Research Article

The potential of *Lantana camara* Linn. from North Sulawesi Province as a source of flavonoids, gallic acid, and tannic acid

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ABSTRACT:

L. camara Linn. as a plant that is invasive and considered a dangerous plant, but contains active substances that are beneficial to health. Active substances contained in the leaves of *L. camara* Linn. including flavonoids, gallic acid, and tannic acid. The purpose of this study was to explore the content of flavonoids, gallic acid, and tannic acid in *L. camara* Linn leaf extract. Ethanol extract of *L. camara* Linn. leaves were tested organoleptic, pH, flavonoid equivalent quercetin (FEQ), phenolic equivalent gallic acid (PEGA), and tannin equivalent tannic acid (TETA). Measurement of flavonoid equivalent quercetin (FEQ), phenolic equivalent gallic acid (PEGA), and tannin equivalent tannic acid (TETA) levels was carried out with a spectrophotometer. The FEQ content in ethanol extract of *L. camara* Linn. leaves 0.428 ± 0.004 mg / g extract. The content of phenolic equivalent gallic acid (PEGA) in ethanol extract of *L. camara* Linn. leaves is 0.288 ± 0.002 mg / g extract, while the tannin equivalent tannic acid (TETA) content is 0.384 ± 0.009 mg / g extract. The content of active substance levels can be used as a reference to explore *L. camara* Linn. as a source of quercetin, gallic acid, and tannic acid.

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

抽象的
L. camara Linn。 作为一种侵入性植物，被认为是危险植物，但含有对健康有益的活性物质。L. camara Linn 叶子中含有的活性物质。 包括类黄酮、没食子酸和鞣酸。本研究的目的是探讨 L. camara Linn 叶提取物中黄酮类化合物、没食子酸和单宁酸的含量。L. camara Linn 的乙醇提取物。 对叶子进行了感官测试、pH 值、类黄酮等效槲皮素 (FEQ)、酚类等效没食子酸 (PEGA) 和单宁等效单宁酸 (TETA)。使用分光光度计测量类黄酮等效槲皮素 (FEQ)、酚类等效没食子酸 (PEGA) 和单宁等效单宁酸 (TETA) 水平。L. camara Linn 乙醇提取物中的 FEQ 含量。 叶 0.428 ± 0.004 mg / g 提取物。L. camara Linn 乙醇提取物中酚当量没食子酸 (PEGA) 的含量。 叶中含 0.288±0.002 mg/g 提取物，而单宁当量单宁酸（TETA）含量为 0.384±0.009 mg/g 提取物。活性物质含量水平可作为探索 L. camara Linn 的参考。 作为槲皮素、没食子酸和单宁酸的来源。
关键词: Lantana camara Linn.，感官，类黄酮等效槲皮素，酚类等效没食子酸，单宁等效单宁酸

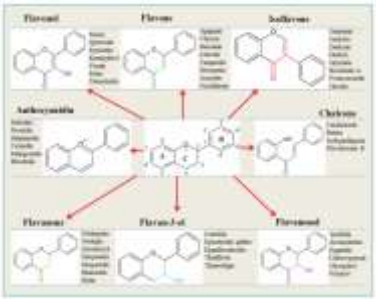
1. Introduction

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

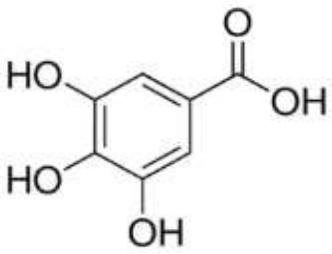
The results of previous studies showed the content of flavonoids, gallic acid, and tannins in the following types of plants. FEQ levels in methanol extract of *Melastoma malabathricum* L. fruit are 6,827 mg/g, while PEGA levels are 154,880 mg/g extract [12]. In addition, it has also been reported that stem bark extract from *M. gigantea* contains flavonoids 25.2 mg/g [13]. It is interesting to note that the content of phenolics catechins is equivalent in various varieties of *Vitis* sp. classified as quite high (>900 mg/L) [14]. The results of other studies showed that various extraction methods against *M. malabathricum* L. shows variation in PEGA levels [15]. The results of this study 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 FEQ levels at various concentrations of *L. camara* Linn. leaf extract cream. *L. camara* Linn. leaf collection was obtained from the area of Tanjakan Cino Mati, Pleret District, Bantul Regency, Special Region of Yogyakarta, Indonesia [16]. *Lantana camara* Linn. is an invasive plant [17], so it is considered a dangerous plant in Indonesia [18]. Several researchers in Indonesia have explored the active ingredients of *L. camara* Linn. to be used in the field of Health [19, 20]. One possible use of *L. camara* Linn. in the field of health, namely 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 to health, we hope that the plant can be used as a source of flavonoids, gallic acid, and tannic acid. Research still needs to be done 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 be used as a reference option about the potential of *L. camara* Linn. as a source of active ingredients in the form of flavonoid equivalent quercetin (FEQ), phenolic equivalent gallic acid (PEGA), and tannin equivalent tannic acid (TETA). The structural formulas of flavonoids, gallic acid, and tannic acid are presented in **Figure 1.**



A.



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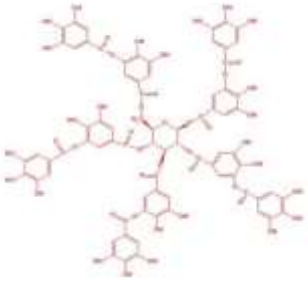

C.

Figure. 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. Structure of tannic acid [22].

2. Material and Methods

2.1. Leaves collection of *L. camara* Linn.

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). The location can be accessed at http://goo.gl/maps/nc1SVYhFU39q8nMz8. The activity was carried out in December 2022. The collected leaves are then washed under running water, then covered with a black cloth, and dried in the hot sun. Leaves of *L. camara* Linn. which had been dried, ground into powder, then sifted to obtain a fine powder. Fine powder of *L. camara* Linn. leaves it is further extracted using 96% ethanol. *L. camara* Linn. leaf extract obtained in a viscous form, dark green in color, then put into sterile bottles, and stored in a refrigerator. The extracts it is ready to be tested.

2.2. Organoleptic, and pH test of *L. camara* Linn. leaf extract

Organoleptic tests performed on *L. camara* Linn. leaf extracts include shape, smell, and color. In addition, pH measurements were also carried out on *L. camara* Linn. leaf extracts [23, 24, 25].

