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Total Phenolic Content of Methanol Extract from Buni Fruits (*Antidesma bunius L.*) Water

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Abstract. Buni Fruit Water was extracted with methanol solvent using Ultrasound technology at a temperature of 50°C for 45 minutes. Total Phenolic Content contained in the methanol extract of buni fruits water is 67 mg / g in GAE. The results of interpretation using GCMS obtained three phenolic compounds, namely 3- (Hydrazinomethyl) Phenol, 5 Allyl-2 Methoxy Phenol and 3 (3,5-di-Tertier Butyl, 4-Hydroxyphenyl) Propionic Acid. IR Spectrum analysis results obtained aromatic and OH group as a marker of the presence of phenolic compounds. These results indicate that the Buni fruit water can be developed as functional food.

1. Introduction

Buni Fruit (*Antidesma bunius L.*) is a plant belonging to the genus of antidesma which is known to have various benefits such as antibacterial, antithrombotic [1], anti-inflammatory [2], antioxidant [3] and cytotoxic [4] properties. Traditional societies often use the Fruit Buni plant to treat high blood pressure, palpitations, anemia, syphilis, anti-cancer, and anti-radicals [5]. Buni Fruit consists of several parts that may have different chemical contents such as Flesh, Seeds and Water.

The water of Buni fruit is one of the parts of the buni fruit that is widely used in traditional medicine. Besides having a distinctive taste, Buni fruit water is also believed to have a high total phenol content. It is indicated because the water of the Buni fruit has a distinctive red-purple color which is one indicator of the presence of phenolic compounds [6].

Phenolic compounds have the characteristic form of an aromatic ring that binds at least one hydroxyl group. Phenolic compounds are good electron donors because their hydroxyl groups can directly contribute to antioxidant activity [7]. The ability of a plant to be used as traditional medicine is influenced by the Total Phenolic Content therein. The higher the Total Phenol Content, the better the ability to fight free radicals.

Optimizing the Total Phenolic Content from Buni fruit water will be extracted using methanol as a solvent using ultrasound technology in the extraction process. Utilization of ultrasound technology in the extraction process provides advantages in time and cost efficiency [6].

2. Methodology

2.1 Material

Buni Fruits (*Antidesma bunius L.*) obtained in the Moncongloe, Maros District, South Sulawesi. Buni fruit that is used is a buni fruit that has been ripe with a purplish red color. Parts of Buni fruit water are



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separated from flesh and seeds. All the chemicals such as Methanol (CH_3OH), Folin-Ciocalteu Phenol, Sodium Carbonate (Na_2CO_3), and Gallic Acid was purchased from Merck.

2.2 Procedure

2.2.1 Buni Fruits Water Extraction. 300 g of Buni fruit water was extracted with methanol at a temperature of 50°C for 45 minutes using sonic power Ultrasound of 405 on a wave of 40 kHz. Then the evaporation process is carried out at a temperature not exceeding 45°C to obtain a thick extract.

2.2.2 Characterization of Extract. The extract was characterized by Prestige-21 Shimadzu Infrared spectroscopy at the range of $400\text{--}4000\text{ cm}^{-1}$ using KBr pellets and Characterization with GCMS using Ultra Shimadzu QP2010 Gas Chromatograph Mass Spectrometer.

2.2.3 Determination of Total Phenolic Content. 1 mL standard gallic acid solution (5, 15, 25, 35, 45, 55 dan 65 mg / L) and Buni Fruits Water extract samples were added with 1 mL of Folin Ciocalteu and 5 mL of 10% Na_2CO_3 . measurement at the wavelength of 765 nm by using Orion Aquamate 8000 UV-Vis Spectroscopy after the sample was stored at room temperature for 1 hour.

3. Results and Discussion

Total Phenolic Content was measured using the Folin-Ciocalteu method. Folin-Ciocalteu will oxidize phenolic hydroxy groups and reduce heteropolyacids (fosfomolibdat- fosfotungstat) contained in the Folin-Ciocalteu reagent into a molybdenum-tungsten blue complex that can be detected with a spectrophotometer [8].

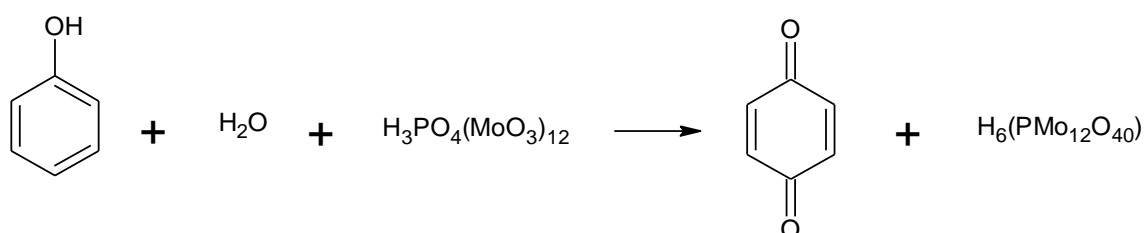


Figure 1. The chemical reaction of Folin-Ciocalteu with Phenol [8]

