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The Potential of *Lantana Camara* Linn. as a Source of Quercetin, Gallic Acid, and Tannic Acid

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Abstract: *L. camara* Linn. is an invasive and dangerous plant that contains active substances beneficial to health. Active substances contained in the leaves of *L. camara* Linn. include flavonoids, gallic acid, and tannic acid. The purpose of this study was to explore the content of quercetin, gallic acid, and tannic acid in *L. camara* Linn leaf extract. The methods of this study include leaf extract of *L. camara* Linn. were tested organoleptic, pH, quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET). Measurement of quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET) levels was carried out with a spectrophotometer. The QEF content of *L. camara* Linn. leaf extract is 0.428 ± 0.004 mg/g. The GAEP content of *L. camara* Linn. leaf extract is 0.288 ± 0.002 mg/g, while the content of TAET is 0.384 ± 0.009 mg/g. This study confirmed the presence of flavonoids, phenols, and tannins in *L. camara* Linn. leaf extract, either extracted with ethanol or with other solvents such as acetone or petroleum ether. The novelty of this study is that the variations in active substance levels can be used as an option in the exploration and use of *L. camara* Linn. Thus, *L. camara* Linn. is not only a wild plant that endangers the environment but can also be a source for exploration of QEF, GAEP, and TAET.

Keywords: *Lantana camara* Linn., organoleptic test, quercetin equivalent flavonoid, gallic acid equivalent phenolic, tannic acid equivalent tannin.

馬纓丹卡馬拉林恩的潛力作為槲皮素、沒食子酸和單寧酸的來源

摘要：馬纓丹卡馬拉林恩。是一種侵入性危險植物，含有對健康有益的活性物質。馬纓丹卡馬拉林恩。葉子中含有的活性物質。包括類黃酮、沒食子酸和鞣酸。本研究的目的是探討馬纓丹卡馬拉林恩。葉提取物中槲皮素、沒食子酸和單寧酸的含量。本研究的方法包括馬纓丹卡馬拉林恩。的葉提取物。測試了感官、酸鹼度、槲皮素等效類黃酮、沒食子酸等效酚

類) 和單寧酸等效單寧。使用分光光度計測量槲皮素等效類黃酮、沒食子酸等效酚類和單寧酸等效單寧水平。馬纓丹卡馬拉林恩。的槲皮素等效類黃酮含量。葉提取物為 0.428 ± 0.004 毫克/克。馬纓丹卡馬拉林恩。的沒食子酸等效酚類磷內容。葉提取物為 0.288 ± 0.002 毫克/克，而和單寧酸等效單寧時間的含量為 0.384 ± 0.009 毫克/克。這項研究證實了馬纓丹卡馬拉林恩。中存在類黃酮、酚類和單寧酸。葉提取物，用乙醇或丙酮或石油醚等其他溶劑提取。這項研究的新穎之處在於，活性物質水平的變化可以作為探索和使用馬纓丹卡馬拉林恩。的一種選擇。因此，馬纓丹卡馬拉林恩。不僅是一種危害環境的野生植物，也是探索槲皮素等效類黃、沒食子酸等效酚類、和單寧酸等效單寧的資源。

关键词：馬纓丹卡馬拉林恩。，感官测试，槲皮素相當於黃酮，沒食子酸相當於酚類，鞣酸相當於單寧。

1. Introduction

Flavonoids are polyphenolic compounds found in various parts of plants. Eight classes of flavonoids exist: flavones, flavonols, flavanones, flavanone, isoflavones, flavantriol, anthocyanidins, and chalcone [1]. The benefits of flavonoids in health include anti-cancer, anti-oxidative, anti-inflammatory, and stimulating bone formation [2]. Recent studies have shown that flavonoids have antiviral activity against SARS-CoV-2 [3]. Gallic acid is a phenol compound known under another name, 3,4,5-trihydroxybenzoic acid, with the chemical structure of $C_6H_2(OH)_3COOH$ [4]. The results of recent studies demonstrate that *Swietenia macrophylla* produces gallic acid [5]. In general, plants produce gallic acid [6]. The benefits of gallic acid in the health sector include antimicrobial, prooxidant, antioxidant, anti-inflammatory, anti-platelet, anti-dengue, anti-cancer, and anti-apoptotic [7]. Tannins are phenolic compounds found in plants. There are two groups of tannins, namely hydrolyzable and condensed tannins. Gallotannins are hydrolyzable tannins, while catechins and gallic acid are condensed tannins [8]. Along with flavonoids and gallic acid, plants also produce tannins, *Hibiscus sabdariffa* tea [9], and *Dimocarpus longan* [10]. The biological activities of tannins include antimicrobial, antidiabetic, antioxidant, and cardioprotective [11].

The results of previous studies showed the content of flavonoids, gallic acid, and tannins in the following types of plants: QEF levels in methanol extract of *Melastoma malabathricum* L. fruit are 6,827 mg/g, while GAEP levels are 154,880 mg/g extract [12]. In addition, stem bark extract from *M. gigantea* contains flavonoids 25.2 mg/g [13]. It is interesting to note that

the content of phenolic catechins is equivalent in various varieties of *Vitis* sp. classified as high enough (> 900 mg/L) [14]. The results of other studies showed that different extraction methods against *M. malabathricum* L. show variation in GAEP levels [15], which are in line with the results of research demonstrating that tannin content is different in various cultivars of *Vitis* species Red Wines measured by various measurement methods [14].

Previously, we measured QEF levels at various concentrations of *L. camara* Linn. leaf extract cream. *L. camara* Linn. leaf collection was obtained from Tanjakan Cino Mati, Pleret District, Bantul Regency, Special Region of Yogyakarta, Indonesia [16]. *L. camara* Linn. is an invasive plant [17] considered dangerous in Indonesia [18]. Several researchers in Indonesia have explored the active ingredients of *L. camara* Linn. to be used for health [19, 20]. One possible use of *L. camara* Linn. for health is utilizing the content of active substances, including flavonoids, gallic acid, and tannic acid.

Since *L. camara* Linn. is invasive and contains active substances that are beneficial for health, we hope that the plant can apply as a source of flavonoids, gallic acid, and tannic acid. Research is still necessary to explore the content of flavonoids, gallic acid, and tannic acid in *L. camara* Linn. leaf extract. We hope that the results of this study can apply as a reference option about the potential of *L. camara* Linn. as a source of active ingredients in the form of quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET). Fig. 1 presents the structural formulas of flavonoids, gallic acid, and tannic acid.

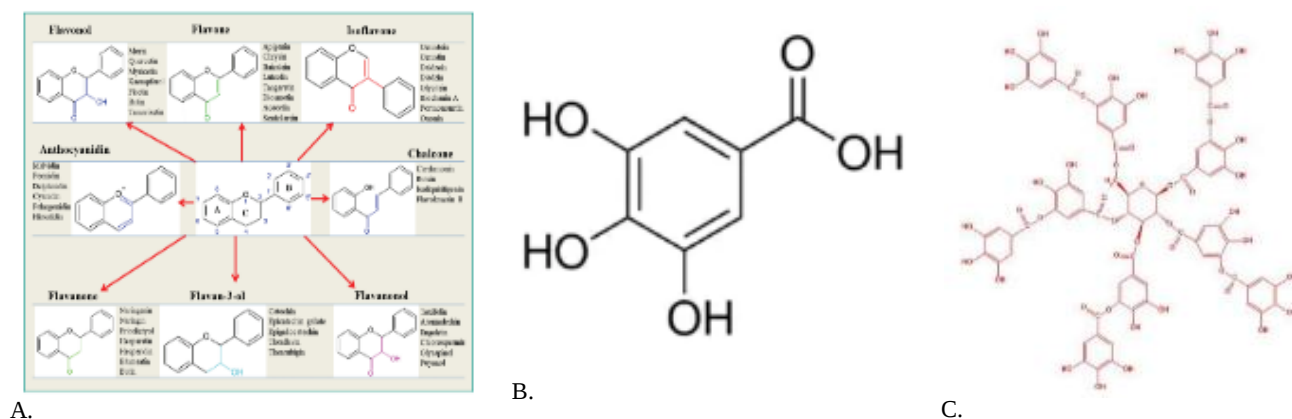


Fig. 1 The structural formula of flavonoids, gallic acid, and tannic acid: A – Basic structure and classification of flavonoids [1]; B – Gallic acid (3,4,5-Trihydroxybenzoic acid) [4, 21]; C – The structure of tannic acid [22]

2. Material and Methods

2.1. Research Design

The design of this research is laboratory experimental research. Fig. 2 shows the main process of this research.