2.3. Qualitative test of flavonoids, gallicacid, and tanic acid in *L. camara* Linn. leaf extract.

2.3.1. Qualitative test of flavonoids

Fifty mg of sample was dissolved with 5 mL ethanol in a test tube, then heated for five minutes. Next, add a few drops of concentrated HCl, then add 0.2 g of Mg powder. A positive result is indicated by the onset of dark red color for 3 minutes.

2.3.2. Qualitative test of phenolic

One mL of sample is dissolved in a test tube containing methanol, then 5% $FeCl_3$ is added. A positive result in the presence of phenolic compounds is indicated by a change in color to orange-brown.

2.3.3. Qualitative test of tannin

Fifty mg of sample was put into a test tube, then added ethanol until the sample was submerged, then added 2-3 drops of 1% $FeCl_3$ solution. A positive result for tannin content is indicated by the formation of a bluish black or green color.

2.4. Quantitative measurement of phytochemical in *L. camara* Linn. leaf extract.

2.4.1. Measurement of flavonoid levels

Measurement of FEQ levels was carried out with aluminum chloride colorimetric assay [26], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). The standard curve of FEQ is 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, then added 0.30 mL 5% NaNO2, and homogenized, then allowed to stand for 5 minutes. Next, add 0.3 mL to 10% $AlCl_3$, and homogenize 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 at a wavelength of 510 nm. The data obtained were used to create a standard curve of flavonoid quercetin equivalent. To measure FEQ levels in the samples, it is done by making a sample solution, namely 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution is reacted with the same reagents used in standard curve making, as well as absorbance readings. Calculation of total FEQ levels by comparing the absorbance of the sample against the standard quercetin curve, the results are expressed as FEQ in mg/g sample.

2.4.2. Measurement of gallic acid levels

Phenolic content measurement was performed with Folin-Ciocalteu assay [26, 27], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). Standard gallic acid curves are

duplicated in volumetric flask. The concentration of gallic acid used is 5, 10, 15, 20, 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 minutes later added 2 mL of 7% Na_2CO_3 solution and 3.6 mL distilled water, then incubated for 90 minutes at room temperature. Absorbance readings with a spectrophotometer at a wavelength of 650 nm. To measure PEGA levels in samples, it is done by making a sample solution, namely 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution is reacted with the same reagents used on the standard curve, as well as the absorbance readings. Total PEGA content was expressed as mg/g sample.

2.4.3. Measurement of tannic acid levels

Measurement of tannin content was performed with Folin-Ciocalteu assay [28], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). Standard tannic acid curves are duplicated in volumetric flask. The concentration of tannic acid used is 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 is put into a flask container containing 7.5 mL of distilled water. To the flask is added 0.5 mL of Follin Denish reagent, allowed to stand 3 minutes, then added 1 mL of saturated Na_2CO_3 solution, then incubated for 15 minutes. Absorbance readings with a spectrophotometer at a wavelength of 740 nm. To measure TETA levels in samples, it is done by making a sample solution, which is 1 mL of *L. camara* Linn. leaf extract instead of standard solutions. The sample solution is reacted with the same reagents used on the standard curve, as well as the absorbance readings. Total TETA content was expressed as mg/g sample.

2.5. Data Analysis

Descriptive analysis was carried out on phytochemical data of *L. camara* Linn leaf extract, namely FEQ, PEGA, and TETA levels. The phytochemical content of *L. camara* Linn. leaf extract presented in table and graphic form using the Microsoft Excel program.

3. Results

3.1. Leaves of *L. camara* Linn.

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) presented in **Figure 1.**

A. B.

Figure 1. *L. camara* Linn. A. *L. camara* Linn. as a wild plant, showing stems, leaves, flowers, and fruit. B. Leaves of *L. camara* Linn. Photographer by Hosea Jaya Edy, December 20, 2022.

3.2. Test results organoleptic, and pH test of *L. camara* Linn. leaf extract

The results of organoleptic test, and pH of *L. camara* Linn. leaf extract is presented in **Table 1.**

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

Type of test	Results
Organoleptic	
• **Shape**	Semi solid
• **smell**	Typical smell of *L. camara* Linn. leaf extract
• **color**	slightly blackish green
pH	5

Abbreviation: pH=potential of hydrogen

The results of qualitative examination of the active substance of *L. camara* Linn. leaf extract is presented in **Table 2.**

Tabel 2. Kadar zat secara kualitatif ekstrak daun *L. camara* Linn.

Compounds tested	Color change results	Result
Flavonoid	Brick red	+
Fenolik	Orange brown	+
Tannin	Greenish-brown transparan	+

Abbreviation: + = positive

3.3. Flavonoid equivalent quercetin

The standard curve of flavonoid equivalent quercetin is presented in **Figure 2**.

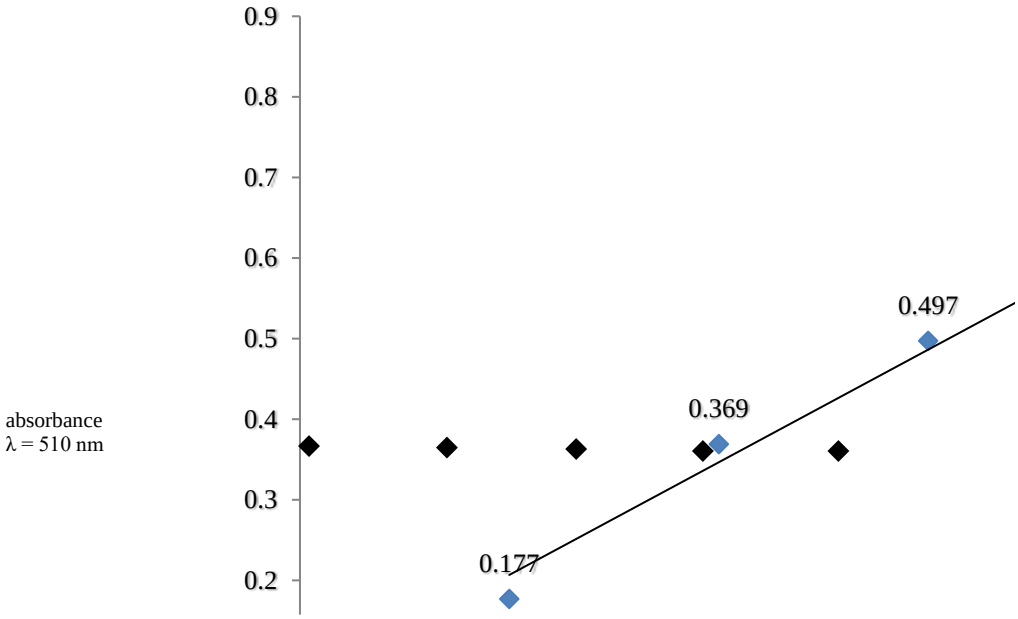

Figure 2. The standard curve of quercetin equivalent flavonoid.

