

DETERMINATION OF SUN PROTECTIVE FACTORS (SPF) AND ANTIOXIDANT ACTIVITY OF ETHANOLIC EXTRACT OF RAMBUTAN RIND (*NEPHELIUM LAPPACEUM L.*)

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ABSTRACT

The fruit peels of rambutan (*Nephelium lappaceum L.*) are organic waste with many potential health benefits but have not been used optimally. The aim of this study were to determine the sun protective factor (SPF) and antioxidant activity of ethanolic extract of rambutan rind. The rambutan rind was extracted by maceration method using ethanol as a solvent. A simple, rapid and reliable in vitro method was used to assess the SPF values by measuring the absorbance between 290-320 nm of diluted extract at every 5 nm intervals using UV spectrophotometry. The SPF values were calculated based on the recorded absorbance using a simple mathematic equation developed by Mansur. The antioxidant activity of rambutan rind was determined by the 1,1-diphenyl-2-picryl-hydrazil (DPPH) assay whereas vitamin C used as a positive control. The SPF values of rambutan rind in concentration series of 50, 100, 150 and 200 $\mu\text{g}/\text{mL}$ were 6.47 ± 0.45 , 9.26 ± 0.28 , 13.01 ± 0.33 , and 16.17 ± 0.63 respectively. The IC_{50} value of rambutan rind was $41.47 \pm 3.89 \mu\text{g}/\text{mL}$ whereas the IC_{50} value of vitamin C was $24.87 \pm 0.69 \mu\text{g}/\text{mL}$. Based on the result, the ethanolic extract of rambutan rind has potency as sunscreen and antioxidant.

Keywords :rambutan rind, SPF, antioxidant, IC_{50}

PENENTUAN NILAI *SUN PROTECTIVE FACTORS* (SPF) DAN AKTIVITAS ANTIOKSIDAN EKSTRAK ETANOL KULIT RAMBUTAN (*NEPHELIUM LAPPACEUM L.*)

ABSTRAK

Kulit buah rambutan (*Nephelium lappaceum L.*) merupakan sampah organik dengan banyak manfaat kesehatan, namun belum dimanfaatkan secara optimal. Penelitian ini bertujuan untuk menentukan nilai SPF dan aktivitas tabir surya ekstrak etanol dari kulit buah rambutan. Kulit buah rambutan diekstraksi dengan menggunakan metode maserasi dimana etanol digunakan sebagai pelarutnya. Nilai SPF ditentukan dengan menggunakan metode *in vitro* yang sederhana, cepat dan dapat diandalkan, yakni dengan mengukur serapan dari ekstrak yang telah diencerkan pada panjang gelombang 290-320 nm. Serapan diukur pada setiap 5 nm dengan menggunakan spektrofotometer UV. Nilai SPF kemudian dihitung berdasarkan serapan yang terekam dengan menggunakan persamaan matematika yang dikembangkan oleh Mansur. Aktivitas antioksidan dari kulit buah rambutan ditentukan dengan menggunakan metode DPPH dan vitamin C sebagai pembanding. Nilai SPF dari kulit buah rambutan yang diukur pada konsentrasi ekstrak 50, 100, 150 dan 200 $\mu\text{g}/\text{mL}$ adalah $6,47 \pm 0,45$; $9,26 \pm 0,28$; $13,01 \pm 0,33$; dan $16,17 \pm 0,63$ secara berurutan. Aktivitas antioksidan dilakukan melalui penentuan nilai IC_{50} . Nilai IC_{50} dari kulit buah rambutan adalah $41,47 \pm 3,89 \mu\text{g}/\text{mL}$ sementara nilai IC_{50} Vitamin C adalah $24,87 \pm 0,69 \mu\text{g}/\text{mL}$. Berdasarkan hasil penelitian ini, ekstrak etanol kulit buah rambutan memiliki potensi sebagai tabir surya dan antioksidan.

Kata kunci : kulit buah rambutan, SPF, antioksidan, IC_{50}

Introduction

Rambutan is a tropical fruit belongs to family *Sapindaceae*. The origin of rambutan is uncertain, but the probable centre of origin are Indonesia and Malaysia (Lim 2013). In Indonesia, there are about twenty two varieties of rambutan derived from pure variety and grafting from two different varieties (Tindall 1994). Some varieties are cultivated for its economic values. Rambutan is mainly eaten fresh while some of them are canned in light syrup without the seed. The rind and the seed often remained as waste.