2.2. *L. Camara* Linn. Leave Collection

Leaves of *L. camara* Linn. were collected from Tondano Kamangta Suluan street, Tombulu District, Minahasa Regency, North Sulawesi Province, Indonesia (1°21'46.6"N; 124°54'13.0"E) in December 2022. The location is available from <http://goo.gl/maps/nc1SVYhFU39q8nMz8>.

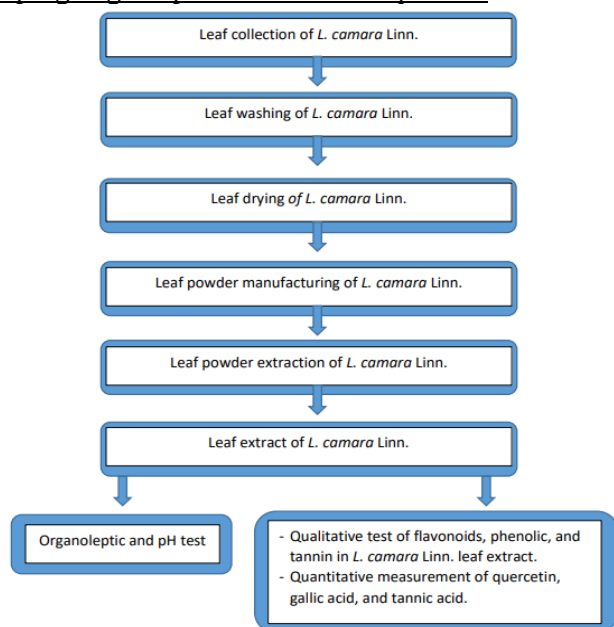


Fig. 2 The main process of this research

The collected leaves are washed under running water, covered with black cloth, and dried in the hot sun. Dried leaves of *L. camara* Linn. were ground into powder and then sifted to obtain a fine powder. The fine powder of *L. camara* Linn. leaves is extracted using 96% ethanol. *L. camara* Linn. leaf extract obtained in a viscous form, dark green color, is then put into sterile bottles and stored in a refrigerator. The

extract is ready for testing.

2.3. The Organoleptic and pH Tests of *L. Camara* Linn. Leaf Extract

Organoleptic tests performed on *L. camara* Linn. leaf extracts included shape, smell, and color. In addition, pH measurements used *L. camara* Linn. leaf extracts [23, 24, 25].

2.4. Qualitative Test of Flavonoids, Phenolics, and Tannins in *L. Camara* Linn. Leaf Extract

2.4.1. The Qualitative Test of Flavonoids

We dissolved 50 mg of the sample in 5 mL ethanol in a test tube, heated for five minutes, added a few drops of concentrated HCl and 0.2 g of Mg powder. The onset of the dark red for 3 min indicates a positive result.

2.4.2. The Qualitative Test of Phenolics

We dissolved one milliliter of the sample in a test tube with methanol and then added 5% FeCl₃. A change in color to orange-brown indicates a positive result in the presence of phenolic compounds.

2.4.3. The Qualitative Test of Tannin

We put 50 mg of the sample into a test tube, added ethanol until submerging the sample, and then added 2-3 drops of 1% FeCl₃ solution. The formation of bluish-black or green indicates a positive result for tannin content.

2.5. The Quantitative Measurement of Phytochemicals in *L. Camara* Linn. Leaf Extract

2.5.1. The Measurement of Flavonoid Levels

Measurement of QEF levels used aluminum chloride colorimetric assay [26] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 116351). The standard curve of QEF was duplicated with concentrations of 2, 4, 6, 8, and 10 µg/mL in 80% methanol solvent. One mL of each series of standard solution plus 4 mL distilled water was added to 0.30

mL 5%NaNO₂ and homogenized, then allowed to stand for 5 min. Next, we added 0.3 mL to 10% AlCl₃ and homogenized using a vortex mixer. After 5 min plus 2 mL of 1 M NaOH plus 2.4 mL of distilled water until a total volume of 10 mL. Absorbance readings for blanks and standard solutions were at a wavelength of 510 nm. We used the data obtained to create the standard curve of the flavonoid quercetin equivalent. To measure QEF levels in the samples, we made a sample solution of 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution reacted with the same reagents used in standard curve-making and absorbance readings. We calculated total QEF levels by comparing the absorbance of the sample against the standard quercetin curve, expressing the results as QEF in mg/g.

2.5.2. The Measurement of Gallic Acid Levels

The phenolic content measurement used the Folin-Ciocalteu assay [26, 27] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 116351). Standard gallic acid curves were duplicated in a volumetric flask. The concentration of gallic acid used is 5, 10, 15, 20, and 25 µg/mL each in 9 mL of distilled water. The blank reagent used is distilled water. 1 ml of Folin-Ciocalteu phenol reagent was added to each of the prepared standard solutions, homogenized, 5 min later added 2 mL of 7% Na₂CO₃ solution and 3.6 mL distilled water, and incubated for 90 min at room temperature. Absorbance readings used a spectrophotometer at a wavelength of 650 nm. To measure GAEP levels in samples, we prepared a sample solution, namely 1 mL of *L. camara* Linn. leaf extract, as a substitute for standard solutions. The sample solution reacted with the same reagents used on the standard curve and the absorbance readings. Total GAEP content was expressed in mg/g.

2.5.3. The Measurement of Tannic Acid Levels

Measurement of tannin content used the Folin-Ciocalteu assay [28] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 116351). Standard tannic acid curves were duplicated in a volumetric flask. The concentrations of tannic acid used are 10, 20, 40, 60, 80 µg/mL, each in 9 mL of distilled water. The blank reagent used is distilled water. One mL of each standard solution was put into a flask container containing 7.5 mL of distilled water. We added 0.5 mL of Follin Denish reagent to the flask, allowed it to stand for 3 minutes, then 1 mL of saturated Na₂CO₃ solution, and incubated for 15 minutes. Absorbance readings used a spectrophotometer at a wavelength of 740 nm. To measure TAET levels in samples, we prepared a sample solution which is 1 mL of *L. camara* Linn. leaf extract instead of standard solutions. The sample

solution reacted with the same reagents used on the standard curve and the absorbance readings. Total TAET content was expressed in mg/g.

2.6. Data Analysis

Descriptive analysis was conducted on phytochemical data of *L. camara* Linn leaf extract, namely QEF, GAEP, and TAET levels. The phytochemical content of *L. camara* Linn. leaf extract is presented in table and graphic form using the Microsoft Excel.

3. Results

3.1. Leaves of *L. Camara* Linn

Fig. 3 presents leaves of *L. camara* Linn. collected from Tondano Kamangta Suluan street, Tombulu District, Minahasa Regency, North Sulawesi Province, Indonesia (1°21'46.6"N; 124°54'13.0"E).

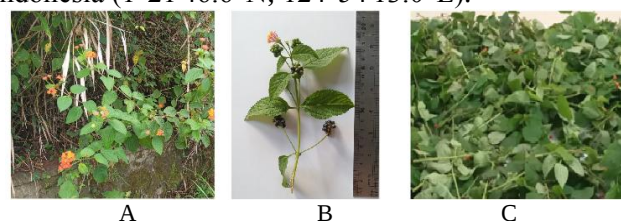


Fig. 3 *L. camara* Linn.: A – *L. camara* Linn. as a wild plant; B – Habitus of *L. camara* Linn. shows stems, leaves, flowers, and fruit; C – Leaves of *L. camara* Linn. as extraction material. Photographer - Hosea Jaya Edy, December 20, 2022

3.2. The Results of Organoleptic Test and pH Test of *L. Camara* Linn. Leaf Extract

Table 1 presents test results of *L. camara* Linn. leaf extract.