The standard curve used for the analysis of FEQ levels in this study was Y=0.1398q+0.0668 (**Fig. 2**). In the equation, Y=absorbance; a=0.1398; b=0.0668; q=flavonoid equivalent quercetin (mg/L) levels. In addition, a standard solution with a concentration of 1.0-10.0 µg/mL obtained a coefficient of determination (R^2)=0.983. To calculate total of FEQ (Q_{FEQ}) per gram of *L. camara* Linn. leaf extract, we used formula Q_{FEQ}=q*v*(p/m). In the equation, q=FEQ levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

FEQ levels of *L. camara* Linn. leaf extract presented in **Table 3.**

Table 3. Flavonoid equivalent quercetin levels of *L. camara* Linn leaf extract.

Sample	Y	a	b	v (L)	p	m (g)	Q_{FEQ} (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**

Abbreviations: Y=absorbance at a wavelength (λ) 510 nm; a=coefficient; b=constant; v=volume (liters); p=dilution; m=sample weight (gram); q_{FEQ}=flavonoid equivalent quercetin levels; Q_{FEQ}=total flavonoid equivalent quercetin; mg/L=milligrams per liter; mg/g=milligrams per gram; SD=standard of deviation.

3.4. Phenolic equivalent gallic acid

The standard curve of PEGA is presented in **Figure 3.**

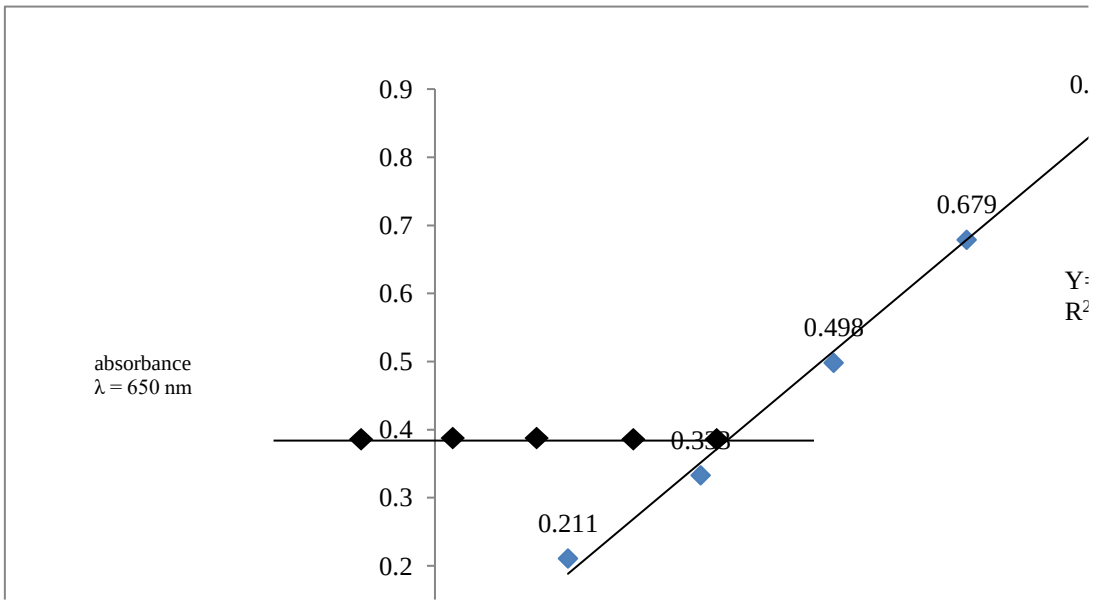

Figure 3. The standard curve of phenolic equivalent gallic acid

The standard curve of PEGA used in this study is Y=0.1634q+0.025 (**Fig. 3**). In the equation, Y=absorbance; a=0.1634; b=0.025; q_{PEGA}=phenolic equivalent gallic acid (mg/L). In addition, a standard solution with a concentration of 5.0-25.0 µg/mL obtained a coefficient of determination (R^2)=0.995. To calculate the amount of PEGA (Q_{PEGA}) per gram of *L. camara* Linn. leaf extract, we used formula $Q_{PEGA}=q*v*(p/m)$. In the equation, q_{PEGA}=PEGA levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

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

Table 4. Levels of phenolic equivalent gallic acid 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**

Abbreviations: Y=absorbance at a wavelength (λ) 650 nm; a=coefficient; b=constant; v=volume (liters); p=dilution; m=sample weight (gram); q $_{PEGA}$=phenolic equivalent to gallic acid levels; Q_{FEGA}=total phenolic equivalent gallic acid in the sample; mg/L=milligrams per liter; mg/g=milligrams per gram; SD=standard of deviation.

