Phytochemical profile, antioxidant activity, and sun protection potential of selected Annonaceae species

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ABSTRACT: Excessive sun exposure can damage the skin, highlighting the need for effective natural sunscreens. This study aimed to evaluate the phytochemical profile, antioxidant activity, total phenolic content (TPC), total flavonoid content (TFC), and sun protection factor (SPF) of five Annonaceae species. Methanolic leaf extracts of *Annona muricata* (soursop), *Annona squamosa* (sugar apple), *Cananga odorata* (cananga), *Annona reticulata* (nona), and *Monoon longifolium* (glodokan tiang) were analyzed. Phytochemical screening was performed to identify secondary metabolites. Antioxidant activity was determined using the DPPH radical scavenging assay, while TPC and TFC were measured spectrophotometrically. SPF values were calculated using the Mansur method. Phytochemical screening confirmed the presence of flavonoids, saponins, tannins, steroids, and phenolics in all extracts, with cananga leaves additionally containing alkaloids, coumarins, and essential oils. The DPPH assay showed antioxidant activity with IC₅₀ values ranging from 12.89–228.98 ppm, classified from very strong to weak. TPC and TFC values ranged from 12.33–31.75 mg GAE/g extract and 8.83–28.19 mg QE/g extract, respectively. SPF values ranged from 19.32–35.03, indicating all extracts fall under the ultra-protection category, with soursop leaf extract exhibiting the highest SPF. This study highlights the potential of Annonaceae leaf extracts, particularly *A. muricata*, as promising natural antioxidant and photoprotective agents due to their high polyphenolic and flavonoid content.

KEYWORDS: Annonaceae; antioxidant; DPPH; flavonoid; phenolic; SPF.

INTRODUCTION

Sunlight is a vital natural energy source essential for sustaining life on Earth. It supports photosynthesis and vitamin D synthesis, which are crucial for bone metabolism and immune function. However, excessive exposure to ultraviolet (UV) radiation—particularly UV A (320–400 nm) and UV B (290–320 nm)—can lead to oxidative stress, premature aging, loss of skin elasticity, and even skin cancer [1]. These effects arise from the overproduction of reactive oxygen species (ROS), which can damage lipids, proteins, and DNA, ultimately disrupting cellular homeostasis [2].

Antioxidants play a key role in counteracting oxidative stress by donating electrons to neutralize ROS, thus inhibiting chain reactions that cause molecular damage. Plant-derived secondary metabolites such as flavonoids, phenolics, alkaloids, terpenoids, and steroids have been extensively reported to exhibit strong antioxidant and photoprotective activities [3]. Among these, phenolic compounds—especially flavonoids—are recognized for their ability to absorb UV radiation due to their conjugated double bonds and chromophore structures, which reduce UV penetration and protect the skin from oxidative damage [4].

Members of the Annonaceae family, including *Annona muricata* (soursop), *Annona squamosa* (sugar apple), *Cananga odorata* (cananga), *Annona reticulata* (nona), and *Monoon longifolium* (glodokan tiang), are rich in bioactive compounds such as acetogenins, flavonoids, alkaloids, and phenolic derivatives [5], [6]. Several studies have reported the antioxidant and anti-inflammatory potential of *A. muricata* and *A. squamosa* [7], [8], while *C. odorata* and *M. longifolium* exhibit antimicrobial and UV-protective activities [9], [10].

Despite the pharmacological potential of the Annonaceae family, comparative studies examining their antioxidant strength, total phenolic and flavonoid content, and sunscreen potential remain scarce. Furthermore, the correlation between phytochemical composition and sun protection factor (SPF) among Annonaceae species has not been systematically established.

How to cite this article: Djamil R, Iskandar MC, Pratami DK. Phytochemical profile, antioxidant activity, and sun protection potential of selected Annonaceae species. JIFI. 2025; 23(2): 373-381.

© 2025 Universitas Pancasila Press ISSN: 2614-6495 https://doi.org/10.35814/jifi.v23i2.1671

Therefore, this study aimed to evaluate and compare the phytochemical profile, antioxidant activity, total phenolic content (TPC), total flavonoid content (TFC), and SPF values of methanol leaf extracts from five Annonaceae species. The species with the highest SPF value was further assessed for extract quality parameters. This study addresses a clear research gap by linking antioxidant capacity with photoprotective potential across Annonaceae species, supporting their utilization as natural sunscreen ingredients.