Table 1 Test results organoleptic and pH of *L. camara* Linn. leaf extract

The type of test	Results
Organoleptic	
Shape	Semi solid
Smell	Typical for <i>L. camara</i> Linn. leaf extract
Color	Slightly blackish green
pH	5

Table 2 presents the results of the qualitative examination of the active substance of *L. camara* Linn. leaf extract.

Table 2 Qualitatively of the active substance of *L. camara* Linn. leaf extract

Compounds tested	Color change results	Result
Flavonoid	Brick red	Positive
Fenolic	Orange brown	Positive
Tannin	Greenish-brown transparent	Positive

3.3. Quercetin Equivalent Flavonoid

Fig. 4 presents the standard curve of quercetin equivalent flavonoid.

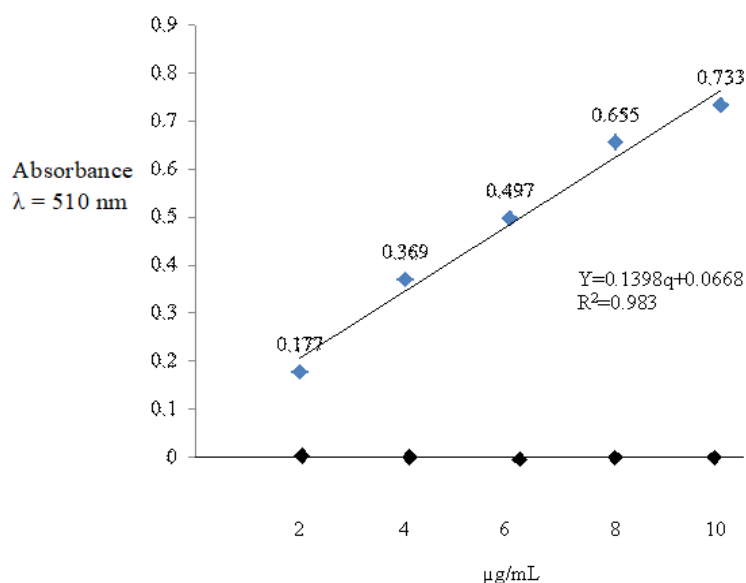


Fig. 4 The standard curve of quercetin equivalent of flavonoid

The standard curve used for the analysis of QEF levels in this study was $Y = 0.1398q + 0.0668$ (Fig. 4), where Y = absorbance; $a = 0.1398$; $b = 0.0668$; q = quercetin equivalent flavonoid (mg/L) levels. In addition, a standard solution with a concentration of 1.0-10.0 $\mu\text{g/mL}$ obtained a coefficient of determination (R^2) = 0.983. To calculate the total QEF (Q_{QEF}) per

gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{\text{QEF}} = q \times v \times (p/m)$$

where q - QEF levels in the sample, v - sample volume, p - dilution, and m - sample mass/weight.

Table 3 presents QEF levels of *L. camara* Linn. leaf extract.

Table 3 Quercetin equivalent flavonoid levels of *L. camara* Linn leaf extract

Sample	Y	a	b	v, L	p	M, g	Q_{QEF} , mg/g
1	0.661	0.1398	0.0668	0.001	10	0.1	0.425036
2	0.659	0.1398	0.0668	0.001	10	0.1	0.423605
3	0.673	0.1398	0.0668	0.001	10	0.1	0.433319
4	0.669	0.1398	0.0668	0.001	10	0.1	0.431107
5	0.661	0.1398	0.0668	0.001	10	0.1	0.425205
6	0.664	0.1398	0.0668	0.001	10	0.1	0.42712
Mean							0.428
SD							0.004

Notes: Y - absorbance at a wavelength $\lambda = 510$ nm; a - coefficient; b - constant; v - volume (liters); p - dilution; m - sample weight (gram); q - quercetin equivalent flavonoid levels; Q_{QEF} - total quercetin equivalent flavonoid; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

3.4. Phenolic Equivalent Gallic Acid

Fig. 5 presents the standard curve of GAEP.

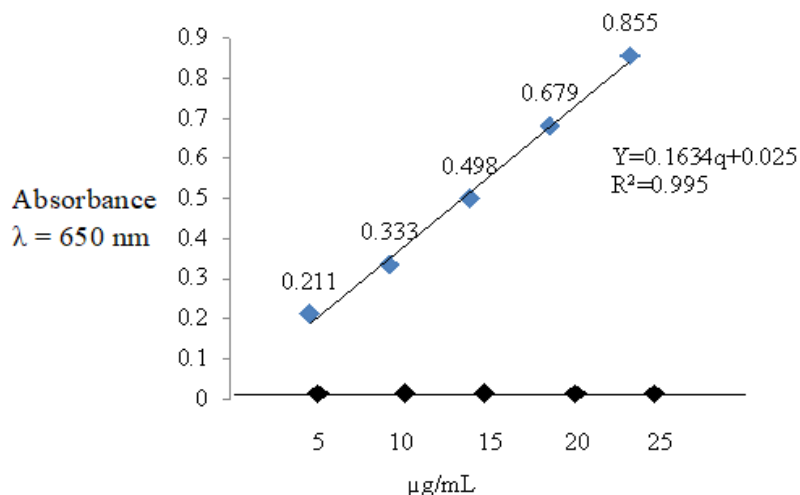


Fig. 5 The standard curve of gallic acid equivalent of phenolics

The standard curve of GAEP used in this study is $Y = 0.1634q + 0.025$ (Fig. 5), where Y = absorbance; $a =$

0.1634; $b = 0.025$; q_{GAEP} = gallic acid equivalent phenolics (mg/L). In addition, a standard solution with

concentration of 5.0–25.0 µg/mL obtained coefficient of determination (R^2) = 0.995. To calculate GAEP (Q_{GAEP}) per gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{GAEP} = q \times v \times (p/m)$$

where q_{GAEP} - GAEP levels in the sample, v - sample volume, p - dilution, and m - sample mass/weight.

Table 4 presents the results of the analysis of gallic acid equivalent phenolic levels of *L. camara* Linn. leaf extract.

Table 4 Levels of gallic acid equivalent phenolic of *L. camara* Linn. leaf extract

Sample	Y	a	b	v, L	p	m, g	Q_{FEGA} , mg/g
1	0.491	0.1634	0.025	0.001	10	0.1	0.285189
2	0.499	0.1634	0.025	0.001	10	0.1	0.290086
3	0.497	0.1634	0.025	0.001	10	0.1	0.288861
4	0.496	0.1634	0.025	0.001	10	0.1	0.288045
5	0.496	0.1634	0.025	0.001	10	0.1	0.288285
6	0.495	0.1634	0.025	0.001	10	0.1	0.287928
Mean							0.288
SD							0.002

Notes: Y - absorbance at a wavelength (λ) 650 nm; a - coefficient; b - constant; v - volume (liters); p - dilution; m - sample weight (gram); q_{GAEP} - phenolic equivalent to gallic acid levels; Q_{FEGA} - total gallic acid equivalent phenolic in the sample; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

3.5. Tannin Equivalent Tannic Acid

Fig. 6 presents the standard curve of TAET.

