Evaluation and Comparison Anti-aging Facial Serum from Algae Extract

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ABSTRACT

The demand for cosmetics in Indonesia is relatively high, so cosmetics can now be considered a primary need. Therefore, various cosmetic products have emerged that are cheap and provide instant results but ignore the health aspects of the user. Several cosmetic products make a breakthrough by using natural ingredients. One natural material that can be used is microalgae, which can produce bioactive compounds and has a relatively faster production process than other natural materials. In this research, the microalgae used were Chlorella sp., Spirulina sp., and Sargassum sp. This research aims to make a serum based on the Chlorella sp., Spirulina sp., and Sargassum sp. microalgae. The research results showed that the standard testing of simplicial facial serum extracts of Spirulina sp., Chlorella sp., and Sargassum was by SNI No. 16-4399-1996. All the metrics analyzed, such as organoleptic characteristics, pH, specific gravity, viscosity, active compounds, and microbiological contamination, have successfully fulfilled the required requirements in SNI No. 16-4399-1996. The results of antioxidant activity testing showed that Chlorella sp. had higher antioxidant activity than the other three types of samples. However, the antioxidant results obtained are still meager and relatively weak, which means this serum does not fully contribute to antiaging. Further research needs to be carried out to obtain serum from microalgae with high levels of antioxidants, including using fresh simplicia, elevating algae concentration, or optimizing the operating conditions.

1. Introduction

Interest in cosmetics now no longer looks at gender and class. This is due to the development of the cosmetics industry, which is no longer segmented and targets all age ranges, young and old, even children, and no longer looks at gender [1]. However, along with the increasing number of cosmetics consumption in Indonesia, various cosmetic products have emerged that provide instant results but ignore the health aspects of the user [2]. Natural ingredients for medical and cosmetic purposes are increasingly being used in developed and developing countries [3]. This is due to increasing public literacy regarding natural ingredients, where natural ingredients will minimize side effects. The natural ingredients in question can be obtained from microalgae [4]. Microalgae can produce primary compounds such as protein, carbohydrates, and fat. Microalgae also contain active components that can be used in industrial fields such as food, pharmaceuticals, nutraceuticals, and cosmetics [5]. Microalgae contain active components such as antimicrobial, antifungal, and antiviral activities, as well as antioxidants and antibacterial substances [6]. Chlorella and spirulina are natural antioxidants that have the potential to delay, slow down, and prevent the oxidation process [7].

Antioxidants have many health benefits, such as protecting against degenerative diseases, the aging process, or antiaging cancer and playing a vital role in maintaining the quality of food products [8]. Several studies in the United States and Europe have proven that *Chlorella sp.* and *Spirulina sp.* have the potential to help the body overcome disorders caused by heavy metals such as Hg, Cd, and Pb by improving the immune system [9]. *Chlorella sp.* It has also been used massively in the clinical world as a detoxification method for individuals exposed to heavy metals, insecticides, pesticides, and hydrocarbons and has provided encouraging results [10]. This research aims to determine the effectiveness of serum antioxidants. The contribution of this study is to compare the results of serum based on the algae *Chlorella sp*., *Spirulina sp*., and *Sargassum sp*.

2. Research Methodology

2.1. Materials

The materials used in this research were N-Hexane (99% purity Merck, Germany), methanol (99.9% purity Merck, Germany), *Spirulina sp*., *Chlorella sp*., *Sargassum sp.* (powder form, obtained from marketplace), meanwhile vaseline, water, plastic wrap, aluminum foil, rose hydrosol, LAA, niacinamide, perfume, propylene glycol, refined glycerin, carboner 940, D-gluticol and DMDM (CV. Fashihul Alfarizki).

2.2. Procedures

1) Extraction of Microalgae

The simplicia (*Spirulina sp., Chlorella sp., and Sargassum sp.*) that had been weighed as much as 10 g was macerated with the help of a shaker using methanol in a ratio simplicial to methanol of 1:5 for 3 hours and produce the first extract. The first extract resulting from maceration was separated by distillation to separate methanol using a distillation flask at 68°C, and the first extract concentrate was produced. Then, the first extract concentrate was extracted with the help of a shaker using N-Hexane in a ratio of 1:5 for 3 hours to produce the second extract. The second extract undergoes the next distillation to separate N-Hexane at a temperature of 69°C until the algae extract is obtained (second extract concentrate).