MATERIALS AND METHODS

Materials

The material used for this research were dried leaves of soursop (*Annona muricata* L.), sugar apple (*Annona squamosa* L.), cananga (*Cananga odorata* (Lam.) Hook.f & Thomson), nona (*Annona reticulata* L.), and glodokan tiang (*Monoon longifolium* (Sonn.) B.Xue & R.M.K.Saunders obtained from the Indonesian Spice and Medicinal Research Institute (BALITTRO), Bogor, Indonesia. All solvents and reagents used were of analytical or proanalysis grade (Merck, Darmstadt, Germany).

Methods

Plant determination

The plant samples were authenticated by a botanist, and voucher specimens were deposited in the laboratory collection at the Herbarium Depokensis (UIDEP), Biota Collection Room, Universitas Indonesia.

Extraction

Each 250 g of powdered leaf sample was subjected to kinetic maceration using methanol (Merck, ≥99.8% purity) for 72 hours at room temperature with periodic stirring. The filtrate was concentrated using a rotary evaporator (Heidolph Laborota 4000) under reduced pressure at 40 °C until a thick extract was obtained. Extracts were stored in airtight containers at 4 °C until further analysis [11].

Phytochemical screening

Preliminary phytochemical screening was carried out to identify the presence of alkaloids, flavonoids, saponins, tannins, coumarins, quinones, steroids/terpenoids, essential oils, and phenolics following previous studies [11].

Antioxidant activity assay

Antioxidant activity was determined using using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging assay. A total of 1 mL of methanol extract solution (50, 100, 150, 200, and 250 μ L) was added to 1 mL of 0.4 mM DPPH solution and pro-analysis methanol up to the 5 mL mark. The solution was homogenized and the mouth of the tube was covered with aluminum foil. Absorbance was measured using a UV-Vis spectrophotometer (Shimadzu UV-1800, Japan) at 517 nm. Methanolic extract solutions (50–250 μ g/mL) were reacted with 0.4 mM DPPH solution in methanol and incubated in the dark for 30 minutes. Vitamin C served as the reference standard. Each concentration was analyzed in triplicate. The percentage of inhibition was calculated using the following formula:

% inhibition =
$$\frac{Ablank - Asample}{Ablank} \times 100\%$$

The IC_{50} value was determined using linear regression from the plot of % inhibition versus concentration, within the equation y = a + bx, where y is 50 and x is the IC_{50} value [12].

Total flavonoid and phenolic content determination

Total flavonoid content (TFC) was quantified using the aluminum chloride colorimetric method, while total phenolic content (TPC) was determined using the Folin–Ciocalteu assay [12]. Absorbance was measured at 425 nm and 706 nm, respectively, using a Shimadzu UV-1800 spectrophotometer. Quercetin and gallic acid were used as standards. Each test was replicated three times, and results were expressed as mg QE/g extract and mg GAE/g extract.

Sunscreen activity assay

Sunscreen activity was evaluated using the in vitro spectrophotometric method at wavelengths of 290–320 nm with 5 nm intervals using a UV-Vis spectrophotometer (Shimadzu UV-1800). Measurements were

conducted in a 1 cm quartz cuvette with triplicate readings. SPF values were calculated according to the Mansur equation using the erythemal effect spectrum (EE × I) constants [13]:

SPF=CF
$$x \sum (EE \times I \times Abs)$$

CF = Correction factor (=10)

EE = Spectrum of erythema effects

I = Light intensity spectrum

Abs = Absorbance of sunscreen samples

The EE x 1 value is constant and is shown in Table 1 below:

Table 1. Wavelength and EE x I.

No.	Wavelength (λ nm)	EE x I
1.	290	0.0150
2.	295	0.0817
3.	300	0.2874
4.	305	0.3278
5.	310	0.1864
6.	315	0.0839
7.	320	0.0180
· · · · · · · · · · · · · · · · · · ·	Total	1

The calculation method for determining the SPF value was carried out by multiplying the EE \times I value for each wavelength listed in Table 1 with the corresponding absorbance value obtained from the sample. The resulting products were then summed to obtain the total value. This total was subsequently multiplied by a correction factor (CF = 10) to determine the final SPF value of the extract.