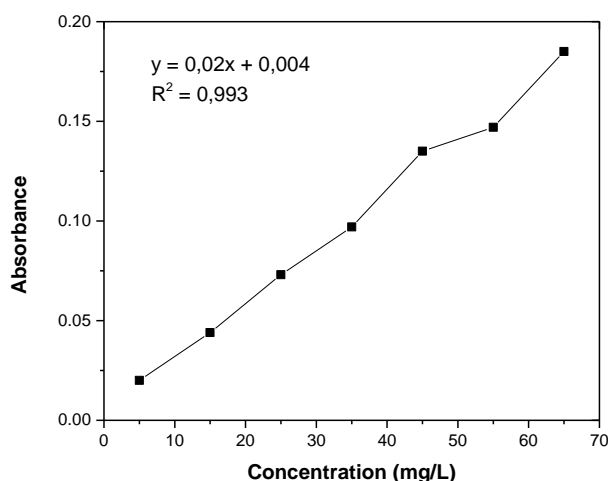


Figure 2. Gallic Acid Standard Curves in the Concentration Range of 5 ppm - 65 ppm

The results showed (Figure 2, Table 1) that Total Phenol Content in the methanol extract of Buni Fruits Water was 67 mg/g in GAE (Gallic Acid Equivalent). Total Phenolic Content of sample is influenced by the number of phenolic compounds contained therein [9]. The higher the consistency of phenolic compounds, the more phenolic ions can reduce heteropoly (phosphomolybdate-

phosphotungstate) into molybdenum-tungsten complex so that the resulting blue color becomes more concentrated.

Table 1. Measurement results of Total Phenolic Content from methanol extract of Buni Fruits Water

No	Name of Sample	Absorbance
1	Standard Solution 5 ppm	0,02
2	Standard Solution 15 ppm	0,044
3	Standard Solution 25 ppm	0,073
4	Standard Solution 35 ppm	0,097
5	Standard Solution 45 ppm	0,135
6	Standard Solution 55 ppm	0,147
7	Standard Solution 65 ppm	0,185
8	Methanol Extract of Buni Fruits Water	0,138

Measurement of samples by using GCMS aims to determine the chemical structure of the compounds contained in the methanol extract of buni fruit water. The results of the analysis using GCMS obtained 20 compounds detected in the sample (Table 2, Figure 3) which are known to contain three phenolic compounds namely 3-(Hydrazinomethyl) Phenols (Figure 4); 5 allyl-2 methoxy phenols (Figure 5) and 3(3,5-di-tertier Butyl, 4-Hydroksyphenyl) Propionic Acid (Figure 6).

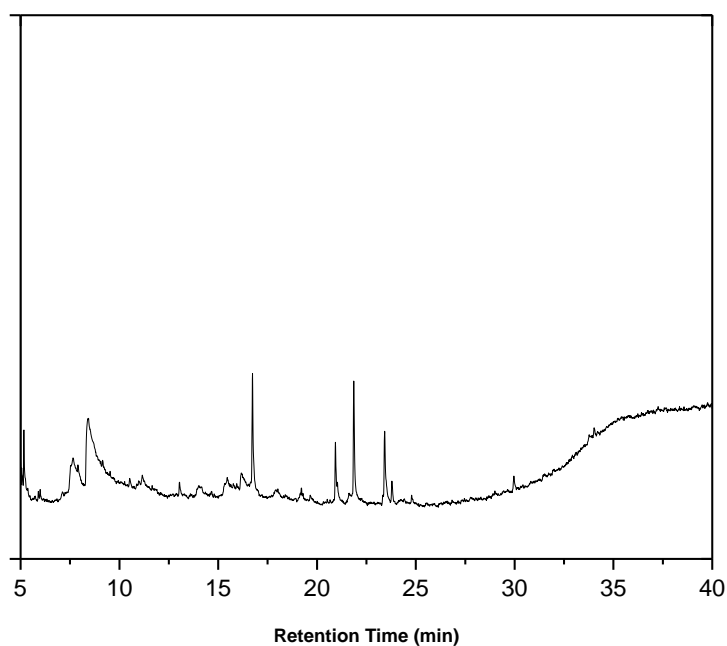


Figure 3. GCMS Spectrum of Methanol Extract from Buni Fruits Water

Table 2. Measurement results with GCMS from methanol extract of Buni Fruits Water

No	Retention Time	Name of Compound
1	5.056	O-Butylsourea
2	5.163	3-(Hydrazinylmethyl) Phenol
3	7.901	Diethoxymethyl Acetate
4	8.435	1,2,3_Propanetriol
5	8.542	N-butyl-N ¹ -(3,4-dichlorophenyl)-N-methyl- Neburon
6	13.039	5-Allyl-2-Methoxyphenol