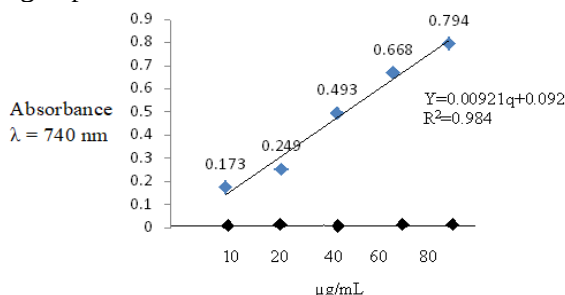


Fig. 6 The standard curve of tannin equivalent tannic acid

The standard curve for TAET used in this study is $Y = 0.00921q + 0.092$. The coefficient of determination (R^2) is 0.984 (Fig. 6). In the equation, Y = absorbance; a = 0.1661; b = 0.0229; q = TAET levels (mg/L). To calculate TAET (Q_{TAET}) per gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{TAET} = q \times v \times (p/m)$$

where q - TAET levels in the sample, v - sample volume, p - dilution, and m - sample mass/weight.

Table 5 presents tannic acid equivalent tannin levels in the *L. camara* Linn. leaf extract.

Table 5 Tannic acid equivalent tannin levels in *L. camara* Linn. leaf extract

Sample	Y	a	b	v, L	p	m, g	Q_{TAET} , mg/g
1	0.124	0.00921	0.0890	0.001	10	0.1	0.3800217
2	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
3	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
4	0.124	0.00921	0.0890	0.001	10	0.1	0.3800217
5	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
6	0.123	0.00921	0.0890	0.001	10	0.1	0.3691640
Mean							0.384
SD							0.009

Notes: Y - absorbance in wavelength (λ) of 740 nm; a - coefficient; b - constant; v - volume (liter); p - dilution; m - sample weight (gram); q_{TAET} - tannic acid equivalent tannin levels; Q_{TAET} - total tannin equivalent tannic acid; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

Fig. 7 presents the phytochemical profile of *L. camara* Linn. leaf extract.

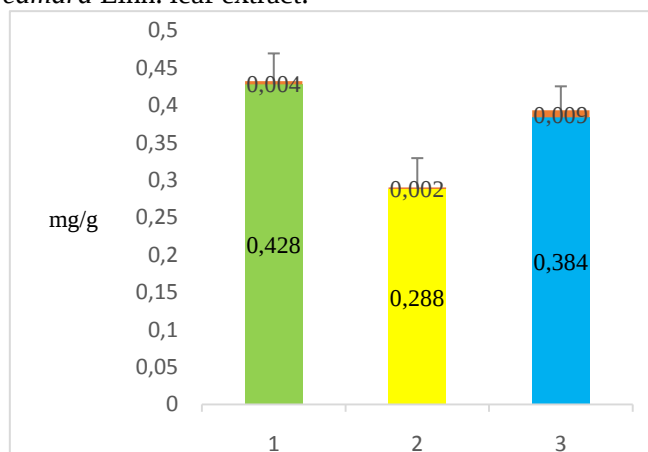


Fig. 7 Phytochemical profile of *L. camara* Linn. leaf extract: 1 –

QEF; 2 – GAEP; 3 – TAET

4. Discussion

The results of organoleptic tests on *L. camara* Linn. leaf extract in this study are similar to the results of our previous research, including the form is semi-solid, the smell is similar to the smell of *L. camara* Linn. leaves, and the color is slightly blackish green [16]. The pH of *L. camara* Linn. leaf extract is normal as it is in the range of 4.5-6.5. The pH of *L. camara* Linn. leaf extract is consistent with the pH of human skin [29]. Compared to the topical formula, the pH of *L. camara* Linn. leaf extract in this study was under the pH of topical preparations containing ibuprofen [30].

The content of flavonoids, phenols, and tannins in *L. camara* Linn. leaf extract in this study was the same

as the results of previous studies [20, 31, 32]. [33] also shows flavonoid content in the leaves of *L. camara* Linn. in extraction using acetone. In addition, methanol extraction of *L. camara* Linn. leaves also showed flavonoid content [32]. In addition, the extract drying method of *L. camara* Linn. leaves also shows flavonoids and tannins [34]. The results of another study demonstrated that the leaves of *L. camara* Linn. extracted using petroleum ether (40°C), chloroform, and methanol also contain flavonoids and tannins [35].

The QEF levels of *L. camara* Linn. leaf extract in this study was lower than the results of other studies [31, 32, 36, 37]. The results of previous studies demonstrated that various varieties of *L. camara* Linn. have QEF content ranging from 16.14 ± 0.21 to 25.22 ± 2.59 mg/g extract [36]. The results of another study showed the QEF content in the dry extract of *L. camara* Linn. 12.44 ± 2.85 mg/g [37]. Another study showed that the methanol extract of *L. camara* Linn. leaves contained QEF 243.89 mg/g extract [31]. The results of another study demonstrated that several fractions of methanol extract of *L. camara* Linn. leaves contained QEF ranging from 19.85 to 97.56 mg/g samples [32]. The results of other studies also revealed that the QEF content of the methanol extract of aerial parts of *L. camara* Linn. from Nepal ranged from 1.87 ± 0.16 to 0 mg/g extract [38]. On the other hand, the results of other studies demonstrate that the ethanol extract of *L. camara* Linn. leaves contains low QEF, which is 0.2423 ± 0.0068 mg/g extract [39]. These results are lower than the QEF content in our study.

GAEP levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies [31, 32, 36-38]. Previous research demonstrated that various varieties of *L. camara* Linn. have GAEP content ranging from 55.57 ± 2.82 to 232.99 ± 15.97 mg/g extract [36]. Other research results also showed that the dry extract of *L. camara* Linn. contains GAEP 144.7 ± 1.34 mg/g [37]. Another study showed that the methanol extract of *L. camara* Linn. leaves contained GAEP 563.57 ± 2.49 mg/g extract, while the GAEP content in flower extract was 614.79 ± 1.54 mg/g extract [31]. The results of another study demonstrated that the GAEP content of *L. camara* Linn. leaf extract was 10.20 ± 0.343 mg/g extract [38]. The results of another study demonstrated the GAEP content in various fractions of methanol leaf extract of *L. camara* Linn. ranging from 20.25 ± 0.41 to 98.81 ± 0.27 mg/g sample [32]. As a reference, the results of research on the content of GAEP in other plants turned out to vary, for example, *Ageratina adenophora* contains GAEP 4.70 ± 0.059 mg/g extract, while *Cupressus sempervirens* contains GAEP 4.31 ± 0.147 mg/g extract [38].

The TAET levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies. The study results demonstrated that the tannin content in *L. camara* Linn. leaf extract was 98.40 ± 6.88 mg/g

[40]. The results of another study showed that the tannin content of *L. camara* Linn. extract was 0.860 ± 0.038 mg/g [41]. On the other hand, some research results demonstrate that the ethanol extract of *L. camara* Linn. leaves contains low tannins in extract, namely 0.2179 ± 0.0056 mg/g [39]. These results were lower than the tannin content in our study. Some studies also demonstrate that tannin levels from the methanol extract of *L. camara* Linn. collected from a semi-arid region of Brazil are not detected [42, 43].

Note the research results demonstrate that the content of GAEP and QEF in *L. rhodesiensis* extract is highest in the leaves and then the stem, while the least is found in the roots [44]. Other research results are the estimation of phenolics, flavonoids, and tannin contents in various solvent extracts of coconut. The results showed that the methanol fraction contained a total phenolic equivalent of gallic acid 822.60 ± 16.36 mg/g sample, a flavonoid equivalent of quercetin 103.30 ± 9.78 mg/g sample, and a tannic acid equivalent of tannin 663.50 ± 19.26 mg/g sample [45].

Based on the data above, there are variations in QEF, GAEP, and TAET levels influenced by variations in plants, environment, and solvents used for extraction. The research results showing that extraction conditions affect flavonoid levels [46] reinforce our statement. Based on the results of these studies as well as other studies [47], flavonoid content was measured in plant extracts [48, 49] and herbal preparations [50, 51, 52, and 53].