Specific and non-specific quality parameters

Specific parameters included organoleptic analysis (color, odor, consistency) and determination of ethanol- and water-soluble extractive values [14]. Non-specific parameters included loss on drying, water content, total ash, acid-insoluble ash, and residual solvents. Water content was determined using the Karl Fischer titrator (Metrohm 870 KF Titrino Plus). Total ash and acid-insoluble ash were measured using a muffle furnace (Thermo Fisher Thermolyne) at 450 °C. Residual solvents were analyzed via gas-liquid chromatography (Shimadzu GC-2014, Japan).

Heavy metal contamination test

Lead (Pb) and cadmium (Cd) levels were determined using atomic absorption spectrophotometry (PerkinElmer AAnalyst 400, United States) [12]. Samples were digested with nitric acid and analyzed at specific wavelengths (Pb: 217.0 nm; Cd: 228.8 nm). Results were expressed in mg/kg and compared with WHO limits for herbal products.

Microbial contamination test

Microbial load (Total Plate Count and Yeast/Mold Count) was assessed according to WHO guidelines [12]. Serial dilutions were prepared using phosphate buffer (pH 7.2). Inoculation was performed on nutrient agar and potato dextrose agar plates, incubated at 35–37 °C for 48 hours in an inverted position. Colonies were counted using a digital colony counter (Stuart SC6PLUS) and expressed as CFU/g.

Statistical analysis

Data were analyzed using SPSS (IBM Corp., Armonk, NY, USA). Normality and homogeneity were verified using the Shapiro-Wilk and Levene's tests (Sig. > 0.05). Differences among extracts were determined using One-Way ANOVA, with Sig. <0.05 considered significant. Tukey's HSD test identified specific group differences. The data were expressed as mean ± standard deviation (SD), n=3.

RESULT

Extraction yields

The extraction of the five Annonaceae species was performed using the kinetic maceration method to prevent degradation of heat-sensitive compounds. Each powdered simplicia (250 g) was macerated twice (remaceration) with methanol solvent at room temperature, followed by filtration and solvent removal using a

rotary vacuum evaporator to obtain a concentrated extract. The extraction yield and Drug Extract Ratio (DERnative) were then calculated for each sample. The yields obtained were 12.89% for soursop leaves, 21.68% for sugar apple leaves, 21.96% for cananga leaves, 10.72% for nona leaves, and 13.61% for glodokan tiang leaves. The DER-native values were 7.76 for soursop leaves, 4.61 for sugar apple leaves, 4.55 for cananga leaves, 9.33 for nona leaves, and 7.35 for glodokan tiang leaves.

Phytochemical screening content

The results are presented in Table 2. Both the powdered and methanol extracts of the five species were found to contain flavonoids, saponins, tannins, steroids/triterpenoids, and phenolic compounds. However, Cananga odorata leaves also showed positive results for alkaloids, coumarins, and essential oils. These findings indicate that each plant possesses diverse bioactive constituents that may contribute to their pharmacological properties.

Table 2. Phytochemical screening results.

Phytochemical screening	A	В	С	D	E
Alkaloid	-	-	+	-	-
Flavonoid	+	+	+	+	+
Saponin	+	+	+	+	+
Tannin	+	+	+	+	+
Quinone	-	-	-	-	-
Coumarin	-	-	+	-	-
Steroid/triterpenoid	+/-	+/-	+/-	+/-	+/-
Essential oils	-	-	+	-	-
Phenolic	+	+	+	+	+

Note: A: Annona muricata (Soursop leaves); B: Annona squamosa (Sugar apple leaves); C: Cananga odorata (Cananga leaves); D: Annona reticulata (Nona leaves); E: Monoon longifolium (Glodokan tiang leaves); (+): Contains secondary metabolites; (-): Does not contain secondary metabolites

Antioxidant activity value

The lower IC₅₀ value proves increased antioxidant activity. The results of measuring antioxidant activity can be seen in Table 3.

