

Sunscreen Activity and Total Phenolic Content of Jengkol Leaves (*Pithecellobium lobatum* Benth.) Ethanolic Extract

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ABSTRACT

Exposure to UV rays can cause several skin problems, such as redness, sunburn, erythema, and ageing. The dangerous effects of UV radiation require more protection of the skin, such as sunscreen. Jengkol leaves (*Pithecellobium lobatum* Benth.) are a plant that contains phenolic compounds that can absorb UV radiation, so they can act as sunscreens. This study aimed to determine the activity of sunscreen using in vitro methods in the ethanol extract of *P. lobatum* leaves based on the sun protection factor (SPF), % transmission of erythema (% Te), % transmission of pigmentation (% Tp) and its total phenolic content by UV-vis spectrophotometry. The best sunscreen activity of the ethanolic extract of *P. lobatum* leaves was at a concentration of 400 ppm with an SPF value of 28.29 ± 0.034 (ultra), % Te and % Tp were included in the sunblock category. The total phenolic content of the ethanolic extract of the leaves of *P. lobatum* was 394.45 ± 6.40 mg of gallic acid equivalent per gram extract (mg GAE/g extract). The ethanolic extract of the *Pithecellobium lobatum* leaves has great potential as a sunscreen product.

INTRODUCTION

The effects of premature ageing can begin with the process of darkening the skin colour (melanogenesis) that occurs due to excessive production of melanin due to increased activity of the tyrosinase enzyme due to exposure to UV light. In addition, some UVA radiation is absorbed by the epidermis, 20-30% of which can reach the dermis of the skin. If it is continuously exposed to the skin, it will cause erythema, photocarcinogenicity, direct pigment darkening (IPD), and photoaging (Avianka et al., 2022). The intensity of excessive exposure to UV rays can increase the likelihood of skin cancer (Jati & Priyambodo, 2010). Ingredients that are sun protection factors are expected to prevent

melanogenesis by preventing enzyme oxidation and inhibiting exposure to excess radicals from sunlight (Gazali et al., 2014).

Jengkol leaves are widely used by the community to treat skin diseases (Bunawan et al., 2013). Previous research stated that *A. jiringa* leaves have very strong antioxidant activity (Alfisyahr, 2023). The jengkol plant (*Archidendron jiringa*, *A. pauciflorum*, *Pithecellobium jiringa*, *P. lobatum*) contains phenolic compounds, flavonoids, tannins, sterols, and alkaloids (Madiabu et al., 2023).

Phenolic compounds are one of the compounds that can function as a sunscreen. Antioxidant activity, sunscreen activity, and phenolic level have a significant correlation (Furi

et al., 2023). The study of jengkol sunscreen activity has never been reported. Therefore, the purpose of this study is to look at the potential activity of sunscreen and determine the total phenolic content of jengkol leaves.

RESEARCH METHODOLOGY

Chemical

The materials used for this study were 96% ethanol (technical grade), 96% ethanol (pro analysis grade, Smart-Lab), 5% FeCl₃, gallic acid (pro analysis grade by Sigma-Aldrich), Folin-Ciocalteu (pro analysis grade, Merck), Na₂CO₃, (technical grade), aquadest.

Extraction

The leaves of *P. lobatum* were determined at Laboratorium Dasar FMIPA ULM (284/LB.LABDASAR/XI/2023) and collected from Landasan Ulin Timur, Banjarbaru City, South Kalimantan on December 2023. The selected leaves were washed, then cut into smaller pieces, and dried in a dryer cabinet (LOKAL) at 50 °C (Fitriana et al., 2020; Helsawati et al., 2023). The dried leaves in small pieces, 26.25% (dark green with a characteristic smell of leaves and tasteless), were mashed with a blender (Miyako®) and then sifted with a 14 mesh sieve (Retsch). The simplicia (250 grams) was macerated with 2.5 L of 96% ethanol (1 : 10 w/v) for 3 x 24 hours (solvent replacement every 24 hours and stirring every 8 hours). The liquid extract collected was concentrated using a rotary evaporator (IKA® RV 10) and a water bath (Memmert®) at a temperature of 50 °C until a fixed weight was obtained (Ministry of Health of the Republic of Indonesia, 2017).

Sunscreen Activity Study

The ethanolic extract of *P. lobatum leaves* (1000 ppm) in 96% ethanol was taken to be diluted into solutions of 100, 200, 400, 600, and 800 ppm in three replications (I-III). The sample solution was measured at a wavelength of 290-320 nm to determine the SPF value (Hasanah et al., 2015; Normaidah et al., 2023), at a wavelength of 292.5-317.5 nm (transmission of erythema, % Te) and 322.5-372.5 nm (pigmentation transmission) with an interval of 5 nm, respectively (Hasanah et al., 2015).