3.5. Tannin equivalent tannic acid

The standard curve of TETA is presented in **Figure 4.**

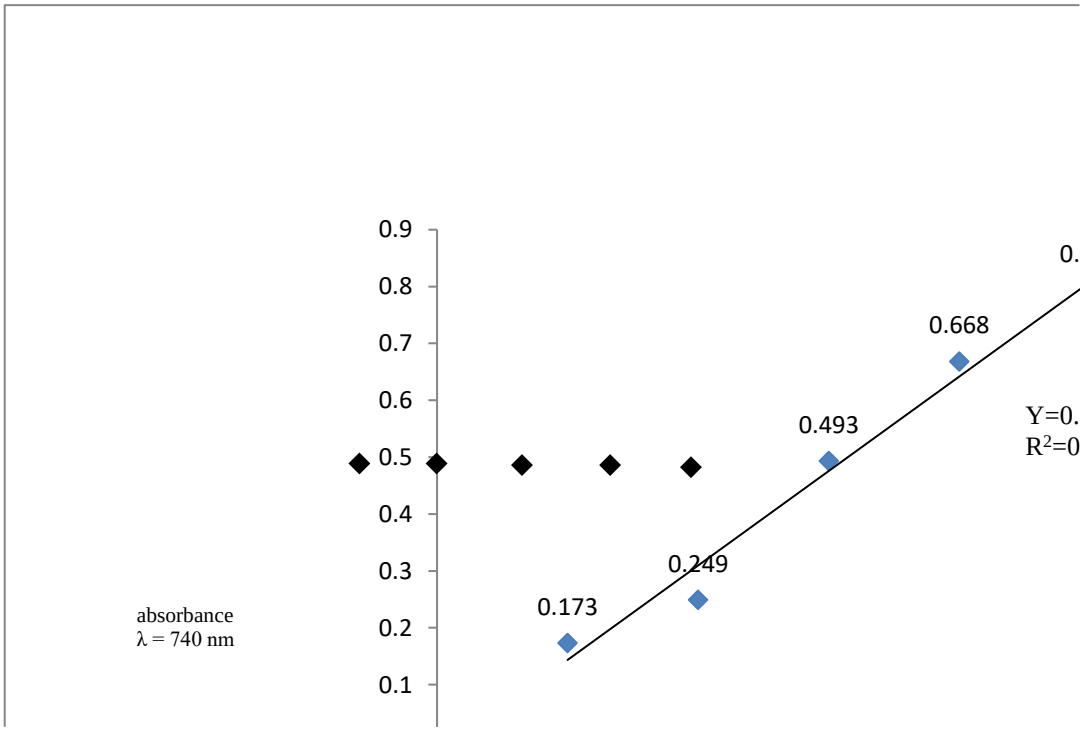

Figure 4. The standard curve of tannin equivalent tannic acid

The standard curve for TETA used in this study is Y=0. 00921q+0.092. The coefficient of determination (R^2) in the equation is 0.984. (**Fig. 4**). In the equation, Y=absorbance; a=0.1661; b=0.0229; q=TETA levels (mg/L). To calculate the amount of TETA (**Q_{TETA}**) per gram of *L. camara* Linn. leaf extract, we used formula Q_{TETA}=q*v*(p/m). In the equation, q=TETA levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

Tannin equivalent tannic acid levels in the *L. camara* Linn. leaf extract is presented in **Table 5.**

Tabel 5. Tannin equivalent tannic acid levels in the *L. camara* Linn. leaf extract

Sample	Y	a	b	v (L)	p	m (g)	Q_{TETA}
							(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**

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

The phytochemical profile of *L. camara* Linn. leaf extract is presented in **Figure 5.**

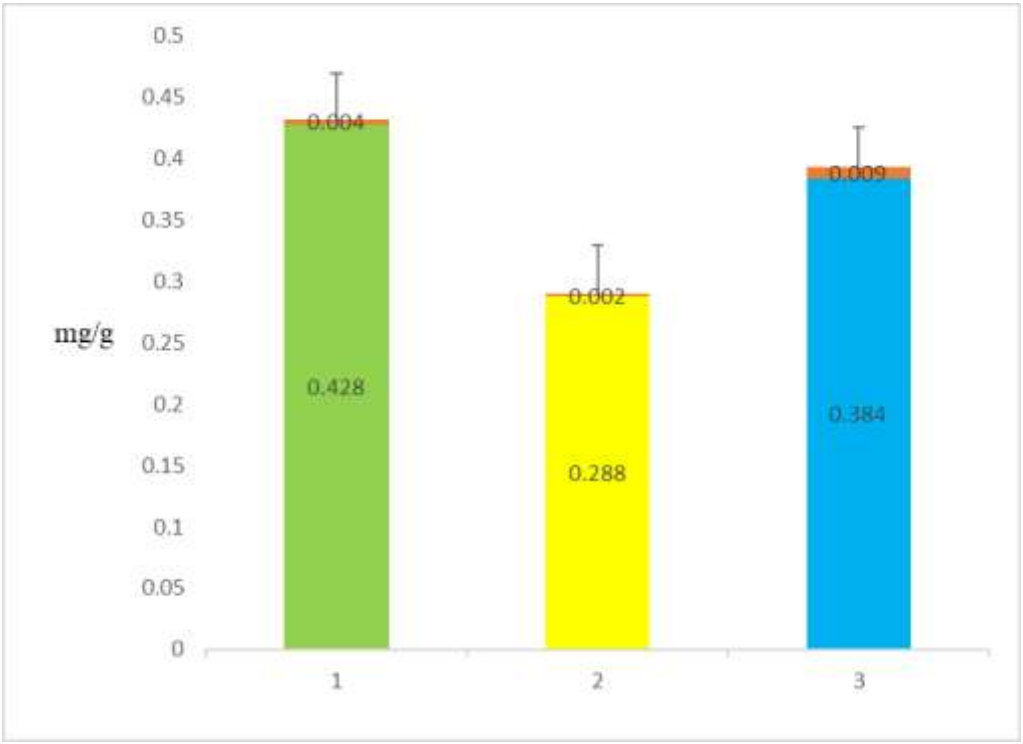

Figure 5. Phytochemical profile of *L. camara* Linn. leaf extract. 1=FEQ (flavonoid equivalent quercetin). 2=PEGA (phenolic equivalent gallic acid). 3=TETA (tannin equivalent tannic acid).

4. Discussion

The results of organoleptic tests on *L. camara* Linn. leaf extract in this time similar with the results of our previous research, including the form is semi-solid, the smell is similar with 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 values of 4.5-6.5. The pH of *L. camara* Linn. leaf extract consistent with pH of human skin [29]. Compared to the topical formula, the pH of *L. camara* Linn. leaf extract in this study was in accordance with 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]. Flavonoid content in the leaves of *L. camara* Linn. also shown with extraction using acetone [33]. 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].

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

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

TETA levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies. The results of the study demonstrated that the tannin content in L. *camara* Linn. leaf extract. 98.40±6.88 mg/g [40]. The results of another study demonstrated that the tannin content of *L. camara* Linn. extract. 0.860±0.038 mg/g [41]. On the other hand, there are research results that demonstrate that ethanol extract of *L. camara* Linn. leaves contain low tannins, namely 0.2179 ± 0.0056 mg/g extract [39]. These results were lower than the tannin content in our study. There are also studies demonstrating that tannin levels from methanol extract of *L. camara* Linn. collected from a semi-arid region of Brazil is not detected [42, 43].