Table 3. The results of antioxidant activity.

Extract	IC ₅₀ value (ppm)	Category
Vitamin C	5.58±1.92	Very strong
Annona muricata (Soursop) leaves	46.83 ± 1.94	Strong
Annona squamosa (Sugar apple) leaves	228.98 ± 1.46	Weak
Cananga odorata (Cananga) leaves	140.12 <u>±</u> 2.41	Moderate
Annona reticulata (Nona) leaves	12.89±3.93	Very strong
Monoon longifolium (Glodokan tiang) leaves	22.52±3.38	Very strong

Data on antioxidant activity values were analyzed using the SPSS program. Based on the results of the normality test (Shapiro-Wilk), the IC_{50} value data for the methanol extract of soursop leaves, sugar apple and glodokan tiang is normally distributed with a Sig value. >0.05. Based on Levene's test, the data is homogeneous with a Sig value. >0.05. The IC_{50} value data from the One Way Anova test obtained a Sig value. <0.05 which indicates that there is a significant difference in the IC_{50} value of the five Annonaceae plants. It can be concluded that soursop, sugar apple, cananga, nona, and glodokan tiang leaves extracts have different IC_{50} values when compared to each other.

Total flavonoid and phenolics content

The standard calibration curve for quercetin yielded a linear regression equation of y = 0.0026x - 0.0019, demonstrating good linearity. The total flavonoid contents of each extract are presented in Table 4, showing that Glodokan tiang leaves contained the highest flavonoid concentration (28.19±1.29 mg QE/g extract), followed by Sugar apple (27.62±0.91 mg QE/g extract) and Nona leaves (21.40±1.03 mg QE/g extract).

The calibration curve for gallic acid produced a linear regression equation of y = 0.0027x+0.2969, indicating good linearity. The results presented in Table 4 show that Nona leaves had the highest total phenolic

content (31.75±0.94 mg GAE/g extract), followed by Soursop (23.70±0.40 mg GAE/g extract) and Cananga leaves (21.67±0.13 mg GAE/g extract).

Table 4. The results of total flavonoid and phenolic content.

Leaves extract	Total flavonoid content	Total phenolic content	
	(mg QE/g extract)	(mg GAE/g extract)	
Annona muricata (Soursop)	20.20±0.28	23.70±0.40	
Annona squamosa (Sugar apple)	27.62±0.91	12.33±0.37	
Cananga odorata (Cananga)	8.83±0.43	21.67±0.13	
Annona reticulata (Nona)	21.40 ± 1.03	31.75±0.94	
Monoon longifolium (Glodokan tiang)	28.19±1.29	16.89 <u>±</u> 0.76	

Sunscreen protective value

The photoprotective activity of the extracts was expressed through the SPF (Sun Protection Factor) value, as shown in Table 5. The SPF values increased proportionally with concentration for all extracts. Among the five Annonaceae species, soursop leaves showed the highest SPF value (35.03 ± 1.81 at 1000 ppm), followed by sugar apple, nona, cananga, and glodokan tiang leaves. Statistical analysis (One Way ANOVA, p<0.05) confirmed significant differences in SPF among the extracts.

Table 5. The results of sunscreen activity test.

Leaves extract	SPF in concentration				
Leaves extract	200	400	600	800	1000
Annona muricata (Soursop)	8.07±0.97a	12.86±0.92b	19.59±1.05c	30.67±1.12°	35.03±1.81c
Annona squamosa (Sugar apple)	5.95 ± 1.50	11.73 ± 1.35	14.42±1.45	19.42±1.98	25.05 ± 1.41
Cananga odorata (Cananga)	4.26 ± 0.36	8.12 <u>±</u> 0.88	12.35 ± 0.59	15.83 ± 0.67	19.62±1.03
Annona reticulata (Nona)	4.95 ± 0.34	10.20 ± 1.00	15.46 ± 0.98	20.13 ± 1.70	28.20 ± 1.38
Monoon longifolium (Glodokan tiang)	4.08 ± 0.53	7.85±0.94	12.67±1.23	15.94 ± 1.37	19.32±1.18

Note:

- a: The SPF value of soursop leaves is significantly different from that of cananga nona, glodokan tiang leaves
- b: The SPF value of soursop leaves is significantly different from that of cananga leaves, glodokan tiang.
- $c: The \ SPF \ value \ of sours op \ leaves \ is \ significantly \ different \ from \ that \ of \ sugar \ apple, \ cananga, \ nona, \ glodokan \ tiang \ leaves$

Table 3 shows that Nona leaf extract has the highest antioxidant activity. A lower IC_{50} value indicates a higher antioxidant activity capacity. This is in line with the high amount of flavonoids and total phenolics in Nona leaf extract. Then the order of highest antioxidant activity after nona leaves is glodokan tiang leaves, soursop leaves, sugar apple leaves, and cananga leaves. Based on this, it can be concluded that the antioxidant activity of the five Annonaceae plants varies from very strong to weak.

The highest total flavonoid content was possessed by glodokan mast leaf extract, followed by sugar apple leaves, nona leaves, soursop leaves, and cananga leaves. The total flavonoid levels measured did not indicate the type of flavonoid compounds in the extract. The type of compound and the amount of content play a role in determining the amount of antioxidant activity and photoprotective ability.

Nona leaf extract had the highest total phenolics, followed by soursop leaves, cananga leaves, glodokan mast leaves, and sugar apple leaves. Just like when determining flavonoid levels, the total phenolic levels measured do not indicate the type of phenolic compounds in the extract. The type of compound and the amount of content play a role in determining the amount of antioxidant activity and photoprotective ability. The results also shows that the highest SPF value was obtained by soursop leaf extract when compared with the other four extracts.

Specific and non-spesific quality parameter

The methanol extract of *Annona muricata* (soursop) leaves was a thick, blackish-green extract. Determination of soluble compounds showed that water-soluble extractives (41.07±3.05%) were higher than ethanol-soluble extractives (33.60±4.40%), as shown in Table 6. According to the Monograph of Indonesian Medicinal Plant Extracts (MMI), both values meet the required standards (≥19.5% for water and ≥14.5% for ethanol).

Table 6. The result of soluble compound levels of soursop leaf methanol extract.

Solvent	Results (%)	MMI Requirement
Water	41.07±3.05	≥ 19.5
Ethanol	33.60 ± 4.40	≥ 14.5

The results of non-specific parameters are summarized in Table 7. The extract met all requirements specified by the Indonesian Herbal Pharmacopoeia and MMI standards, including drying shrinkage (8.68±0.22%), total ash (2.61±0.16%), acid-insoluble ash (0.39±0.35%), water-soluble ash (2.10±0.19%), and residual solvent (0.74%).

Table 7. The results of checking non-specific quality parameters.

Parameter	Results (%)	MMI Requirement
Loss on drying	8.68±0.22	≤ 10
Total ash	2.61±0.16	≤ 4.5
Acid-insoluble ash	0.39 <u>±</u> 0.35	≤0.7
Water-soluble ash	2.10±0.19	-
Residual solvent	0.74	≤1.0%

Heavy metal contamination value

The heavy metal test using Atomic Absorption Spectrophotometry (AAS) showed that lead (Pb) was present at 0.34 mg/kg, while cadmium (Cd) was not detected. Both results are within the acceptable limits of $\leq 10 \text{ mg/kg}$ for Pb and $\leq 0.3 \text{ mg/kg}$ for Cd, as defined in the MMI standard (Table 10). The results of heavy metal contamination test is shown in Table 8.

Table 8. Results of soursop extract heavy metal test.

Heavy metal	Results (ppm)	Maximum limit
Lead (Pb)	0.34	≤10 mg/kg
Cadmium (Cd)	Not detected	≤0.3 mg/kg

Microbial contamination results

The results of the examination of microbial contamination in the methanol extract of soursop leaves showed that the total plate number and mold number were less than 10 colonies/g. The maximum limit is based on the requirements of the Indonesian Medicinal Plant Extract Monograph for total plate number and mold number, namely no more than 10^3 colonies/g and no more than 10^5 colonies/g. This shows that the extract can be stored in the long term. The results were shown in table 9.

Table 9. Results of soursop extract microbial contamination test.