2) Synthesis of Antioxidant Serum

Serum synthesis was started by making a serum base using distilled water (79.37%), propylene glycol (1.19%), refined glycerin (11.90%), carboner 940 (1.19%), D-glucitol (2.38%), and D-Glucitol, Dimethyol-dimethyl (DMDM) (3.97%). The serum base is made by mixing the ingredients individually, stirring until homogeneous, then leaving it for 24 hours. The serum base that has been obtained is mixed with distilled water (20.97%), rose hydrosol (6.53%), L-Ascorbic Acid (LAA) (2.54%), niacinamide (3.81%), algae extract (2.54%), and jasmine perfume (0.25%) to obtain an antioxidant serum from algae extract.

3) Serum Standard Test

Serum standard tests are based on SNI 16-4399-1996 [11], including pH, viscosity, density, organoleptic, homogeneity, and angka limping total (ALT) tests. ALT test refers to the microbiological analysis method (MA PPOM 61/MIK/06). As for antioxidant testing using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method at PT. Saraswanti Indo Genetech.

3. Results and Discussion

Algae and microalgae are used as active and primary ingredients of cosmetic products. Algae like *Chlorella sp*., *Spirulina sp*., and *Sargassum sp*. have antioxidant or anti-aging functions. This research was conducted to determine the effectiveness of serum antioxidants based on *Chlorella sp*., *Spirulina sp.*, and *Sargassum sp.* and determine the antioxidant activity using the DPPH (2,2-diphenyl-1-picrylhydrazyl) testing method and qualification based on SNI 16-4399-1996. Fig. 1. shows the results of the algae extract of *Chlorella sp*., *Spirulina sp*. *Chlorella sp*. and *Sargassum sp*. are light brown, while Spirulina sp. produces a dark brown extract.

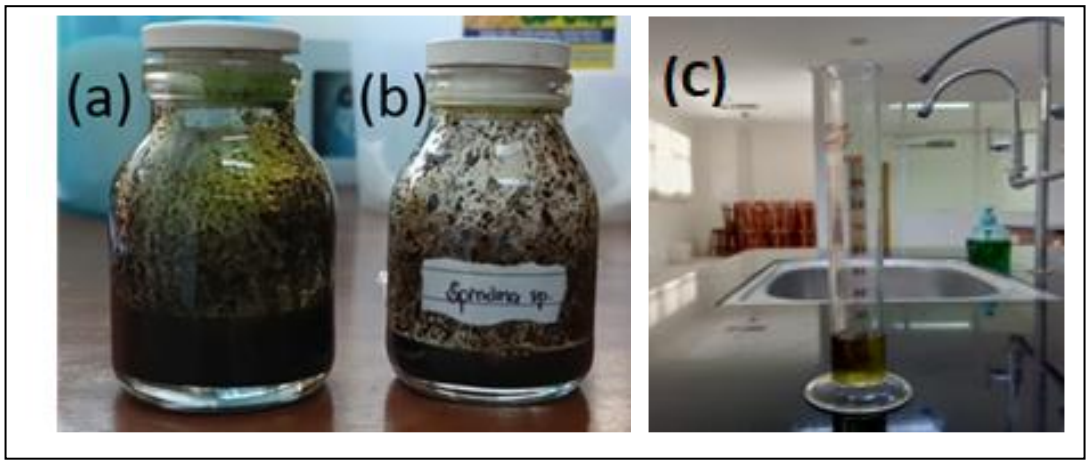

Fig. 1. Results of algae extract. a*) Chlorella sp*. b) *Spirulina sp.* c) *Sargassum sp.*

3.1. Extraction Process

In this research, extraction was carried out with the organic solvent methanol. The choice of extraction method using organic solvent was based on literature studies where previous research, such as research by Fasya et al. [12], which extracted Chlorella sp. using methanol solvent, and Firdayani et al. [13], who extracted *Spirulina sp.* with acetone solvent. Table 1 shows that the extraction process produces extracts of *Chlorella sp*., *Spirulina sp*., and *Sargassum sp*. This result is lower than research conducted by Ahmad Ganain [14], which obtained the yield of *Chlorella sp*. extract at 80.95%.