The SPF value is calculated by the Mansur equation from the absorbed value obtained and incorporated into **equation 1**.

$$SPF=CF \times \sum_{290}^{320} EE \times I(\lambda) \times \text{abs}(\lambda) \dots\dots(1)$$

Information:

CF = correlation factor (10)
 EE x I = efficient erythema and Sunlight simulation spectrum (**Table 1**)
 Abs = absorbance

The %Te value was calculated from the erythema transmission value of each wavelength (292.5-317.5 nm) obtained and included in **equation 2**.

$$\% Te = \frac{Ee}{\sum Fe} = \frac{\sum(T \times Fe)}{\sum Fe} \dots\dots\dots(2)$$

Information:

Te = percent of erythema transmission value
 Fe = flux eritema (**Table 1**)
 Ee = The amount of erythema flux that the sunscreen carries on
 T = Transmission

The % Tp value was calculated from the erythema transmission value of each wavelength (322.5-372.5 nm) obtained and included in equation 3.

$$\% Tp = \frac{Ep}{\sum Fp} = \frac{\sum(T \times Fp)}{\sum Fp} \dots\dots\dots(3)$$

Information:

Tp = Pigmentation Transmission Percentage Value
 Fp = pigmentation flux (**Table 1**)
 Ep = The amount of pigmentation flux that the sunscreen carries on
 T = Transmission

Phenol-Phytochemical Screening

The extract was subjected to a phytochemical screening study to determine the presence of secondary metabolites, especially phenols. The extract (0.2 grams in 10 ml of 96% ethanol) was added 5% FeCl₃ about 5 drops (Ministry of Health of the Republic of Indonesia, 2017).

Determination of the Total Phenolic Content

Determination of Maximum Wavelength and Operating Time and Standard Curve Series

This study used gallic acid as a standard (Da'i et al., 2012; Rizki et al., 2022). A total of 500 µL (40 ppm) in 96% ethanol was added to 2.5 ml of 7.5% Folin-Ciocalteu reagent and allowed to stand for 8 minutes. After that, 2 mL of 7.5% Na₂CO₃ solution was added, homogeneously shaken, and left at room temperature in the operating time range, then the absorption was measured at a wavelength of 600-850 nm and the absorbance was measured at the maximum wavelength in the range of 0-60 minutes with a

time interval of 2 minutes until the absorption was stable using the UV-vis spectrophotometer (PerkinElmer UV/Vis Lambda 356). The determination of the standard curve was carried out with concentrations of 10, 20, 30, 40, 50 and 60 ppm gallic acid to obtain **equation 4** (Al-Amin, 2019; Ministry of Health of the Republic of Indonesia, 2017).

$$y = bx + a \text{ (4)}$$

Determination of the total phenolic content of the ethanol extract of *P. lobatum* leaves

The ethanolic extract of the leaves of *P. lobatum* (150 ppm) was treated the same as the

treatment in the standard curve solution and measured by UV-vis spectrophotometer in three replications. The total phenolic content has been calculated using **equation 5**.

$$\text{TPC} = \frac{C.V.fp}{g} \text{ (5)}$$

Information:

TPC = total phenolic content (mg EAG/g)

C = phenolic concentration (ppm).

V = sample volume (mL)

fp = dilution factor

g = sample weight (gram)

Table 1. Standard values in sunscreen study

| Wavelength (nm) | SPF | Wavelength (nm) | %Te | Wavelength (nm) | %Tp |
|-----------------|---------------|-----------------|---------------|-----------------|---------------|
| | EE XI | | Ee | | Fe |
| 290 | 0.0150 | 292.5 | 0.1105 | 322.5 | 0.1079 |
| 295 | 0.0817 | 297.5 | 0.672 | 327.5 | 0.102 |
| 300 | 0.2874 | 302.5 | 1 | 332.5 | 0.0936 |
| 305 | 0.3278 | 307.5 | 0.2008 | 337.5 | 0.0798 |
| 310 | 0.1864 | 312.5 | 0.1364 | 342.5 | 0.0669 |
| 315 | 0.0839 | 317.5 | 0.1125 | 347.5 | 0.057 |
| 320 | 0.0180 | | | 352.5 | 0.0448 |
| | | | | 357.5 | 0.0456 |
| | | | | 362.5 | 0.0356 |
| | | | | 367.5 | 0.031 |
| | | | | 372.5 | 0.026 |
| Total | 1.0000 | Total | 2.2322 | Total | 0.6902 |