It is noteworthy that there are research results demonstrate that the content of PEGA and FEQ in *L. rhodesiensis* extract is highest in the leaves, then the stem, while the least is found in the roots [44]. Other research results to note are about estimation of phenolics, flavonoids and tannin contents in various solvent extracts of coconut. The results showed that 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 tannin equivalent tannic acid 663.50±19.26 mg/g sample [45].

Based on the data above, there are variations in FEQ, PEGA, and TETA levels that are influenced by variations in plants, environment, and solvents used for extraction. Our statement is reinforced by research results showing that extraction conditions affect flavonoid levels [46]. Based on the results of these study as well as the results of other studies [47], flavonoid content was measured in plant extracts [48, 49], and herbal preparations [50, 51, 52].

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 are related to the levels of substances

in *L. camara* Linn. leaf extract. The levels of these two minerals have been shown to affect the stability of FEQ levels in *L. camara* Linn leaf extract cream [16].

Based on the results of our research, also the results of other studies, and we have described above, it is shown that the leaf extract of *L. camara* Linn. contains FEQ, PEGA, and TETA, but levels vary. Variation of FEQ, PEGA, and TETA levels in *L. camara* Linn leaf extract. This is influenced by the type and place of life. Nonetheless, we hope that the variations in FEQ, PEGA, and TETA levels in the *L. camara* Linn. can be used as an option in the exploration and utilization of *L. camara* Linn. Thus *L. camara* Linn. not only considered as a wild plant that endangers the environment, but can be used as a source of FEQ, PEGA, and TETA exploration.

5. Conclusion

The ethanolic extract of *L. camara* Linn. contains levels of FEQ, PEGA, and TETA 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 be used as a reference to explore *L. camara* Linn. as a source of quercetin, gallic acid, and tannic acid.

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Conflict of Interest

No conflict of interest.

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Subject: Paper Submission
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Title of your paper: The potential of Lantana camara Linn. as a source of quercetin, gallic acid, and tannic acid
Corresponding Author's Email Address: edy.parwanto@gmail.com
Author(s): Edy Parwanto, David Tjahyadi, Husnun Amalia, Hosea Jaya Edy, Ashaolu Victoria Oladimeji, Joey Joshua Vidova Tjahyadi, Laurentia Gabrielle
Keywords: Lantana camara Linn., organoleptic, quercetin equivalent flavonoid, gallic acid equivalent phenolic, tannic acid equivalent tannin
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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. as a plant that is invasive and considered a dangerous plant, but contains active substances that are beneficial to health. Active substances contained in the leaves of *L. camara* Linn. including 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 including 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 that the variations in active substances levels, that is can be used as an option in the exploration and utilization of *L. camara* Linn. Thus, *L. camara* Linn., not only considered as a wild plant that endangers the environment, but can be used as a source for exploration of QEF, GAEP, and TAET.

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

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马缨丹的潜力。 作为槲皮素、没食子酸和单宁酸的来源

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抽象的

L. camara Linn。 作为一种侵入性植物，被认为是危险植物，但含有对健康有益的活性物质。 L. camara Linn 叶子中含有的活性物质。 包括类黄酮、没食子酸和鞣酸。 本研究的目的是探讨 L. camara Linn 叶提取物中槲皮素、没食子酸和单宁酸的含量。 本研究的方法包括 L. camara Linn 的叶提取物。 测试了感官、pH、槲皮素等效类黄酮 (QEF)、没食子酸等效酚类 (GAEP) 和单宁酸等效单宁 (TAET)。 使用分光光度计测量槲皮素等效类黄酮 (QEF)、没食子酸等效酚类 (GAEP) 和单宁酸等效单宁 (TAET) 水平。 L. camara Linn 的 QEF 含量。 叶提取物为 0.428 ± 0.004 mg/g。 L. camara Linn 的 GAEP 内容。 叶提取物为 0.288±0.002 mg/g，而 TAET 的含量为 0.384±0.009 mg/g。 该研究证实了 L. camara Linn 中存在类黄酮、酚类和单宁酸。 叶提取物，用乙醇或其他溶剂（如丙酮或石油醚）提取。 本研究的新颖之处在于活性物质水平的变化，即可以作为探索和利用 L. camara Linn 的一种选择。 因此，L. camara Linn. 不仅被认为是一种危害环境的野生植物，而且可以作为探索 QEF、GAEP 和 TAET 的资源。

关键词: *Lantana camara* Linn.， **感官**， **槲皮素等效**类黄酮，没食子酸等效酚类，单宁酸等效单宁

1. Introduction

Flavonoids are polyphenolic compounds, found in various parts of plants. There are 8 classes of flavonoids, namely: flavones, flavonols, flavanones, flavanonol, isoflavones, flavantriol, anthocyanidins, and chalcone [1]. The benefits of flavonoids in health include being anti-cancer, anti-oxidative, anti-inflammatory, stimulating bone formation [2]. Recent studies have shown that flavonoids have antiviral activity against SARSCoV-2 [3]. Gallic acid is a phenol compound, and is known by another name, 3,4,5-trihydroxybenzoic acid. Gallic acid has the chemical formula (chemical structure) $C_6H_2(OH)_3COOH$ [4]. The results of recent studies demonstrate that gallic acid is produced by *Swietenia macrophylla* [5]. In general, plants produce gallic acid [6]. The benefits of gallic acid in the health sector include anti-microbial, prooxidant, anti-oxidant, anti-inflammatory, anti-platelet, anti-dengue, anti-cancer, and anti-apoptotic [7]. Tannins are phenolic compounds found in plants. There are 2 groups of tannins, namely hydrolysable and condensed tannins. Gallotannins are examples of hydrolysable tannins, while catechins and gallocatechins are examples of condensed tannins [8]. As is the case with flavonoids and gallic acid, tannins are also produced by plants, i.e. *Hibiscus sabdariffa* tea [9], *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, it has also been reported that stem bark extract from *M. gigantea* contains flavonoids 25.2 mg/g [13]. It is interesting to note that the content of phenolics catechins is equivalent in various varieties of *Vitis* sp. classified as quite high (>900 mg/L) [14]. The results of other studies showed that various extraction methods against *M. malabathricum* L. shows variation in GAEP levels [15]. The results of this study 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 the area of Tanjakan Cino Mati, Pleret District, Bantul Regency, Special Region of Yogyakarta, Indonesia [16]. *Lantana camara* Linn. is an invasive plant [17], so it is considered a dangerous plant in Indonesia [18]. Several researchers in Indonesia have explored the active ingredients of *L. camara* Linn. to be used in the field of Health [19, 20]. One possible use of *L. camara* Linn. in the field of health, namely 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 to health, we hope that the plant can be used as a source of flavonoids, gallic acid, and tannic acid. Research still needs to be done 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 be used 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). The structural formulas of flavonoids, gallic acid, and tannic acid are presented in **Figure 1.**

