

Determination of Total Phenolic Content and Sunscreen Activity of 96% Ethanol Extract of Sea Grape (*Caulerpa racemosa*) Using the UAE Method (Ultrasound-Assisted Extraction)

Dwi Susiloningrum^{1*}, Vita Permansari²

Institut Teknologi Kesehatan Cendekia Utama Kudus, Indonesia

*Corresponding Author: dsusiloningrum@gmail.com

Abstract. Sea grapes, with the Latin name *Caulerpa racemosa*, are a marine plant in the *Caulerpaceae* family. Sea grapes are known to contain many active compounds, one of which is phenolics, which also play a role in sunscreen activity. The purpose of this study was to determine the total phenolic content and sunscreen activity of 96% ethanol extract of fresh sea grapes (*Caulerpa racemosa*) using the UAE (*Ultrasound Assisted Extraction*) method. This research is an experimental study, namely the determination of total phenolic content and sunscreen activity of 96% ethanol extract of sea grapes (*Caulerpa racemosa*) using the UAE (Ultrasound Assisted Extraction) extraction method with 96% ethanol solvent and phytochemical screening tests were carried out. Secondary metabolites found in fresh sea grapes positively contain phenolic compounds. The results of the total phenolic content contained in sea grape ethyl acetate extract using the Ultrasound Assisted Extraction (UAE) method showed an average total phenolic content of 1.30%. The Sun Protection Factor (SPF) value for the 96% ethanol extract of sea grapes (*Caulerpa racemosa*) is 4.43, which is included in the minimal protection category.

Key Words : Sea Grape (*Caulerpa racemosa*), total phenolic, sunscreen

INTRODUCTION

Sea grapes are a leading commodity with potential as a functional food. They are rich in fiber and a source of natural antioxidants that are readily available and abundant (Novaczek ; Chew *et al.*, 2008). *Caulerpa racemosa* or better known by the local name Latoh is a type of green seaweed (Chlorophyta) which is widely distributed in tropical to subtropical waters (Tapotubun *et al.*, 2020).

Sea grapes can be used for health as a medicinal ingredient because they contain chemical compounds that have biological activity or bioactive substances such as antibacterial, antioxidant, anticancer, antifungal. Some secondary metabolites contained in sea grapes include phenolics (Nursandi, 2014). Phenolics are secondary metabolites characterized by aromatic rings containing one or two hydroxyl groups (OH) directly bound to the aromatic ring. The bioactive phenol compound is the compound responsible for sunscreen activity. The phenol compound content in *Caulerpa racemosa* can function as a defense against ultraviolet (UV) radiation (Ersalina, 2019). Sunscreen is a preparation that contains chemical compounds in conditions where ultraviolet (UV) radiation energy does not directly hit the skin, thus avoiding the negative effects of ultraviolet (UV) rays (Widyawati *et al.*, 2019). Sunscreen functions to absorb or scatter sunlight so that the intensity of the rays that can reach the skin is much less than it should be (Ulfa *et al.*, 2022).

The extraction process for secondary metabolites is influenced by several factors, including the extraction method and the type of solvent used. The method used in this study was the UAE method. Ultrasound-Assisted Extraction (UAE) is an accelerated method (Ayu *et al.*, 2020). Phenolic compounds tend to dissolve in polar solvents (Julianto, 2019). The choice of solvent is adjusted to the polarity of the desired compound, based on the principle of "like dissolves like," where a solvent will dissolve compounds with the same polarity. The type of solvent and temperature used in extraction affect the extract yield and chemical content (Suryani *et al.*, 2016). Based on this, a study was conducted to determine the total phenolic content and sunscreen activity of a 96% ethanol extract of sea grape (*Caulerpa racemosa*) using the Ultrasound-Assisted Extraction (UAE) method.

METHODS

Type of Research

This research is an experimental study with quantitative analysis, namely the determination of total phenolic content of 96% ethanol extract of sea grapes (*Caulerpa racemosa*) using the Ultrasound Assisted Extraction (UAE) method, phytochemical screening, and determination of total phenolic content using UV-Vis Spectrophotometry.

Research Instruments

The tools used were analytical balance (Ohaus), glass bottles (dark), blender (Philips), filter paper (Whatman-50), funnel, measuring cylinder (Pyrex), Erlenmeyer flask (Pyrex), rotary evaporator, water bath (DHH-4), porcelain cup, spatula, label paper, spectrophotometer (Shimadzu), ultrasonic bath (Branson 2002). The materials used in the study were sea grape samples, distilled water (H₂O), 96% ethanol, sodium carbonate (Na₂CO₃), 10% lead acetate, 31% FeCl, Folin-Ciocalteu reagent, and gallic acid standard.

Data Collection Techniques

Research permit application, plant identification, raw material preparation, sea grape extraction, organoleptic examination of the extract, phytochemical screening, and determination of total phenolic content.

Data Analysis Techniques

Data analysis was conducted using quantitative descriptions, with phytochemical screening based on the formation of color and sediment in sea grape (*Caulerpa racemosa*) extract. SPF value test: The SPF value can be calculated using the Mansur equation (Karina, 2015).

$$SPF = CF \times \sum_{290}^{320} EE(\lambda) \times I(\lambda) \times abs(\lambda)$$

Information :

EE : Erythema Spectrum of Effects

CF : Correction Factor (=10)

I : Light Intensity Spectrum

Abs : Absorbance of Sunscreen Samples

RESULTS AND DISCUSSION

Preparation of Simplisia

Fresh sea grapes (*Caulerpa racemosa*) were obtained from Blebak Beach in the Mlonggo area of Jepara. The freshest, dark green sea grapes were selected, harvested 30-40 days after harvest. The simple drugs were wet sorted using running water, then drained.