The limitations of this study include not examining the mineral content, which can affect the levels of active substances in *L. camara* Linn. leaf extract. Therefore, we suggest for research that it is necessary to measure mineral levels, especially Fe and Zn, which relate to the levels of substances in *L. camara* Linn. leaf extract. The levels of these two minerals affected the stability of QEF levels in *L. camara* Linn leaf extract cream [16].

5. Conclusion

The ethanolic extract of *L. camara* Linn. contains levels of QEF, GAEP, and TAET as well as 0.428 ± 0.004 mg/g extract, 0.288 ± 0.002 mg/g extract, and 0.384 ± 0.009 mg/g extract, respectively. The content of active substance levels can apply as a reference to explore *L. camara* Linn. as a source of quercetin, gallic acid, and tannic acid.

Based on the results of our research and other studies, and as described above, it turns out that the leaf extract of *L. camara* Linn. contains QEF, GAEP, and TAET, but levels vary. The type and place of life influence the variation of QEF, GAEP, and TAET levels in *L. camara* Linn. leaf extract. Nonetheless, we hope that the variations in QEF, GAEP, and TAET levels in *L. camara* Linn. can be used as an option in the exploration and use of *L. camara* Linn. Thus, *L. camara* Linn. is not only a wild plant that endangers

the environment but can also apply as a source of QEF, GAEP, and TAET exploration.

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The Potential of lantana camara linn. As a source of quercetin, gallic acid, and tannic acid

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The Potential of *Lantana Camara* Linn. as a Source of Quercetin, Gallic Acid, and Tannic Acid

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Abstract: *L. camara* Linn. is ¹⁴invasive and dangerous plant that contains active substances beneficial to ¹⁴alth. Active substances contained in the leaves of *L. camara* Linn. include flavonoids, gallic acid, and tannic acid. The purpose of this study was to explore the content of quercetin, gallic acid, and tannic acid in *L. camara* Linn leaf extract. The methods of this study include leaf extract of *L. camara* Linn. were tested organoleptic, pH, quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET). Measurement of quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET) levels was carried out with a spectrophotometer. The QEF content of *L. camara* Linn. leaf extract is 0.428 ± 0.004 mg/g. The GAEP content of *L. camara* Linn. leaf extract is 0.288 ± 0.002 mg/g, while the content of TAET is 0.384 ± 0.009 mg/g. This study confirmed the presence of flavonoids, phenols, and tannins in *L. camara* Linn. leaf extract, either extracted with ethanol or with other solvents such as acetone or petroleum ether. The novelty of this study is that the ¹⁵variations in active substance levels can be used as an option in the exploration and use of *L. camara* Linn. Thus, *L. camara* Linn. is not only a wild plant that endangers the environment but can also be a source for exploration of QEF, GAEP, and TAET.

Keywords: *Lantana camara* Linn., organoleptic test, quercetin equivalent flavonoid, gallic acid equivalent phenolic, tannic acid equivalent tannin.

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馬纓丹卡馬拉林恩的潛力作為槲皮素、沒食子酸和單寧酸的來源

26

摘要：馬纓丹卡馬拉林恩。是一種侵入性危險植物，含有對健康有益的活性物質。馬纓丹卡馬拉林恩。葉子中含有的活性物質。包括類黃酮、沒食子酸和鞣酸。本研究的目的是探討馬纓丹卡馬拉林恩。葉提取物中槲皮素、沒食子酸和單寧酸的含量。本研究的方法包括馬纓丹卡馬拉林恩。的葉提取物。測試了感官、酸鹼度、槲皮素等效類黃酮、沒食子酸等效酚

類) 和單寧酸等效單寧。使用分光光度計測量槲皮素等效類黃酮、沒食子酸等效酚類和單寧酸等效單寧水平。馬纓丹卡馬拉林恩。的槲皮素等效類黃酮含量。葉提取物為 0.428 ± 0.004 毫克/克。馬纓丹卡馬拉林恩。的沒食子酸等效酚類磷內容。葉提取物為 0.288 ± 0.002 毫克/克，而和單寧酸等效單寧時間的含量為 0.384 ± 0.009 毫克/克。這項研究證實了馬纓丹卡馬拉林恩。中存在類黃酮、酚類和單寧酸。葉提取物，用乙醇或丙酮或石油醚等其他溶劑提取。這項研究的新穎之處在於，活性物質水平的變化可以作為探索和使用馬纓丹卡馬拉林恩。的一種選擇。因此，馬纓丹卡馬拉林恩。不僅是一種危害環境的野生植物，也是探索槲皮素等效類黃、沒食子酸等效酚類、和單寧酸等效單寧的資源。

关键词：馬纓丹卡馬拉林恩。，感官测试，槲皮素相當於黃酮，沒食子酸相當於酚類，鞣酸相當於單寧。

1. Introduction

Flavonoids are polyphenolic compounds found in various parts of plants. Eight classes of flavonoids exist: flavones, flavonols, flavanones, flavanol, isoflavones, flavantriol, anthocyanidins, and chalcone [1]. The benefits of flavonoids in health include anti-cancer, anti-oxidative, anti-inflammatory, and stimulating bone formation [2]. Recent studies have shown that flavonoids have antiviral activity against SARS-CoV-2 [3]. Gallic acid is a phenol compound known under another name, 3,4,5-trihydroxybenzoic acid, with the chemical structure of $C_6H_2(OH)_3COOH$ [4]. The results of recent studies demonstrate that *Swietenia macrophylla* produces gallic acid [5]. In general, plants produce gallic acid [6]. The benefits of gallic acid in the health sector include antimicrobial, prooxidant, antioxidant, anti-inflammatory, anti-platelet, anti-dengue, anti-cancer, and anti-apoptotic [7]. Tannins are phenolic compounds found in plants. There are two groups of tannins, namely hydrolyzable and condensed tannins. Gallotannins are hydrolyzable tannins, while catechins and gallocatechins are condensed tannins [8]. Along with flavonoids and gallic acid, plants also produce tannins, *Hibiscus sabdariffa* tea [9], and *Dimocarpus longan* [10]. The biological activities of tannins include antimicrobial, antidiabetic, antioxidant, and cardioprotective [11].

The results of previous studies showed the content of flavonoids, gallic acid, and tannins in the following types of plants: QEF levels in methanol extract of *Melastoma malabathricum* L. fruit are 6,827 mg/g, while GAEP levels are 154,880 mg/g extract [12]. In addition, stem bark extract from *M. gigantea* contains flavonoids 25.2 mg/g [13]. It is interesting to note that

the content of phenolic catechins is equivalent in various varieties of *Vitis* sp. classified as high enough (> 900 mg/L) [14]. The results of other studies showed that different extraction methods against *M. malabathricum* L. show variation in GAEP levels [15], which are in line with the results of research demonstrating that tannin content is different in various cultivars of *Vitis* species Red Wines measured by various measurement methods [14].

Previously, [19] measured QEF levels at various concentrations of *L. camara* Linn. leaf extract cream. *L. camara* Linn. leaf collection was obtained from Tanjakan Cino Mati, Pleret District, Bantul Regency, Special Region of Yogyakarta, Indonesia [16]. *L. camara* Linn. is an invasive plant [17] considered dangerous in Indonesia [18]. Several researchers in Indonesia have explored the active ingredients of *L. camara* Linn. to be used for health [19, 20]. One possible use of *L. camara* Linn. for health is utilizing the content of active substances, including flavonoids, gallic acid, and tannic acid.

Since *L. camara* Linn. is invasive and contains active substances that are beneficial for health, we hope that the plant can apply as a source of flavonoids, gallic acid, and tannic acid. Research is still necessary to explore the content of flavonoids, gallic acid, and tannic acid in *L. camara* Linn. leaf extract. We hope that the result of this study can apply as a reference option about the potential of *L. camara* Linn. as a source of active ingredients in the form of quercetin equivalent flavonoid (QEF), gallic acid equivalent phenolic (GAEP), and tannic acid equivalent tannin (TAET). Fig. 1 presents the structural formulas of flavonoids, gallic acid, and tannic acid.