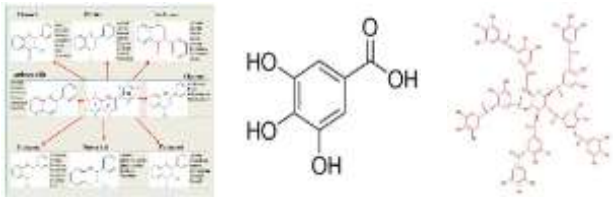

Figure. 1. The structural formula of flavonoids, gallic acid, and tannic acid. A. Basic structure and classification of flavonoids [1]. B. Gallic acid (3,4,5-Trihydroxybenzoic acid) [4, 21]. C. Structure of tannic acid [22].

2. Material and Methods

2.1. Research design

The design of this research is a laboratory experimental research. The main process of these research showed in **Figure 2.**

2.2. Leaves collection of *L. camara* Linn.

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). The location can be accessed at http://goo.gl/maps/nc1SVYhFU39q8nMz8.

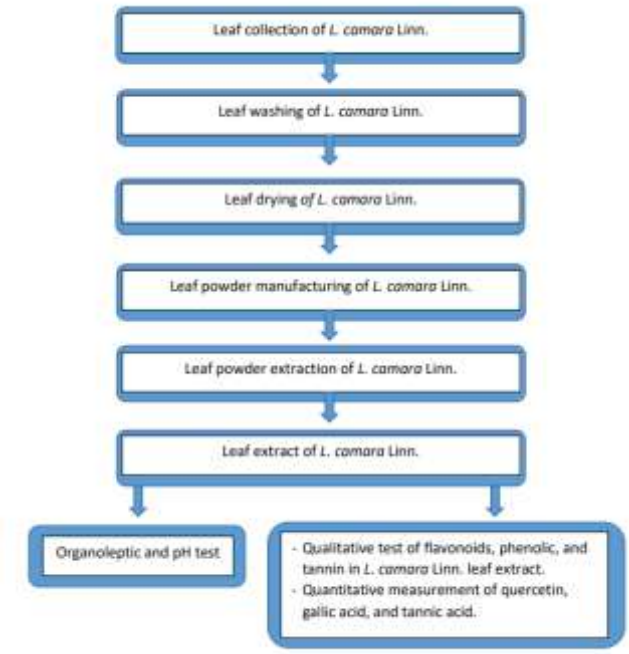

Figure 2. The main process of these research

The activity was carried out in December 2022. The collected leaves are then washed under running water, then covered with a black cloth, and dried in the hot sun. Leaves of *L. camara* Linn. which had been dried, ground into powder, then sifted to obtain a fine powder. Fine powder of *L. camara* Linn. leaves it is further extracted using 96% ethanol. *L. camara* Linn. leaf extract obtained in a viscous form, dark green in color, then put into sterile bottles, and stored in a refrigerator. The extracts it is ready to be tested.

2.3. Organoleptic, and pH test of *L. camara* Linn. leaf extract

Organoleptic tests performed on *L. camara* Linn. leaf extracts include shape, smell, and color. In addition, pH measurements were also carried out on *L. camara* Linn. leaf extracts [23, 24, 25].

2.4. Qualitative test of flavonoids, phenolic, and tannin in *L. camara* Linn. leaf extract.

2.4.1. Qualitative test of flavonoids

Fifty mg of sample was dissolved with 5 mL ethanol in a test tube, then heated for five minutes. Next, add a few drops of concentrated HCl, then add 0.2 g of Mg powder. A positive result is indicated by the onset of dark red color for 3 minutes.

2.4.2. Qualitative test of phenolic

One mL of sample is dissolved in a test tube containing methanol, then 5% $FeCl_3$ is added. A positive result in the presence of phenolic compounds is indicated by a change in color to orange-brown.

2.4.3. Qualitative test of tannin

Fifty mg of sample was put into a test tube, then added ethanol until the sample was submerged, then added 2-3 drops of 1% $FeCl_3$ solution. A positive result for tannin content is indicated by the formation of a bluish black or green color.

2.5. Quantitative measurement of phytochemical in *L. camara* Linn. leaf extract.

2.5.1. Measurement of flavonoid levels

Measurement of QEF levels was carried out with aluminum chloride colorimetric assay [26], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). The standard curve of QEF is 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, then added 0.30 mL 5% NaNO2, and homogenized, then allowed to stand for 5 minutes. Next, add 0.3 mL to 10% $AlCl_3$, and homogenize 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 at a wavelength of 510 nm. The data obtained were used to create a standard curve of flavonoid quercetin equivalent. To measure QEF levels in the samples, it is done by making a sample solution, namely 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution is reacted with the same reagents used in standard curve making, as well as absorbance readings. Calculation of total QEF levels by comparing the absorbance of the sample against the standard quercetin curve, the results are expressed as QEF in mg/g sample.

2.5.2. Measurement of gallic acid levels

Phenolic content measurement was performed with Folin-Ciocalteu assay [26, 27], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). Standard gallic acid curves are duplicated in volumetric flask. The concentration of gallic acid used is

5, 10, 15, 20, 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 minutes later added 2 mL of 7% Na_2CO_3 solution and 3.6 mL distilled water, then incubated for 90 minutes at room temperature. Absorbance readings with a spectrophotometer at a wavelength of 650 nm. To measure GAEP levels in samples, it is done by making a sample solution, namely 1 mL of *L. camara* Linn. leaf extract as a substitute for standard solutions. The sample solution is reacted with the same reagents used on the standard curve, as well as the absorbance readings. Total GAEP content was expressed as mg/g sample.