Table 1. Yield of Simplicial extraction of algae

Simplicia	Solvent	Yield microalgae extract (%)
Chlorella sp.	Methanol	48
Spirulina sp.	Methanol	44.2
Sargassum sp.	Methanol	50

3.2. Homogeneity Test

The homogeneity test is a test to see the stability of the serum through direct observation (visual) methods [15]. The homogeneity test is carried out to test whether any clots form to determine the quality of the serum formula. This homogeneity test was carried out through direct observation for one month with variables of 3, 7, 14, and 30 days. The results of homogeneity test observations on serum of *Spirulina sp*., *Sargassum sp*., and *Chlorella sp*., at intervals of 3, 7, 14, and 30 days are presented in Table 2.

Table 2. The result of the homogeneity test at different time

Serum Sources	Time (day)	Temperature (℃)	Homogeneity
Spirulina sp.			Homogenous
Chlorella sp.	3	25	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	7	26	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	14	25	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	30	27	Homogenous
Sargassum sp.			Homogenous

Apart from that, a homogeneity test was also carried out based on temperature reduction. The results of homogeneity test observations on the serum of *Spirulina sp*., *Sargassum sp*., and *Chlorella sp*. at different temperatures with time intervals of 10, 15, 20, and 25 minutes are shown in Table 3.

Table 3. The result of the homogeneity test at different temperature

Serum Sources	Time (min)	Temperature (℃)	Homogeneity
Spirulina sp.			Homogenous
Chlorella sp.	10	25	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	15	18	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	20	15	Homogenous
Sargassum sp.			Homogenous
Spirulina sp.			Homogenous
Chlorella sp.	25	13	Homogenous
Sargassum sp.			Homogenous

The results of observations in the homogeneity test are presented in Tables 2 and 3. The results show that the serum of *Spirulina sp*., *Chlorella sp*., and *Sargassum sp*. was classified as homogeneous and good; this is proven by the absence of separation in the form of lumps or residues in the serum formulation. Cahya and Fitri [15] stated in their research that the lack of oil and sediment phase separation in the serum proved good homogeneity.

3.3. pH Testing

pH testing is a test carried out to measure the acidity level of a product. This pH test is mandatory because the facial serum formulation with *Spirulina sp*., *Chlorella sp*., and *Sargassum sp*. is used topically. Therefore, the pH of this preparation must be the same as the pH of facial skin. The goal is that the facial serum with microalgae and algae extract does not irritate the skin. Based on the results of the tests, the pH values of *Spirulina sp*., *Chlorella sp*., and *Sargassum sp*. are 7, 6, and 7, respectively. Thus, based on the results of these observations, it can be concluded that the pH of the facial serum of *Spirulina sp*., *Chlorella sp*., and *Sargassum sp*. is by the standard pH of facial skin, namely between 4.5 and 6.5 [16] and SNI regulations that the appropriate pH range is 4.5–8.

3.4. Organoleptic Test

Organoleptic tests were carried out to observe the scent, color, and consistency of the serum, which were carried out by direct observation (visual) tests on days 0, 3, 5, and 7, and the results are presented in Table 4.

Table 4. The result of the organoleptic test

Serum Sources	Parameter	Day of observation			Description
		0	3	7	
Spirulina sp.	Scent	*Spirulina* typical	*Spirulina* typical	*Spirulina* typical	No alteration
	Color	Dark brown	Dark brown	Dark brown	
	Consistency	Viscous	Viscous	Viscous	
Chlorella sp.	Scent	*Chlorella* typical	*Chlorella* typical	*Chlorella* typical	No alteration
	Color	Light brown	Light brown	Light brown	
	Consistency	Viscous	Viscous	Viscous	
Sargassum sp.	Scent	Fishy (*Sargassum* typical)	Fishy (*Sargassum* typical)	Fishy (*Sargassum* typical)	No alteration
	Color	Clear cloudy	Clear cloudy	Clear cloudy	
	Consistency	Viscous	Viscous	Viscous	

Table 4 shows that the serum formula of the three types of microalgae tends to be consistent, and there is no change in scent, color, and consistency between days 0, 3, and 7. This shows that the serum formula has good quality.

3.5. Density Test

This density test was carried out on day 0 when the extraction was finished and was carried out on each serum extract. Based on the results obtained, the serums show the same density value, namely 1.02 g/mL. This indicates that the three facial serums meet the standard density requirements for facial serum, and SNI 16-4399-1996 states that the normal specific gravity is 0.95-1.05 g/mL.

3.6. Viscosity Test

The viscosity test aims to determine the product's consistency, which affects the product's spreadability, such as being easy to remove from the packaging but not easily flowing from the hand [17]. This viscosity test uses an Ostwald viscometer. The results of viscosity observations are presented in Table 5.