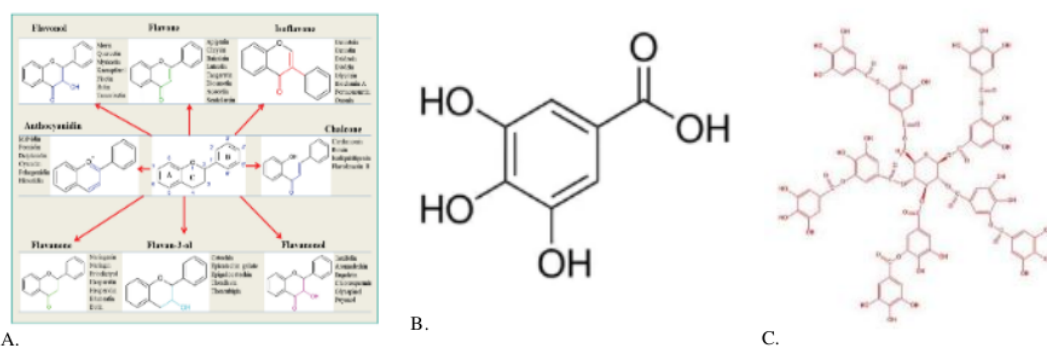


Fig. 1 The structural formula of flavonoids, gallic acid, and tannic acid: A – Basic structure and classification of flavonoids [1]; B – Gallic acid (3,4,5-Trihydroxybenzoic acid) [4, 21]; C – The structure of tannic acid [22]

2. Material and Methods

2.1. Research Design

The design of this research is laboratory experimental research. Fig. 2 shows the main process of this research.

2.2. *L. Camara* Linn. Leave Collection

Leaves of *L. camara* Linn. were collected from Tondano Kamangta Suluan street, Tombulu District, Minahasa Regency, North Sulawesi Province, Indonesia (1°21'46.6"N; 124°54'13.0"E) in December 2022. The location is available from <http://goo.gl/maps/nc1SVYhFU39q8nMz8>.

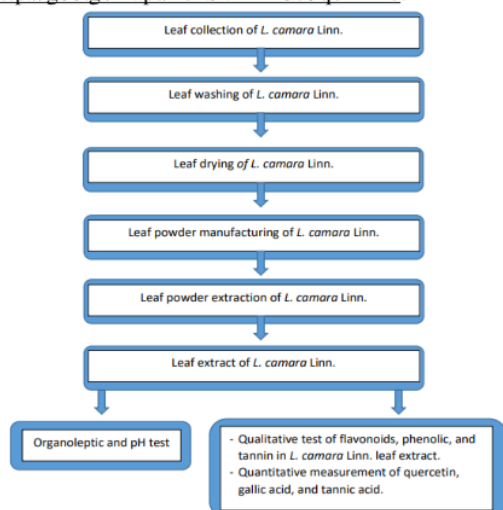


Fig. 2 The main process of this research

The collected leaves are washed under running water, covered with black cloth, and dried in the hot sun. Dried leaves of *L. camara* Linn. were ground into powder and then sifted to obtain a fine powder. The fine powder of *L. camara* Linn. leaves is extracted using 96% ethanol. *L. camara* Linn. leaf extract obtained in a viscous form, dark green color, is then put into sterile bottles and stored in a refrigerator. The

extract is ready for testing.

2.3. The Organoleptic and pH Tests of *L. Camara* Linn. Leaf Extract

Organoleptic tests performed on *L. camara* Linn. leaf extracts included shape, smell, and color. In addition, pH measurements used *L. camara* Linn. leaf extracts [23, 24, 25].

2.4. Qualitative Test of Flavonoids, Phenolics, and Tannins in *L. Camara* Linn. Leaf Extract

2.4.1. The Qualitative Test of Flavonoids

We dissolved 50 mg of the sample in 5 mL ethanol in a test tube, heated for five minutes, added a few drops of concentrated HCl and 0.2 g of Mg powder. The onset of the dark red for 3 min indicates a positive result.

2.4.2. The Qualitative Test of Phenolics

We dissolved one milliliter of the sample in a test tube with methanol and then added 5% FeCl₃. A change in color to orange-brown indicates a positive result in the presence of phenolic compounds.

2.4.3. The Qualitative Test of Tannin

We put 50 mg of the sample into a test tube, add ethanol until submerging the sample, and then added 2-3 drops of 1% FeCl₃ solution. The formation of bluish-black or green indicates a positive result for tannin content.

2.5. The Quantitative Measurement of Phytochemicals in *L. Camara* Linn. Leaf Extract

2.5.1. The Measurement of Flavonoid Levels

Measurement of QEF levels used aluminum chloride colorimetric assay [26] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 11634). The standard curve of QEF was duplicated with concentrations of 2, 4, 6, 8, and 10 µg/mL in 80% methanol solvent. One mL of each series of standard solution plus 4 mL distilled water was added to 0.30

mL 5%NaNO₂ and homogenized, then allowed to stand for 5 min. Next, we added 0.3 mL to 10% AlCl₃ and homogenized using a vortex mixer. After 5 min plus 9 mL of 1 M NaOH plus 2.4 mL of distilled water until a total volume of 10 mL. Absorbance readings for blanks and standard solutions were at a wavelength of 510 nm. We used the data obtained to create the standard curve of the flavonoid quercetin equivalent. To measure QEF levels in the samples, we made a sample solution of 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution reacted with the same reagents used in standard curve-making and absorbance readings. We calculated total QEF levels by comparing the absorbance of the sample against the standard quercetin curve, expressing the results as QEF in mg/g.

2.5.2. The Measurement of Gallic Acid Levels

The phenolic content measurement used the Folin-Ciocalteu assay [26, 27] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 116351). Standard gallic acid curves were duplicated in a volumetric flask. The concentration of gallic acid used is 5, 10, 15, 20, and 25 µg/mL each in 17 mL of distilled water. The blank reagent used is distilled water. 1 mL of Folin-Ciocalteu phenol reagent was added to each of the prepared standard solutions, homogenized, 5 min after added 2 mL of 7% Na₂CO₃ solution and 3.6 mL distilled water, and incubated for 90 min at room temperature. Absorbance readings used a spectrophotometer at a wavelength of 650 nm. To measure GAEP levels in samples, we prepared a sample solution, namely 1 mL of *L. camara* Linn. leaf extract, as a substitute for standard solutions. The sample solution reacted with the same reagents used on the standard curve and the absorbance readings. Total GAEP content was expressed in mg/g.

2.5.3. The Measurement of Tannic Acid Levels

Measurement of tannin content used the Folin-Ciocalteu assay [28] using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, serial No. 116351). Standard tannic acid curves were duplicated in a volumetric flask. The concentrations of tannic acid used are 10, 20, 40, 60, 80 µg/mL, each in 9 mL of distilled water. The blank reagent used is distilled water. One mL of each standard solution was put into a flask container containing 7.5 mL of distilled water. We added 0.7 mL of Follin Denish reagent to the flask, allowed it to stand for 3 minutes, then 1 mL of saturated Na₂CO₃ solution, and incubated for 15 minutes. Absorbance readings used a spectrophotometer at a wavelength of 740 nm. To measure TAET levels in samples, we prepared a sample solution which is 1 mL of *L. camara* Linn. leaf extract instead of standard solutions. The sample

solution reacted with the same reagents used on the standard curve and the absorbance readings. Total TAET content was expressed in mg/g.

2.6. Data Analysis

Descriptive analysis was conducted on phytochemical data of *L. camara* Linn leaf extract, namely QEF, GAEP, and TAET levels. The phytochemical content of *L. camara* Linn. leaf extract is presented in table and graphic form using the Microsoft Excel.

3. Results

3.1. Leaves of *L. Camara* Linn

Fig. 3 presents leaves of *L. camara* Linn. collected from Tondano Kamangta Suluan street, Tombulu District, Minahasa Regency, North Sulawesi Province, Indonesia (1°21'46.6"N; 124°54'13.0"E).