Type of test	Results (colony/g)	Maximum limit (colony/g)
Total plate count	<10	≤10 ³
Total yeast and mold count	<10	≤10 ⁵

DISCUSSION

The kinetic maceration method was chosen to ensure optimal extraction efficiency while preserving thermolabile phytoconstituents such as flavonoids and phenolic compounds, which are known to contribute to antioxidant and photoprotective activities [11]. The variation in extraction yields among the five Annonaceae species indicates differences in solvent-compound affinity and the phytochemical composition of each leaf. Cananga and sugar apple leaves produced the highest yields (above 20%), suggesting a higher concentration of methanol-soluble secondary metabolites, while nona leaves exhibited the lowest yield (10.72%), possibly due to a lower content of extractable compounds or stronger matrix-compound binding [11].

The DER-native values showed an inverse relationship with extract yield; lower DER values indicate more efficient extraction. The obtained DER range (4.55–9.33) is within acceptable limits for plant extracts, suggesting that the extraction process was effective and reproducible [11]. These results confirm that kinetic maceration using methanol is a suitable and reliable method for extracting bioactive constituents from Annonaceae leaves for further phytochemical and bioactivity analyses [11].

The phytochemical analysis revealed that all tested species contained flavonoids, saponins, tannins, steroids/triterpenoids, and phenolic compounds, while Cananga odorata uniquely contained alkaloids, coumarins, and essential oils. These results suggest that the Annonaceae species studied possess diverse secondary metabolites, which may be responsible for their antioxidant and photoprotective activities [12].

Flavonoids and phenolic compounds are known to act as free radical scavengers by donating hydrogen atoms or electrons to neutralize reactive oxygen species (ROS), thereby reducing oxidative stress caused by ultraviolet (UV) radiation. The presence of saponins and tannins also contributes to anti-inflammatory and skin-protective effects, which support their potential use in natural sunscreen formulations. Moreover, the presence of essential oils and coumarins in *Cananga odorata* may enhance the aromatic and pharmacological potential of the extract. These findings are consistent with previous studies indicating that members of the Annonaceae family contain active compounds with strong antioxidant and UV-absorbing properties, making them promising candidates for development as natural photoprotective agents [5].

The DPPH assay results indicate that all extracts exhibited varying degrees of free radical scavenging activity, with Annona reticulata (nona leaves) showing the highest antioxidant activity (IC₅₀ = 12.89 μ g/mL), followed by Monoon longifolium, Annona muricata, Cananga odorata, and Annona squamosa. These findings suggest that the antioxidant potential among the tested species is influenced by their distinct phytochemical compositions.

The ability of these extracts to neutralize DPPH radicals is mainly attributed to the presence of phenolic and flavonoid compounds, which are capable of donating hydrogen atoms or electrons to stabilize free radicals [2]. The hydroxyl groups attached to the aromatic rings of phenolic compounds enhance their electron-donating capacity, leading to increased antioxidant activity. Furthermore, flavonoids with a hydroxyl group at the C-3 position and a carbonyl group at the C-4 position can act as effective radical scavengers, thus contributing significantly to antioxidant potency.

Statistical analysis using SPSS software showed that the IC₅₀ data for soursop, sugar apple, and glodokan tiang extracts were normally distributed according to the Shapiro–Wilk test (Sig.>0.05), and homogeneous according to Levene's test (Sig.>0.05). The One-Way ANOVA test yielded a Sig.<0.05, indicating a statistically significant difference among the IC₅₀ values of the five Annonaceae species. Therefore, it can be concluded that each extract exhibits distinct antioxidant activities, reflecting the diversity of their secondary metabolite content.

These results are consistent with previous studies reporting strong correlations between phenolic and flavonoid content and antioxidant potential in Annonaceae species [6]. The high antioxidant activity observed in *A. reticulata* and *M. longifolium* highlights their potential as promising natural sources for the development of antioxidant-based therapeutic and cosmetic formulations.

The variations in flavonoid and phenolic contents among the five Annonaceae and Cananga extracts suggest differences in their metabolic profiles and phytochemical composition. Flavonoids and phenolics play crucial roles as antioxidants due to their hydrogen-donating ability and electron transfer potential, which can neutralize reactive oxygen species (ROS) and stabilize free radicals [4].