RESULT AND DISCUSSION

The ethanolic extract of the *P. lobatum* leaves obtained a percentage yield of 20.08%. (50.19 grams) with the blackish-green colour, thick shape, had a distinctive odour, and taste very bitter. This result was greater than the Alfisyahr (2023), which was 14.43%. The obtained extraction yield was greater because there was a difference in the ratio of the simplicia with the solvents for maceration. The Alfisyahr (2023) study was 1:4 (w/v), while in this study the ratio was 1:10 (w/v). The ratio affected the obtained yield because the larger the ratio of solvents to samples made the higher concentration difference, the more optimally the discharge of the target compound into the solvent, so the yield increased (Noviyanty et al., 2019; Senduk et al., 2020; Wathan et al., 2024).

The sunscreen activity of this study were divided into 3 value, SPF value, %Te and %Tp. The sunscreen activity of this extract has never

been reported before. The SPF value can be seen in **Table 2**. The study is in line with the research of Risman et al. (2022), who stated that the higher concentration given the higher SPF value. SPF (Sun Protecting Factor) is a comparison between the amount of energy needed to cause minimal erythema on skin protected by sunscreen and the amount of energy needed to cause the same effect on skin not protected by UVB sunscreen (Isfardiyana & Safitri, 2014).

The %Te and %Tp can be seen in **Table 3** and **Table 4** respectively. The value of the percentage of transmission of erythema (% Te) describes the ability of a sunscreen to transmit UVB that can cause erythema. The value of the percentage of transmission of pigmentation (% Tp) describes the ability of a sunscreen to transmit UVA that can cause pigmentation or skin discolouration to become darker. The lower the transmission value, the less UV light is passed to the skin (Hasanah et al., 2015).

Table 2. The SPF value of *P. lobatum* ethanolic extract of *P. lobatum* leaves

| Concentration (ppm) | SPF value | | | Average | SD | RSD (%) | Category* |
|---------------------|-----------|-------|-------|---------|-------|---------|-----------|
| | I | II | III | | | | |
| 60 | 3.26 | 3.27 | 3.27 | 3.27 | 0.006 | 0.18 | Minimum |
| 80 | 5.46 | 5.45 | 5.44 | 5.45 | 0.010 | 0.18 | Moderate |
| 100 | 6.38 | 6.44 | 6.44 | 6.42 | 0.035 | 0.54 | Extra |
| 200 | 12.73 | 12.72 | 12.77 | 12.74 | 0.026 | 0.21 | Maximum |
| 400 | 28.31 | 28.25 | 28.31 | 28.29 | 0.035 | 0.12 | Ultra |

*(Hasanah et al., 2015; Kasitowati et al., 2021)

Table 3. The % Te of *P. lobatum* ethanolic extract of *P. lobatum* leaves

| Concentration (ppm) | %Te | | | Average | SD | RSD (%) | Category* |
|---------------------|-------|-------|-------|---------|--------|---------|------------------|
| | I | II | III | | | | |
| 60 | 44.35 | 44.24 | 44.23 | 44.27 | 0.069 | 0.15 | - |
| 80 | 26.29 | 26.47 | 26.48 | 26.41 | 0.1088 | 0.41 | - |
| 100 | 21.37 | 21.03 | 20.97 | 21.13 | 0.220 | 1.04 | - |
| 200 | 5.75 | 5.72 | 5.56 | 5.68 | 0.098 | 1.72 | Extra Protection |
| 400 | 0.65 | 0.65 | 0.64 | 0.64 | 0.007 | 1.13 | Sunblock |

*(Hasanah et al., 2015; Kasitowati et al., 2021)

Table 4. The % Tp of *P. lobatum* ethanolic extract of *P. lobatum* leaves

| Concentration (ppm) | %Tp | | | Average | SD | RSD (%) | Category* |
|---------------------|-------|-------|-------|---------|-------|---------|------------------|
| | I | II | III | | | | |
| 60 | 81.90 | 82.21 | 82.19 | 82.10 | 0.175 | 0.21 | Fast tanning |
| 80 | 74.38 | 74.94 | 74.96 | 74.76 | 0.332 | 0.44 | Fast tanning |
| 100 | 69.67 | 69.61 | 70.69 | 69.99 | 0.609 | 0.87 | Fast tanning |
| 200 | 50.53 | 50.55 | 49.29 | 50.13 | 0.722 | 1.44 | Extra Protection |
| 400 | 23.64 | 23.70 | 22.95 | 23.43 | 0.416 | 1.77 | Sunblock |