Fig. 3 *L. camara* Linn.: A – *L. camara* Linn. as a wild plant; B – Habit of *L. camara* Linn. shows stems, leaves, flowers, and fruit; C – Leaves of *L. camara* Linn. as extraction material. Photographer - Hosea Jaya Edy, December 20, 2022

2.2. The Results of Organoleptic Test and pH Test of *L. Camara* Linn. Leaf Extract

Table 1 presents test results of *L. camara* Linn. leaf extract.

Table 1 Test results organoleptic and pH of *L. camara* Linn. leaf extract

The type of test	Results
Organoleptic	
Shape	Semi solid
Smell	Typical for <i>L. camara</i> Linn. leaf extract
Color	Slightly blackish green
pH	5

Table 2 presents the results of the qualitative examination of the active substance of *L. camara* Linn. leaf extract.

Table 2 Qualitatively of the active substance of *L. camara* Linn. leaf extract

Compounds tested	Color change results	Result
Flavonoid	Brick red	Positive
Fenolic	Orange brown	Positive
Tannin	Greenish-brown transparent	Positive

3.3. Quercetin Equivalent Flavonoid

Fig. 4 presents the standard curve of quercetin equivalent flavonoid.

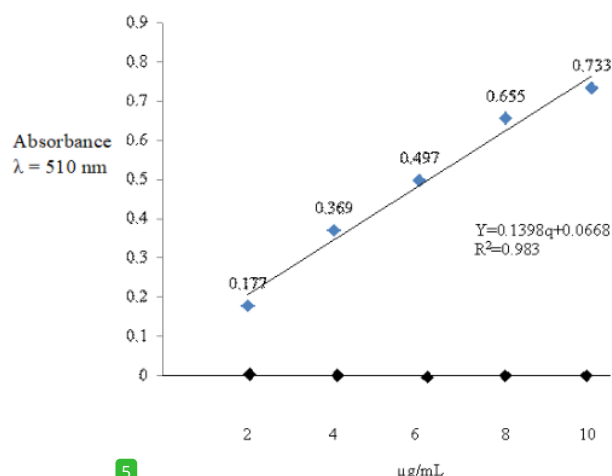


Fig. 4 The standard curve of quercetin equivalent of flavonoid

The standard curve used for the analysis of QEF levels in this study was $Y = 0.1398q + 0.0668$ (Fig. 4), where Y = absorbance; $a = 0.1398$; $b = 0.0668$; q = quercetin equivalent flavonoid (mg/L) levels. In addition, a standard solution with a concentration of 1.0-10.0 $\mu\text{g/mL}$ obtained a coefficient of determination (R^2) = 0.983. To calculate the total QEF (Q_{QEF}) per

gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{\text{QEF}} = q \times v \times (p/m)$$

where q - QEF levels in the sample, v - sample volume, p - dilution, and m - sample mass weight.

Table 3 presents QEF levels of *L. camara* Linn. leaf extract.

Sample	Y	a	b	v, L	p	M, g	Q_{QEF} , mg/g
1	0.661	0.1398	0.0668	0.001	10	0.1	0.425036
2	0.659	0.1398	0.0668	0.001	10	0.1	0.423605
3	0.673	0.1398	0.0668	0.001	10	0.1	0.433319
4	0.669	0.1398	0.0668	0.001	10	0.1	0.431107
5	0.661	0.1398	0.0668	0.001	10	0.1	0.425205
6	0.664	0.1398	0.0668	0.001	10	0.1	0.42712
Mean							0.428
SD							0.004

Notes: Y - absorbance at a wavelength $\lambda = 510$ nm; a - coefficient; b - constant; v - volume (liters); p - dilution; m - sample weight (gram); q - quercetin equivalent flavonoid levels; Q_{QEF} - total quercetin equivalent flavonoid; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

3.4. Phenolic Equivalent Gallic Acid

Fig. 5 presents the standard curve of GAEP.

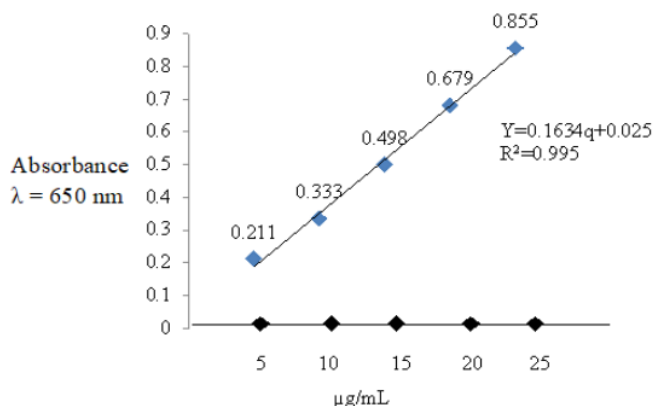


Fig. 5 The standard curve of gallic acid equivalent of phenolics

The standard curve of GAEP used in this study is $Y = 0.1634q + 0.025$ (Fig. 5), where Y = absorbance; $a =$

0.1634; $b = 0.025$; q_{GAEP} = gallic acid equivalent phenolics (mg/L). In addition, a standard solution with

concentration of 5.0–25.0 µg/mL obtained coefficient of determination (R^2) = 0.995. To calculate GAEP (Q_{GAEP}) per gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{GAEP} = q \times v \times (p/m)$$

where q_{GAEP} - GAEP levels in the sample, v - sample volume, p - dilution, and m - sample mass/weight.

Table 4 presents the results of the analysis of gallic acid equivalent phenolic levels of *L. camara* Linn. leaf extract.

6 Table 4 Levels of gallic acid equivalent phenolic of *L. camara* Linn. leaf extract

Sample	Y	a	b	v, L	p	m, g	Q_{FEGA} , mg/g
1	0.491	0.1634	0.025	0.001	10	0.1	0.285189
2	0.499	0.1634	0.025	0.001	10	0.1	0.290086
3	0.497	0.1634	0.025	0.001	10	0.1	0.288861
4	0.496	0.1634	0.025	0.001	10	0.1	0.288045
5	0.496	0.1634	0.025	0.001	10	0.1	0.288285
6	0.495	0.1634	0.025	0.001	10	0.1	0.287928
Mean							0.288
SD							0.002

Notes: Y - absorbance at a wavelength (λ) 650 nm; a - coefficient; b - constant; v - volume (liters); p - dilution; m - sample weight (gram); q_{GAEP} - phenolic equivalent to gallic acid levels; Q_{FEGA} - total gallic acid equivalent phenolic in the sample; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

3.5. Tannin Equivalent Tannic Acid

Fig. 6 presents the standard curve of TAET.

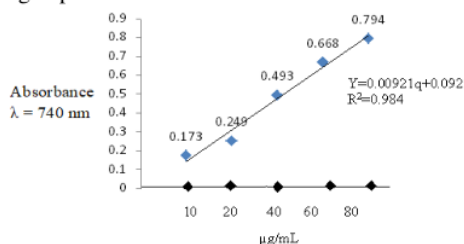


Fig. 6 The standard curve of tannin equivalent tannic acid

The standard curve for TAET used in this study is $Y = 0.00921q + 0.092$. The coefficient of determination (R^2) is 0.984 (Fig. 6). In the equation, Y = absorbance; a = 0.1661; b = 0.0229; q = TAET levels (mg/L). To calculate TAET (Q_{TAET}) per gram of *L. camara* Linn. leaf extract, we used the formula

$$Q_{TAET} = q \times v \times (p/m)$$

where q - TAET levels in the sample, v - sample volume, p - dilution, and m - sample mass/weight

Table 5 presents tannic acid equivalent tannin levels in the *L. camara* Linn. leaf extract.