7	15.325	3-(Tert-Butylsulfanyl)-4,4-Bis(Trifluoromethyl)-4H-Pyrazole
8	15.459	1-Octanol,3,7- Dimetil
9	16.168	Propanoic Acid, 2-Methyl-, 1-(1,1-Dimethylethyl)-2-Methyl-1,3-Propanediyl Ester
10	16.731	Propanoic acid, 2-methyl-, 1-(1,1-dimethylethyl)-2-methyl-1,3-propanediyl ester
11	19.204	1-Octadecene
12	20.933	Hexadecanoic Acid
13	21.029	3-(3,5-di-Tert-Butyl-4-Hydroxyphenyl)-Propionic Acid
14	21.608	Hexadecanoic Acid
15	21.858	Hexadecanoic Acid
16	23.325	9,12-Octadecadienoic Acid
17	23.420	9-Octadecenoic Acid
18	23.792	Octadecanoic Acid
19	29.959	1,2-Benzenedicarboxylic Acid
20	34.019	2,6,10,14,18,22-Tetracosahexaene

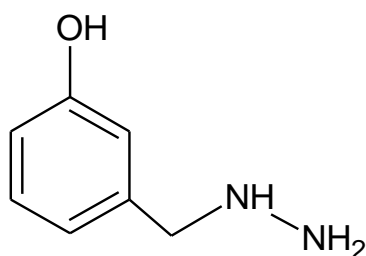


Figure 4. 3-(Hydrazinylmethyl) Phenol

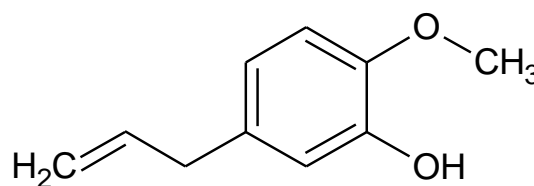


Figure 5. 5-Allyl-2-Methoxy Phenols

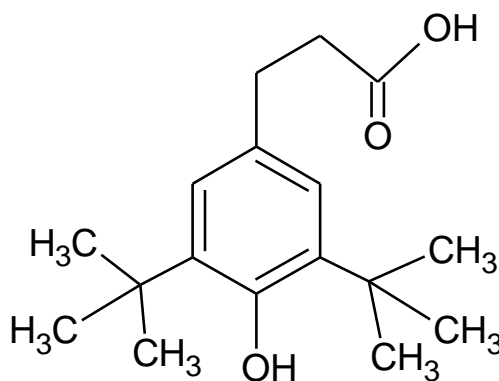


Figure 6. 3-(3,5-di-tert-butyl, 4-Hydroxyphenyl) Propionic Acid

The results of the interpretation using FTIR showed the presence of phenol groups. The aromatic group of phenolic compounds sometimes appears in the range of wave numbers 1800 - 900 cm^{-1} [10]. There are the typical signal aromatic ether group at wave number 1230,84 cm^{-1} [11]. The aromatic group is also in the range of wave numbers 2000 - 1660 cm^{-1} in various bond combinations. In contrast, the presence of OH groups is in the range of 3570 - 3200 cm^{-1} [12]. Other functional groups identified through GCMS such as carboxylic acids were detected by reading with FTIR at wave number 1728,28 cm^{-1} [11] (Figure 7). These results corroborate the analysis using GCMS, which detects the presence of aromatic and OH functional groups as markers for the presence of phenolic compounds.

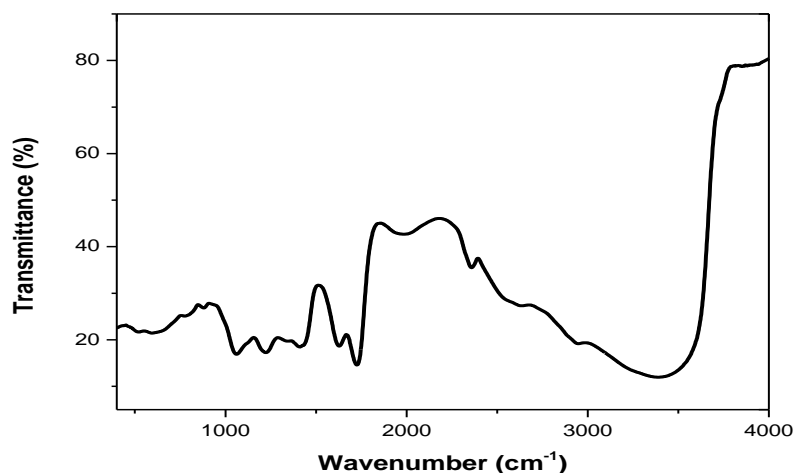


Figure 7. IR Spectrum of Methanol Extract from Buni Fruits Water

4. Conclusion

Methanol extract from the water of Buni fruit with a total phenolic content of 67 mg/g in GAE has the potential to be developed as a functional food. It is supported by the presence of several phenolic compounds contained in the Water of Buni fruits. The three phenolic compounds that are indicated contained in the water of Buni Fruits are 3- (Hydrazinomethyl) Phenols; 5 allyl-2 methoxy phenols and 3 (3,5-di-tertier Butyl, 4-Hydroxyphenyl) Propionic Acid.

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