Table 5. The result of the viscosity test

Serum Sources	Viscosity		SNI 16--4399-1996
	Ns/m²	*cp*	
Spirulina sp.	2.54	2540	
Chlorella sp.	2.44	2440	2000-50000 (cp)
Sargassum sp.	2.3	2300	

The data in the table shows the average viscosity of serum. These values align with SNI 16-4399-1996, which regulates the standard for serum to have a viscosity range of 2000-50000 cp.

3.7. Total Plate Number test (ALT/Angka Lempeng Total)

The principle of ALT is to count the growth of aerobic mesophyll bacterial colonies on the right media. After incubation at the appropriate temperature and time, the number of bacterial colonies growing on the agar plate was counted [18]. The analysis of ALT microbial contamination refers to the SNI method (Indonesian National Standard (SNI) 16-4399-1996, 1996). Samples were tested by the Yogyakarta Health and Calibration Laboratory Center, and the results obtained are presented in Table 6. The table shows that the number of mesophyll bacterial colonies (ALT) is still below the maximum value required by SNI 16-4399-1996. Variations in the number of bacterial colonies observed in each serum can be attributed to varying degrees of cleanliness in the materials, containers, and instruments employed throughout the serum preparation process. In addition, harmful microbial contamination can arise during the production and preservation of serum.

Table 6. The result of the total plate number (ALT) test

Analysis	Chlorella Serum (CFU/gr)	Spirulina Serum (CFU/gr)	Sargassum Serum (CFU/gr)	SNI 16-4399-1996
ALT (kol/g)	27	34	67	Maks. 102

3.8. Antioxidants Test

A quantitative antioxidant activity test using the DPPH method is expressed in the IC50 (inhibition concentration) value. According to Andayani et al. [19], the amount of antioxidant activity can be identified through the IC50 value, namely the concentration of the sample solution needed to inhibit 50% of DPPH free radicals. The test results for facial serum made from fresh *Spirulina* platensis extract with methanol solvent had antioxidant activity with an IC50 value of 251664 ppm. In comparison, the IC50 value for facial serum made from *Chlorella sp.* extract was 285105 ppm, and the IC50 value for facial serum made from *Sargassum sp.* extract was 335830 ppm. *Spirulina sp.* platensis crude extract has the strongest antioxidant activity, followed by *Chlorella sp.* extract, and the weakest is *Sargassum sp.*. However, the antioxidant activity of these three serums is significantly deficient. The serum obtained lacks antiaging properties due to its insufficient antioxidant value.

An activity is vital if the IC50 value is 50-100 ppm. Mardawati et al. [20] stated that a compound is said to be a powerful antioxidant if the IC50 value is <50 ppm, strong for an IC50 value of 50-100 ppm, moderate level at an IC50 value of 100-150 ppm, and classified as weak if the IC50 value is

more than 200 ppm. Bariyyah et al. [21] found in their research that *Chlorella sp*. has an IC50 antioxidant activity value of 18.61 ppm. Sedjati [22], in his study, found the antioxidant activity value of *Sargassum sp.* amounted to 69.27 ppm. Anam et al. [23] stated in their research that *Spirulina sp.* platensis powder extract had an IC50 value of 11.203 ppm using ethyl acetate solvent. Firdiyani [13] noted that the IC50 value of fresh *Spirulina sp.* extract with Acetone solvent was 65.89 ppm and 76.36 ppm with ethanol solvent.

4. Conclusion

Spirulina sp., *Chlorella sp*., and *Sargassum sp.* can be natural sources of facial serum. The research results showed that the standard testing of Simplicia facial serum extracts of *Spirulina sp*., *Chlorella sp*., and *Sargassum sp.* align with SNI No. 16-4399-1996. The metrics examined, including organoleptic properties, pH, specific gravity, viscosity, active substances, and microbiological contamination, met the requirements specified in SNI No. 16-4399-1996. Nevertheless, due to its significantly low antioxidant value, the algae serum did not acquire its antiaging capabilities. The results of antioxidant activity testing showed that *Chlorella sp.* had higher antioxidant activity than the other three types of samples. Additional investigation is required to acquire serum with very high (IC 50 <50) or elevated (50 < IC 50 <100) amounts of antioxidants from algae. This can be achieved through the selection of algae, utilization of fresh simplicia, augmentation of algal extract concentration, and appropriate storage and processing of the serum. Enhance or optimize the working conditions for testing serum.

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