Rambutan rind are organic waste with many potential health benefits, such as antioxidant activity (Thitilertdecha *et al.* 2008, Palanisamy *et al.* 2008, Sun *et al.* 2011, Ling *et al.* 2010, Sun *et al.* 2012), antihyperglycemic activity (Palanisamy, 2011), antimicrobial activity (Zhao *et al.* 2011), anticancer activity (Wan. 2011), and antiobesity activity (Zhao *et al.* 2011 and Lestari, 2013). Rambutan rind contains flavonoids, tannins, ellagic acid and the major functional group of CH₃, aliphatic CH₃, and C=O compounds (Lestari, 2013). Palanisamy *et al.* (2011) have isolated geraniin, an ellagitannin as the major bioactive compound with strong antioxidant activity and showed antihyperglycemic activity. Sun *et al.* (2011) in another study have found that free phenolic compounds which was isolated from rambutan pericarp have higher antioxidant activity compare to soluble and insoluble-bound phenolic compounds.

Phenolic compounds has the ability to scavenge free radicals from UV exposure, thus it can be used as sunscreen (Svobovoda 2003). Some studies have shown that the photoprotective and antioxidant activity of natural plants were related to the phenolic compounds isolated from the plants (Fatmawati *et al.* 2014 dan Souza *et al.* 2014). This recent study aimed to determine the sun protective factor (SPF) and antioxidant activity of ethanolic extract of rambutan rind.

Materials and Methods

Plant material and preparation of extract

Rambutan fruit were collected in Palopo, South Sulawesi, Indonesia. Rambutan rind was

separated from its fruit, then washed out and cut into small pieces to prepare for the drying step.

The pieces were dried on straw paper and later grinded and sieved using 24 mesh. 200 g of powder of Rambutan rind powder (200 g) was macerated using ethanol for 5 x 24 hours. Every 24 hours, the solvent was replaced. The result of the consecutive maceration was filtered and evaporated using rotary evaporator at 50 °C to get concentrated ethanolic extract. Ethanolic extract 25 mg was weighed and transferred into 50 mL volumetric flask, then it was diluted by ethanol to make stock solution of 0.5 mg/ml for further assays.

In Vitro SPF Assay

Concentration series of 50, 100, 150, and 200 µg/mL, were prepared from aforementioned stock solution. The absorbance of each concentration was measured between 290-320 nm using 1 cm quartz cell at an interval of 5 nm using UV-Vis Spectrophotometer (Agilent 8453). Each concentration was replicated three times. The SPF value was calculated using Mansur equation (Mansur *et al.* 1986).

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

Where, CF = 10 (correction factor), EE(λ) = Erythemalogenic effect of radiation of wavelength λ, I(λ) = Intensity of solar radiation at a wavelength and Abs(λ) = Spectrophotometric absorbance at wavelength λ. The values of EE(λ) × I(λ) are constants as listed in Table 1 (Sayre *et al.* 1979).

Table 1 Normalized product function used in the calculation of SPF

Wavelength (nm)	EE × I (λ)
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0837
320	0.0180
Total	1

Antioxidant Activity Assay

The antioxidant activity was measured by determining the free radical scavenging activity of the extract using 2,2-diphenyl-1-picryl-hydrazyl (DPPH) radical (Mensor *et al.* 2001). The ability of the extract in scavenging the DPPH free radical was showed by the discoloration of the DPPH solution. Stock solution of 0.5 mg/mL of the extract were diluted in ethanol to final concentrations of 5, 10, 15, 20 and 25 µg/mL. DPPH solution were added to the diluted extract and allowed to react at room temperature for 30 minutes. Each concentration was replicated three times. The absorbance values were measured at 518 nm and converted into percentage of antioxidant activity (%AA) using the following formula:

$$\% \text{ AA} = \left(\frac{\text{absorbance control} - \text{absorbance sample}}{\text{absorbance control}} \right) \times 100$$

Absorbance control is the absorbance of DPPH solution in ethanol, ethanol was used as blank. IC₅₀ was calculated by linear regression using GraphPad Prism® 7.01 program. The IC₅₀ values represent the concentration of extract to scavenge 50% DPPH free radicals. The ability of vitamin C in scavenging the DPPH solution was measured as a positive control.