2.5.3. Measurement of tannic acid levels

Measurement of tannin content was performed with Folin-Ciocalteu assay [28], using the UV-1800 spectrophotometer (Shimadzu Corp. 00787, Serial No. 116351). Standard tannic acid curves are duplicated in volumetric flask. The concentration of tannic acid used is 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 is put into a flask container containing 7.5 mL of distilled water. To the flask is added 0.5 mL of Follin Denish reagent, allowed to stand 3 minutes, then added 1 mL of saturated Na_2CO_3 solution, then incubated for 15 minutes. Absorbance readings with a spectrophotometer at a wavelength of 740 nm. To measure TAET levels in samples, it is done by making a sample solution, which is 1 mL of *L. camara* Linn. leaf extract instead of standard solutions. The sample solution is reacted with the same reagents used on the standard curve, as well as the absorbance readings. Total TAET content was expressed as mg/g sample.

2.6. Data Analysis

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

3. Results

3.1. Leaves of L. camara Linn.

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) presented in **Figure 3.**

A. B. C.

Figure 3. *L. camara* Linn. A. *L. camara* Linn. as a wild plant. B. Habitus of *L. camara* Linn. showing stems, leaves, flowers, and fruit. B. Leaves of *L. camara* Linn. as an extraction material. Photographer by Hosea Jaya Edy, December 20, 2022.

3.2. Test results organoleptic, and pH test of *L. camara* Linn. leaf extract

The results of organoleptic test, and pH of *L. camara* Linn. leaf extract is presented in **Table 1.**

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

Type of test	Results
Organoleptic	
• **Shape**	Semi solid
• **smell**	Typical smell of *L. camara* Linn. leaf extract
• **color**	slightly blackish green
pH	5

Abbreviation: pH=potential of hydrogen

The results of qualitative examination of the active substance of *L. camara* Linn. leaf extract is presented in **Table 2.**

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

Compounds tested	Color change results	Result
Flavonoid	Brick red	+
Fenolik	Orange brown	+
Tannin	Greenish-brown transparan	+

Abbreviation: + = positive

3.3. Quercetin equivalent flavonoid

The standard curve of quercetin equivalent flavonoid is presented in **Figure 4**.

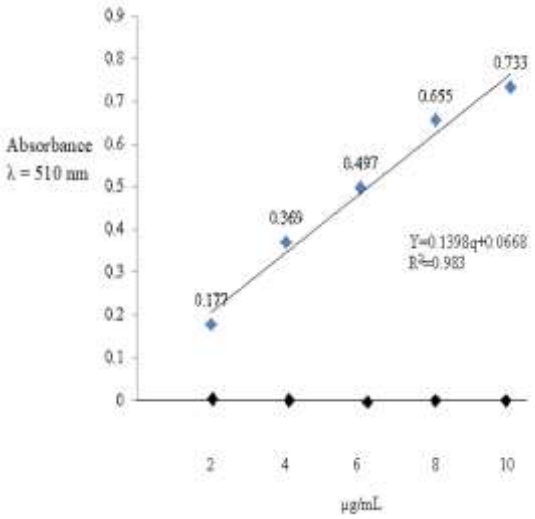

Figure 4. The standard curve of quercetin equivalent of flavonoid.

The standard curve used for the analysis of QEF levels in this study was Y=0.1398q+0.0668 (**Fig. 4**). In the equation, 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 µg/mL obtained a coefficient of determination (R^2) =0.983. To calculate total of QEF (Q_{QEF}) per gram of *L. camara* Linn. leaf extract, we used formula Q_{QEF}=q*v*(p/m). In the equation, q=QEF levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

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

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**

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

3.4. Phenolic equivalent gallic acid

The standard curve of GAEP is presented in **Figure 5.**

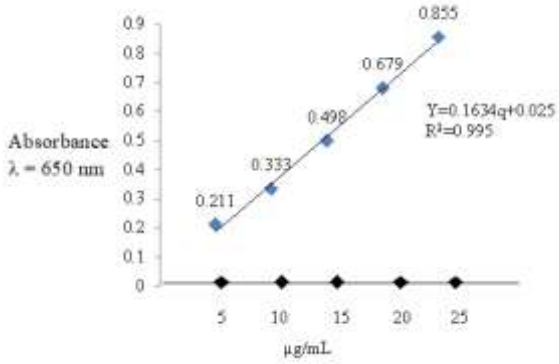

Figure 5. The standard curve of gallic acid equivalent of phenolic

The standard curve of GAEP used in this study is Y=0.1634q+0.025 (**Fig. 5**). In the equation, Y=absorbance; a=0.1634; b=0.025; q $_{GAEP}$=gallic acid equivalent phenolic (mg/L). In addition, a standard solution with a concentration of 5.0-25.0 µg/mL obtained a coefficient of determination (R^2) =0.995. To calculate the amount of GAEP (Q_{GAEP}) per gram of *L. camara* Linn. leaf extract, we used formula Q_{GAEP}=q*v*(p/m). In the equation, q $_{GAEP}$=GAEP levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

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

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**

Abbreviations: 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 of deviation.

3.5. Tannin equivalent tannic acid

The standard curve of TAET is presented in **Figure 6.**

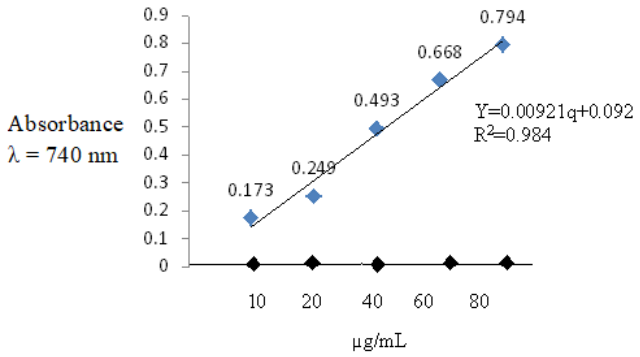

Figure 6. The standard curve of tannin equivalent tannic acid.

The standard curve for TAET used in this study is Y=0. 00921q+0.092. The coefficient of determination (R2) in the equation is 0.984. (**Fig. 6**). In the equation, Y=absorbance; a=0.1661; b=0.0229; q=TAET levels (mg/L). To calculate the amount of TAET (QTAET) per gram of *L. camara* Linn. leaf extract, we used formula Q_{TAET}=q*v*(p/m). In the equation, q=TAET levels in the sample, v=sample volume, p=dilution, and m=sample mass/weight.

