process of homogenization. The larger particles emerged as the consequence of longer duration during the analysis which permits the agglomeration of the particles.¹²

Physicochemical and Biological Properties of Anti-dandruff Shampoo

Table-2: Characteristics of Anti-dandruff Shampoo Products

Sample	Organoleptic	pH	Viscosity (Poise)	Foam Height (cm)	Diameter of Inhibition Zone (mm)
F1	Homogenous, Translucent, Green tea scent	7	1.2277	8.5	27.25 ± 0.21
F2	Homogenous, Translucent, Green tea scent	7	0.6995	7	27.30 ± 0.98
F3	Homogenous, Translucent, Green tea scent	7	0.2144	7.2	24.80 ± 0.42

Sensory Evaluation

As can be seen in Fig.-2, the anti-dandruff shampoo products formulated using nano chitosan and green tea oil were acceptable in terms of product quality based on their texture (homogeneity), colors (translucent), and retain the green tea scent during preparation.

pH

As can be seen in Fig.-2, the pH value of each formulated product (F1, F2, F3) was similar or at 7.0. The pH value is congruent to the range quality of shampoo products based on the Indonesian National Standard (SNI) 06-4085-1996, which lies between 5.0-9.0.

Viscosity

The results from the F1, F2, and F3 formulations displayed the viscosity at 1.2277 Poise, 0.6995 Poise, and 0.2144 Poise, respectively using the Ostwald viscometer. It was demonstrated that the lower the viscosities, the more chitosan was added.

Foam Height

The results from the F1, F2, and F3 formulations produced foam heights of 8.5 cm, 7 cm, and 7.2 cm when reacted with distilled water. It was demonstrated that the more chitosan concentration, the less foam of the shampoo being produced.

Anti-Dandruff Activity

The most effective concentration of nano chitosan in the formulation of anti-dandruff shampoo was obtained at 3 gr of nano chitosan (F2) against *C. Albicans*¹³ with the diameter of inhibitory zone of 27.30 ± 0.98 mm.

CONCLUSION

Goldfish scales were used to extract chitosan, which was then characterized and transformed into nano chitosan. The organoleptic test, pH values, foam heights, viscosities, and anti-dandruff activities are used to evaluate the anti-dandruff properties of shampoo. The results demonstrate that the shampoo has a high anti-dandruff activity, with a *Candida albicans* inhibition zone of 27.30 ± 0.98 mm at 3 g of nano chitosan concentration, with acceptable physical properties for commercial shampoo products.

ACKNOWLEDGEMENT

The authors would like to express the highest gratitude to the Indonesian Ministry of Education, Culture, Research, and Technology under the scheme of *Program Kreativitas Mahasiswa* (PKM) which has funded this research, and Universitas Sumatera Utara which has facilitated a laboratory for this research.

REFERENCES

1. V. Yogaswari, Manado(2009).
2. N. Aziz, M. Fikri, F. Bill, and W. U. Pitoyo, *Hasanuddin Student Journal*, **1** (2017).
3. A. Kara, S. Tamburaci, F. Tihminlioglu, and H. Havitcioglu, *International Journal of Biological Macromolecules*, **130**, 266(2019), <https://doi.org/10.1016/j.ijbiomac.2019.02.067>
4. R. Jayakumar, N. T. Nwe, S. Tokura, and H. Tamura, *International Journal of Biological Macromolecules*, **40**, 3(2007), <https://doi.org/10.1016/j.ijbiomac.2006.06.021>
5. T. Bourtoom and M. S. Chinnan, *Food Science and Technology International*, **41**, (2018)
6. Z. Abedian, N. Jenabian, A. A. Moghadamnia, E. Zabihi, R. Pourbagher, and M. Rajabnia, *International Journal of Infectious Diseases*, **101**, 1(2020), <https://doi.org/10.1016/j.ijid.2020.09.154>
7. A. T. Utami, *International Journal of Infectious Diseases*, **101**, 1(2020), <https://doi.org/10.1016/j.ijid.2020.09.553>
8. P. K. Dutta, S. Tripathi, G. K. Mehrotra, and J. Dutta, *Food Chemistry*, **39**, 29(2011), <https://doi.org/10.1007/s12034-011-0032-5>
9. E. I. Nweze, *Medical Mycology*, **39**, 2(2001), <https://doi.org/10.1080/mmy.39.2.181.184>
10. S. C. Widowati, Bogor, (2016).
11. R. Hardiyanti, L. Marpaung, I. K. Adnyana, and P. Simanjuntak, *Rasayan Journal Chemistry*, **12(3)**, (2019), <https://doi.org/10.31788/RJC.2019.1235272>
12. I. F. M. Rumengan, P. Suptijah, N. Salindeho, S. Wullur, and A. H. Luntungan, Manado, (2018).
13. I. E. Mba, E. I. Nweze, E. A. Eze, Z. K. G. Anyaegbunam, *Infection, Genetics and Evolution*, **99**, 105256(2022), <https://doi.org/10.1016/j.meegid.2022.105256>

[RJC-6755/2021]