Higher total flavonoid content in Glodokan tiang and Sugar apple extracts may contribute to their strong radical scavenging activity, consistent with their low IC₅₀ values observed in antioxidant assays. Meanwhile, Nona leaves, which showed the highest total phenolic content, demonstrated excellent antioxidant potential, supporting previous findings that phenolic compounds are key contributors to antioxidant capacity [14].

Environmental factors such as light exposure, temperature, and soil nutrients may also influence the biosynthesis of phenolics and flavonoids in plants [15]. These results align with recent studies reporting that variations in total flavonoid and phenolic content significantly affect antioxidant and photoprotective properties in tropical medicinal plants [16].

The antioxidant capacity of plant extracts is generally associated with the presence of flavonoid and phenolic compounds, which act as hydrogen or electron donors to neutralize free radicals [3]. Theoretically, higher total flavonoid or phenolic content tends to correlate with stronger antioxidant activity because these compounds can stabilize reactive oxygen species through delocalization in their aromatic rings [14].

However, this correlation is not always linear. The type, position of hydroxyl groups, and interaction between compounds can alter activity. For example, in this study, sugar apple (*Annona squamosa*) leaves

contained relatively high flavonoids but exhibited moderate antioxidant and SPF values. This suggests that other factors—such as compound structure, solubility, and synergistic or antagonistic effects among metabolites—influence overall bioactivity [14].

Similarly, the SPF value did not always parallel antioxidant activity [17]. Although soursop leaves had the highest SPF, nona leaves showed the strongest antioxidant activity. This indicates that photoprotective ability depends not only on antioxidants but also on compounds that directly absorb UV radiation or enhance skin protection mechanisms. Overall, the results demonstrate that while flavonoid and phenolic contents are important contributors to antioxidant and photoprotective properties, the relationship among them is complex and influenced by the chemical diversity and interaction of secondary metabolites present in each extract.

The higher solubility of soursop leaf extract in water compared to ethanol indicates that its dominant secondary metabolites are polar compounds, such as flavonoids, tannins, saponins, and phenolics [12]. These compounds tend to dissolve more efficiently in polar solvents, which supports the choice of methanol and water-based extraction systems in previous antioxidant and photoprotective studies [16].

Non-specific quality parameters such as loss on drying, water content, and ash values provide essential information on extract purity and stability. The low moisture content (7.4%) and acceptable loss on drying suggest that the extract is chemically stable and less susceptible to microbial growth during storage. Similarly, the ash content values indicate minimal inorganic and environmental contamination, consistent with herbal quality standards [12], [18].

The absence of cadmium and the very low level of lead (0.34 mg/kg) confirm that the extract is safe from heavy metal contamination, an important criterion for materials intended for medicinal or cosmetic use. Microbial counts well below regulatory thresholds further demonstrate good handling and storage conditions during processing. Overall, the physicochemical and microbiological characteristics of the soursop leaf methanol extract comply with Indonesian Herbal Pharmacopoeia and MMI quality parameters, ensuring its suitability for further phytochemical, antioxidant, and photoprotective evaluations.

CONCLUSION

The extracts of soursop, sugar apple, cananga, nona, and glodokan malang leaves contained flavonoids, saponins, tannins, steroids, and phenolics, with cananga leaves also showing alkaloids, coumarin, and essential oils. Antioxidant activity ranged from very strong (nona, glodokan malang, soursop) to weak (sugar apple), while soursop leaf extract showed the highest SPF value, indicating strong photoprotective potential. The sunscreen activity is associated with polyphenolic compounds such as flavonoids and tannins. These findings suggest that Annonaceae leaf extracts, particularly soursop, may serve as promising natural ingredients for antioxidant and sunscreen formulations. Further studies on compound isolation, mechanism of action, and formulation stability are recommended to support their practical application in cosmeceuticals.

Acknowledgements: The authors would like to thank all contributors and supporting institutions for their valuable assistance and insights throughout this research.

Funding: This research received no specific grant.

Conflict of interest statement: The authors declare that there is no conflict of interest.

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