*(Hasanah et al., 2015; Kasitowati et al., 2021)

In this investigation by phytochemical screening, the ethanolic extract of *P. lobatum* contained phenolic. The total phenolic content (TPC) was measured with the standard gallic acid calibration curve. The TPC was analysed at 732.5 nm in operation time 54-56 minutes. This is related to previous research on the determination of TPC (Gultom et al., 2021; Rivai et al., 2019). The concentration series (10-60 ppm) had given absorbance between 0.207-0.977 with $y=0.0152x + 0.056$ ($r = 0.9998$) (Figure 1). The data show that this sample had a total phenolic content of 39.445 ± 0.64 % b/b EAG or equal to 394.45 ± 6.40 mg/ EAG (Table 5). This TPC was greater than Al-Amin (2019).

The study showed that the TPC of extract was 340,703 mg of EAG / gram of extract. The results of this difference are probably due to different treatments in the samples and in the collecting place. Al-Amin (2019) determined the total phenolic content using Na_2CO_3 7%, while in this study Folin-Ciocalteu of 7.5% and Na_2CO_3 of 7.5%, but the collection site was not reported.

From the data, this ethanolic extract with 400 ppm has a great potential to develop as a sunscreen product. The presence of conjugated double bonds in phenolic compounds is specifically capable of absorbing UV rays and will reduce their intensity (Furi et al., 2023).

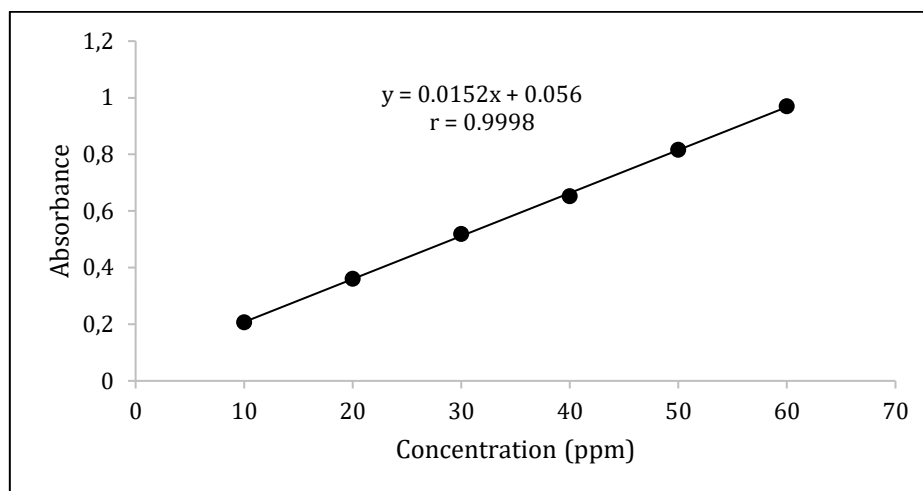


Figure 1. Calibration curve of gallic acid standard

Table 5. Total phenolic content of ethanol extract of *P. lobatum* leaves

| Absorbance | Concentration ($\mu\text{g/mL}$) | TPC ($\mu\text{g EAG/mg extract}$) | TPC (%b/b EAG) | \bar{x} TPC (%b/b EAG) | SD | RSD (%) |
|------------|---------------------------------------|---|-------------------|-----------------------------|------|------------|
| 0.949 | 58.750 | 391.672 | 39.167 | | | |
| 0.972 | 60.263 | 401.759 | 40.176 | 39.445 | 0.64 | 1.62 |
| 0.945 | 58.487 | 389.917 | 38.992 | | | |

CONCLUSIONS

The best sunscreen activity is at a concentration of 400 ppm with an SPF value of 28.29 ± 0.034 (ultra), % Te and % Tp are included in the sunblock category with the total phenolic content of 394.45 ± 6.40 mg of GAE / g of extract. The *Pithecellobium lobatum* leaves ethanolic extract has great potential as a sunscreen product.

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AUTHORS' CONTRIBUTIONS

Normaidah contributed as the head of the research team, contributed to data collection guidance and writing manuscript, Mia Fitriana contributed to proofreading, Fadlilaturrahmah contributed to data collection guidance, Winda Tri Kurniasari contributed to data collection, Hayatun Izma contributed to script preparation, Prima Happy Ratnapuri contributed to review data analysis.

CONFLICT OF INTERESTS

All authors in this manuscript declared that there was no conflict of interest in this study.

ETHICAL CONSIDERATION

Ethical issues (including plagiarism, data fabrication, double publication, etc) have been completely observed by the authors.

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