6 Table 5 Tannic acid equivalent tannin levels in *L. camara* Linn. leaf extract

Sample	Y	a	b	v, L	p	m, g	Q_{TAET} , mg/g
1	0.124	0.00921	0.0890	0.001	10	0.1	0.3800217
2	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
3	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
4	0.124	0.00921	0.0890	0.001	10	0.1	0.3800217
5	0.125	0.00921	0.0890	0.001	10	0.1	0.3908795
6	0.123	0.00921	0.0890	0.001	10	0.1	0.3691640
Mean							0.384
SD							0.009

Notes: Y - absorbance in wavelength (λ) of 740 nm; a - coefficient; b - constant; v - volume (liter); p - dilution; m - sample weight (gram); q_{TAET} - tannic acid equivalent tannin levels; Q_{TAET} - total tannin equivalent tannic acid; mg/L - milligrams per liter; mg/g - milligrams per gram; SD - standard deviation

Fig. 7 presents the phytochemical profile of *L. camara* Linn. leaf extract.

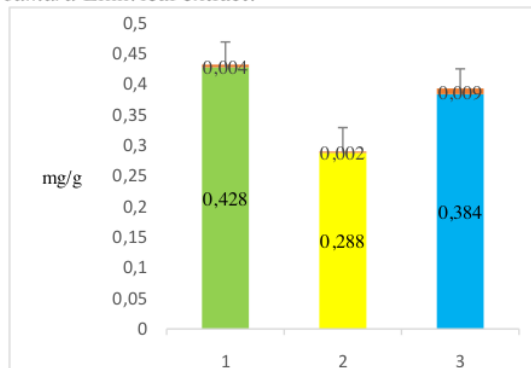


Fig. 7 Phytochemical profile of *L. camara* Linn. leaf extract: 1 –

QEF; 2 – GAEP; 3 – TAET

4. Discussion

The results of organoleptic tests on *L. camara* Linn. leaf extract in this study are similar to the results of our previous research, including the form is semi-solid, the smell is similar to the smell of *L. camara* Linn. leaves and the color is slightly blackish green [16]. The pH of *L. camara* Linn. leaf extract is normal as it is in the range of 4.5–6.5. The pH of *L. camara* Linn. leaf extract is consistent with the pH of human skin [29]. Compared to the topical formula, the pH of *L. camara* Linn. leaf extract in this study was under the pH of topical preparations containing ibuprofen [30].

The content of flavonoids, phenols, and tannins in *L. camara* Linn. leaf extract in this study was the same

as the results of previous studies [20, 31, 32]. [33] also shows flavonoid content in the leaves of *L. camara* Linn. in extraction using acetone. In addition, methanol extraction of *L. camara* Linn. leaves also showed flavonoid content [32]. In addition, the extract drying method of *L. camara* Linn. leaves also shows flavonoids and tannins [34]. The results of another study demonstrated that the leaves of *L. camara* Linn. extracted using petroleum ether (40°C), chloroform, and methanol also contain flavonoids and tannins [35].

The QEF levels of *L. camara* Linn. leaf extract in this study was lower than the results of other studies [31, 32, 36, 37]. The results of previous studies demonstrated that various varieties of *L. camara* Linn. have QEF content ranging from 16.14 ± 0.21 to 25.22 ± 2.59 mg/g extract [36]. The results of another study showed the QEF content in the dry extract of *L. camara* Linn. 12.44 ± 2.85 mg/g [37]. Another study showed that the methanol extract of *L. camara* Linn. leaves contained QEF 243.89 mg/g extract [31]. The results of another study demonstrated that several fractions of methanol extract of *L. camara* Linn. leaves contained QEF ranging from 19.85 to 97.56 mg/g samples [32]. The results of other studies also revealed that the QEF content of the methanol extract of aerial parts of *L. camara* Linn. from Nepal ranged from 1.87 ± 0.16 to 0 mg/g extract [38]. On the other hand, the results of other studies demonstrate that the ethanol extract of *L. camara* Linn. leaves contains low QEF, which is 0.2423 ± 0.0068 mg/g extract [39]. These results are lower than the QEF content in our study.

GAEP levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies [31, 32, 36–38]. Previous research demonstrated that various varieties of *L. camara* Linn. have GAEP content ranging from 55.57 ± 2.82 to 232.99 ± 15.93 mg/g extract [36]. Other research results also showed that the dry extract of *L. camara* Linn. contains GAEP 144.7 ± 1.34 mg/g [37]. Another study showed that the methanol extract of *L. camara* Linn. leaves contained GAEP 563.57 ± 2.49 mg/g extract, while the GAEP content in flower extract was 614.79 ± 1.54 mg/g extract [31]. The results of another study demonstrated that the GAEP content of *L. camara* Linn. leaf extract was 10.20 ± 0.343 mg/g extract [38]. The results of another study demonstrated the GAEP content in various fractions of methanol leaf extract of *L. camara* Linn. ranging from 20.25 ± 0.41 to 98.81 ± 0.27 mg/g sample [32]. As a reference, the results of research on the content of GAEP in other plants turned out to vary, for example, *Ageratina adenophora* contains GAEP 4.70 ± 0.059 mg/g extract, while *Cupressus sempervirens* contains GAEP 4.31 ± 0.147 mg/g extract [38].

The TAET levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies. The study results demonstrated that the tannin content in *L. camara* Linn. leaf extract was 98.40 ± 6.88 mg/g

[40]. The results of another study showed that the tannin content of *L. camara* Linn. extract was 0.860 ± 0.038 mg/g [41]. On the other hand, some research results demonstrate that the ethanol extract of *L. camara* Linn. leaves contains low tannins in extract, namely 0.2179 ± 0.0056 mg/g [39]. These results were lower than the tannin content in our study. Some studies also demonstrate that tannin levels from the methanol extract of *L. camara* Linn. collected from a semi-arid region of Brazil are not detected [42, 43].

Note the research results demonstrate that the content of GAEP and QEF in *L. rhodesiensis* extract is highest in the leaves and then the stem, while the least found in the roots [44]. Other research results are the estimation of phenolics, flavonoids, and tannin contents in various solvent extracts of coconut. The results showed that the methanol fraction contained a total phenolic equivalent of gallic acid 822.60 ± 16.36 mg/g sample, a flavonoid equivalent of quercetin 103.30 ± 9.78 mg/g sample, and a tannic acid equivalent of tannin 663.50 ± 19.26 mg/g sample [45].

Based on the data above, there are variations in QEF, GAEP, and TAET levels influenced by variations in plants, environment, and solvents used for extraction. The research results showing that extraction conditions affect flavonoid levels [46] reinforce our statement. Based on the results of these studies as well as other studies [47], flavonoid content was measured in plant extracts [48, 49] and herbal preparations [50, 51, 52, and 53].

The limitations of this study include not examining the mineral content, which can affect the levels of active substances in *L. camara* Linn. leaf extract. Therefore, we suggest for research that it is necessary to measure mineral levels, especially Fe and Zn, which relate to the levels of substances in *L. camara* Linn. leaf extract. The levels of these two minerals affected the stability of QEF levels in *L. camara* Linn. leaf extract cream [16].

5. Conclusion

The ethanolic extract of *L. camara* Linn. contains levels of QEF, GAEP, and TAET as well as 0.428 ± 0.004 mg/g extract, 0.288 ± 0.002 mg/g extract, and 0.384 ± 0.009 mg/g extract, respectively. The content of active substance levels can apply as a reference to explore *L. camara* Linn. as a source of quercetin, gallic acid, and tannic acid.

Based on the results of our research and other studies, and as described above, it turns out that the leaf extract of *L. camara* Linn. contains QEF, GAEP, and TAET, but levels vary. The type and place of life influence the variation of QEF, GAEP, and TAET levels in *L. camara* Linn. leaf extract. Nonetheless, we hope that the variations in QEF, GAEP, and TAET levels in *L. camara* Linn. can be used as an option for the exploration and use of *L. camara* Linn. Thus, *L. camara* Linn. is not only a wild plant that endangers

the environment but can also apply as a source of QEF, GAEP, and TAET exploration.

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