Sea Grape Extraction (*Caulerpa racemosa*)

Factors affecting the extraction rate are extraction time, sample size, temperature, and solvent type. During the extraction process, the active ingredient will be dissolved by the solvent, which matches its polarity (Ministry of Health of the Republic of Indonesia, 2000). This study used 100 grams of finely ground sea grapes (*Caulerpa racemosa*) and 425 ml of 96% ethanol. The solution was stirred and sonicated three times for 2 minutes. Afterward, the filtrate was filtered, separating the residue. The residue was added with 375 ml of solvent, then sonicated again for 2 minutes and repeated 3 times. The filtrate obtained was then concentrated with a rotary evaporator and water bath at a temperature of 40o to produce a thick extract. From 100 grams of Sea Grape (*Caulerpa racemosa*) simplicia that had been extracted and concentrated, the weight of the thick extract was 15.52 grams and the yield was 15.52%.

Table 1. The organoleptic test results

Parameters	Result
Shape	Thick
Color	Blackish green
Odor	Slightly fishy
Taste	-

Determination of Maximum Wavelength

The research results showed that the maximum wavelength obtained was 766 nm. This is a 6 nm difference from the study by Dewantara, Ananto, & Andayani (2021), which obtained a wavelength of 760 nm. The obtained wavelength was then used to determine the operating time, standard curve, and sample content.

Determination of Operating Time (OT)

The operating time obtained differed by 11 minutes from the 45th minute of the study by Dewantara, Ananto, & Andayani (2021). The operating time results were then used to determine the standard curve. To determine the operating time, 0.3 ml of a standard gallic acid solution at a concentration of 30 ppm was taken, 1.5 ml of Follin-Ciocalteu reagent was added, allowed to stand for 3 minutes, and 1.2 ml of 7.5% Na₂CO₃ was added. The measurement was performed at a wavelength of 766 nm for 1 hour. The absorbance was 0.299, which was stable at 56 minutes.

Total Phenolic Yield of 96% Ethanol Extract of Sea Grape (*Caulerpa racemosa*)

The results of the total phenolic research of 96% ethanol extract of Sea Grapes (*Caulerpa racemosa*) obtained an average total phenolic content of 1.30% extract, which means that in every gram of 96% ethanol extract of Sea Grapes (*Caulerpa racemosa*) there are phenolics equivalent to 13.30 mg of gallic acid. The results of the total phenolic value of 96% ethanol extract of Sea Grapes (*Caulerpa racemosa*) can be seen in table 2.

Table 2. Results of Total Phenol Value in 96% Ethanol Extract of Sea Grapes (*Caulerpa racemosa*)

Sample	Extract Weight (mg)	Absorbansi Ppm	Total Phenolic Content (%)	Average
Etanol 96%	10 mg	0,271	1,28	1,30 %
		0,276	1,33	
		0,273	1,30	

Research conducted using a quantitative test of total phenolic content was conducted to determine the total phenolic content in 96% ethanol extract of Sea Grapes (*Caulerpa racemosa*) using the Follin Ciocalteu method. The more intense the color, the greater the phenol content in the fraction (Lestari *et al.*, 2018).

Sunscreen Activity of 96% Ethanol Extract of Sea Grapes (*Caulerpa racemosa*)

Sea grape (*Caulerpa racemosa*) samples at a concentration of 200 ppm were measured using a UV-Vis spectrophotometer. Absorbance was measured at a wavelength of 290-320 nm at 5 nm intervals. The average Sun Protection Factor (SPF) value for the 96% ethanol extract of sea grapes (*Caulerpa racemosa*) at a concentration of 200 ppm was 4.430, as shown in Table 3.

Table 3. SPF value of 96% ethanol extract of Sea Grape (*Caulerpa racemosa*)

Concentration ppm	Replication			Average	Protection Category
	I	II	III		
200	4,387	4,426	4,477	4,430	Minimum Protection

Sun Protection Factor (SPF) is an indicator to protect the skin from UV exposure. Where the higher the SPF value of a sunscreen ingredient, the better its protective ability to the skin (Schalka & Manoel, 2011). SPF measurements have been classified based on the Food and Drug Administration (FDA), which groups the effectiveness of sunscreen preparations in the SPF value category. According to the Food and Drug Administration (FDA), the requirements for a good sunscreen preparation are >15.

The average measurement of the SPF value of 96% ethanol extract at a concentration of 200 ppm is 4.43 so this is included in the minimal protection category.

CONCLUSION

Based on the research results, it can be concluded that:

1. The secondary metabolites of the 96% ethanol extract of sea grapes (*Caulerpa racemosa*) using the Ultrasound-Assisted Extraction (UAE) method positively contained phenolic compounds and tannins.
2. The results of the total phenolic content in the 96% ethanol extract of sea grapes (*Caulerpa racemosa*) using the Ultrasound-Assisted Extraction (UAE) method obtained an average total phenolic content of 1.30%.
3. The SPF value of the 96% ethanol extract of fresh sea grapes (*Caulerpa racemosa*) at a concentration of 200 ppm was 4.43, which falls within the minimal protection category.

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