Results and Discussion

The result of the in vitro SPF assay was shown in Table 2, at the concentration of 50 µg/mL, the SPF value was more than 6. The SPF value of the rambutan rind ethanolic extract increased along with the increasing concentration of extract.

Table 2 SPF Value of Varies concentration of Rambutan rind ethanolic extract

Extract Concentration (ppm)	SPF Value (mean ± SD)
50	6.47 ± 0.45
100	9.26 ± 0.28
150	13.01 ± 0.33
200	16.17 ± 0.63

In vitro SPF Assay

Antioxidant activity was measured using DPPH. DPPH is a free radical because of the unpaired electron. With the presence of hydrogen donor, the unpaired electron of the radical becomes paired, thus decreasing the absorbance in the wavelength of 518 nm. The antioxidant activity of rambutan rind ethanolic extract was presented in Figure 1, in % AA. IC₅₀ represents the concentration in which the inhibition of free radical DPPH by 50% as shown in Table 3.

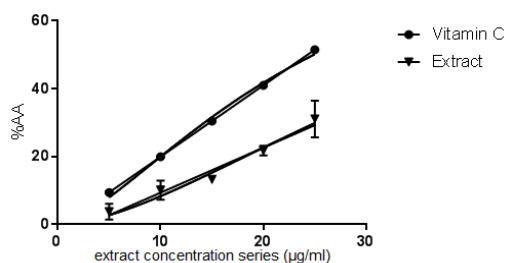


Figure 1 Percentage of Antioxidant activity (%AA) of Rambutan rind ethanolic extract

Table 3 IC₅₀ value of rambutan rind ethanolic extract and vitamin C as positive control

Sample	IC ₅₀ value
Vitamin C	24.87 ± 0.69
Rambutan Rind ethanolic Extract	41.47 ± 3.89

Rambutan is a sweet seasonal tropical fruit. The peel and the seed are organic waste of rambutan possessing many health benefits but have not used optimally. SPF value has become the international standard for quantitative measurement of sunscreen products. In cosmetics, SPF of 15 is mainly used to protect the skin from the UV radiation. This present study showed that rambutan rind ethanolic extract has the capacity as UV filter. At a concentration range of 50-200 µg/mL, the SPF value was ranging from 6 to 16. The capacity of natural SPF plants acts as sunscreen is related to its phenolic compounds. Previous studies have shown that the higher the phenolic compound the better value. Wungkana *et al.* (2013) showed that the phenolic fraction of waste corn cobs have high SPF value.

Another study by Martono *et al.* (2010) described that the phenolic content of black tea industrial waste related to its potent UV filter effectiveness. Thus in this research the SPF value of rambutan rind ethanolic extract could be related to its phenolic content, however future research are needed to confirm this statement.

The percentage of antioxidant activity of rambutan rind ethanolic extract at concentration of 25 µg/mL was around 31.11%. The IC₅₀ of rambutan rind ethanol extract was 41.47 ± 3.89 µg/mL which was about two folds higher than IC₅₀ value of vitamin C (24.87 ± 0.69 µg/mL). This data showed that the antioxidant activity of rambutan rind ethanolic extract was two folds lower than vitamin C. In another study, IC₅₀ of methanolic fraction of rambutan rind was 4.94 µg/mL (Thitilertdecha 2010). This value indicated that the methanolic fraction from the later study showed better antioxidant activity than the ethanolic extract. The location source of plant, extraction method and the solvent used could be different, resulting in different value of IC₅₀. Despite of that, this study was in line with each other in supporting the fact that rambutan rind is a potent antioxidant.

The main bioactive compounds of rambutan rind are phenolic compound, flavonoids and tannis. Phenolic compound and flavonoids have been related to the antioxidant activity of this extract.

Conclusion

Rambutan is a tropical fruit contained high in flavonoids and phenolic compounds. The ethanolic extract of rambutan rind showed both UV filter activity and antioxidant activity. Its activities possibly related to its bioactive components. Thus, further research in relating phenolic and flavonoid content to sunscreen activity is needed.

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