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

Tabel 5. Tannic acid equivalent tannin levels in the *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

Abbreviation: Y=absorbance in wave length (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 of deviation.

The phytochemical profile of *L. camara* Linn. leaf extract is presented in **Figure 7.**

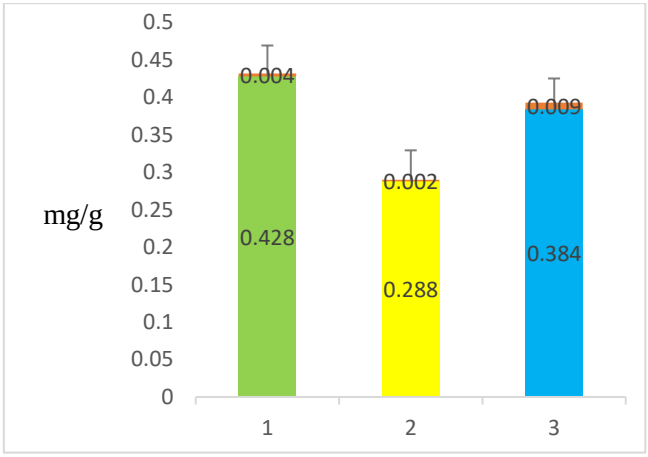

Figure 7. Phytochemical profile of *L. camara* Linn. leaf extract. 1=QEF (quercetin equivalent flavonoid). 2=GAEP (phenolic equivalent gallic acid). 3=TAET (tannin equivalent tannic acid).

4. Discussion

The results of organoleptic tests on *L. camara* Linn. leaf extract in this time similar with the results of our previous research, including the form is semi-solid, the smell is similar with 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 values of 4.5-6.5. The pH of *L. camara* Linn. leaf extract consistent with pH of human skin [29]. Compared to the topical formula, the pH of *L. camara* Linn. leaf extract in this study was in accordance with 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]. Flavonoid content in the leaves of *L. camara* Linn. also shown with extraction using acetone [33]. 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].

QEF levels of *L. camara* Linn. leaf extract in this study were 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 that the content of QEF in 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 contain 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 19.85-97.56 mg/g samples [32]. The results of other studies also revealed that the QEF content of methanol extract of aerial parts of *L. camara* Linn. from Nepal ranged from 1.87±0.16 0 mg/g extract [38]. On the other hands, the results of other studies demonstrate that ethanol extract of *L. camara* Linn. leaves contain 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, 37, 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 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 contain GAEP 563.57±2.49 mg/g extract, while the GAEP content in flower extract 614.79±1.54 mg/g extract [31]. The results of another study demonstrated that the GAEP content of *L. camara* Linn. leaf extract 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].

TAET levels of *L. camara* Linn. leaf extract in this study were lower than the results of other studies. The results of the study demonstrated that the tannin content in L. *camara* Linn. leaf extract. 98.40±6.88 mg/g [40]. The results of another study demonstrated that the tannin content of *L. camara* Linn. extract. 0.860±0.038 mg/g [41]. On the other hand, there are research results that demonstrate that ethanol extract of *L. camara* Linn. leaves contain low tannins, namely 0.2179 ± 0.0056 mg/g extract [39]. These results were lower than the tannin content in our study. There are also studies demonstrating that tannin levels from methanol extract of *L. camara* Linn. collected from a semi-arid region of Brazil is not detected [42, 43].

It is noteworthy that there are research results demonstrate that the content of GAEP and QEF in *L. rhodesiensis* extract is highest in the leaves, then the stem, while the least is found in the roots [44]. Other research results to note are about estimation of phenolics, flavonoids and tannin contents in various solvent extracts of coconut. The results showed that 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 tannic acid equivalent tannin 663.50±19.26 mg/g sample [45].

Based on the data above, there are variations in QEF, GAEP, and TAET levels that are influenced by variations in plants, environment, and solvents used for extraction. Our statement is reinforced by research results showing that extraction conditions affect flavonoid levels [46]. Based on the results of these study as well as the results of other studies [47], flavonoid content was measured in plant extracts [48, 49], and herbal preparations [50, 51, 52, 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 are related to the levels of substances in *L. camara* Linn. leaf extract. The levels of these two minerals have been shown to affect 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 be used 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, as well as the results of other studies, and we have described above, it turns out that the leaf extract of *L. camara* Linn. contains QEF, GAEP, and TAET, but levels vary. Variation of QEF, GAEP, and TAET levels in *L. camara* Linn. leaf extract. This is influenced by the type and place of life. Nonetheless, we hope that the variations in QEF, GAEP, and TAET levels in the *L. camara* Linn. can be used as an option in the exploration and utilization of *L. camara* Linn. Thus *L. camara* Linn. not only

considered as a wild plant that endangers the environment, but can be used as a source of QEF, GAEP, and TAET exploration.

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Conflict of Interest

No conflict of interest.

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Cover letter

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[2023-05-06]

Dear Editor the Journal of Hunan University Natural Sciences,

I/We wish to submit an original research article entitled "The potential of *Lantana camara* Linn. as a source of quercetin, gallic acid, and tannic acid" for consideration by the Journal of Hunan University Natural Sciences.

I/We confirm that this work is original and has not been published elsewhere, nor is it currently under consideration for publication elsewhere.

In this paper, I/we report on / show that *Lantana camara* Linn. as a source of quercetin, gallic acid, and tannic acid. This is significant because *L.camara* Linn. which is considered a wild plant and harmful to the environment, but can be used as a source of exploration for quercetin, gallic acid, and tannic acid which are beneficial for health.

We believe that this manuscript is appropriate for publication by Journal of Hunan University Natural Sciences because it…

[Based on the facts show that *L.camara* Linn. considered wild plants and harmful to the environment, even though these plants contain active substances that are beneficial to health. Exploitation of wild plants that are harmful to the environment needs to be explored, among others by exploring the content of active substances. *L. camara* Linn. it turns out that it contains the active substances quercetin, gallic acid, and tannic acid which are beneficial to human health. In our opinion, readership will be interested in this, because of the importance of reducing the environmental burden caused by *L. camara* Linn. Moreover, the benefits of the presence of active substances in the form of quercetin, gallic acid, and tannic acid can be obtained].

We have no conflicts of interest to disclose.

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Thank you for your consideration of this manuscript.

Sincerely,

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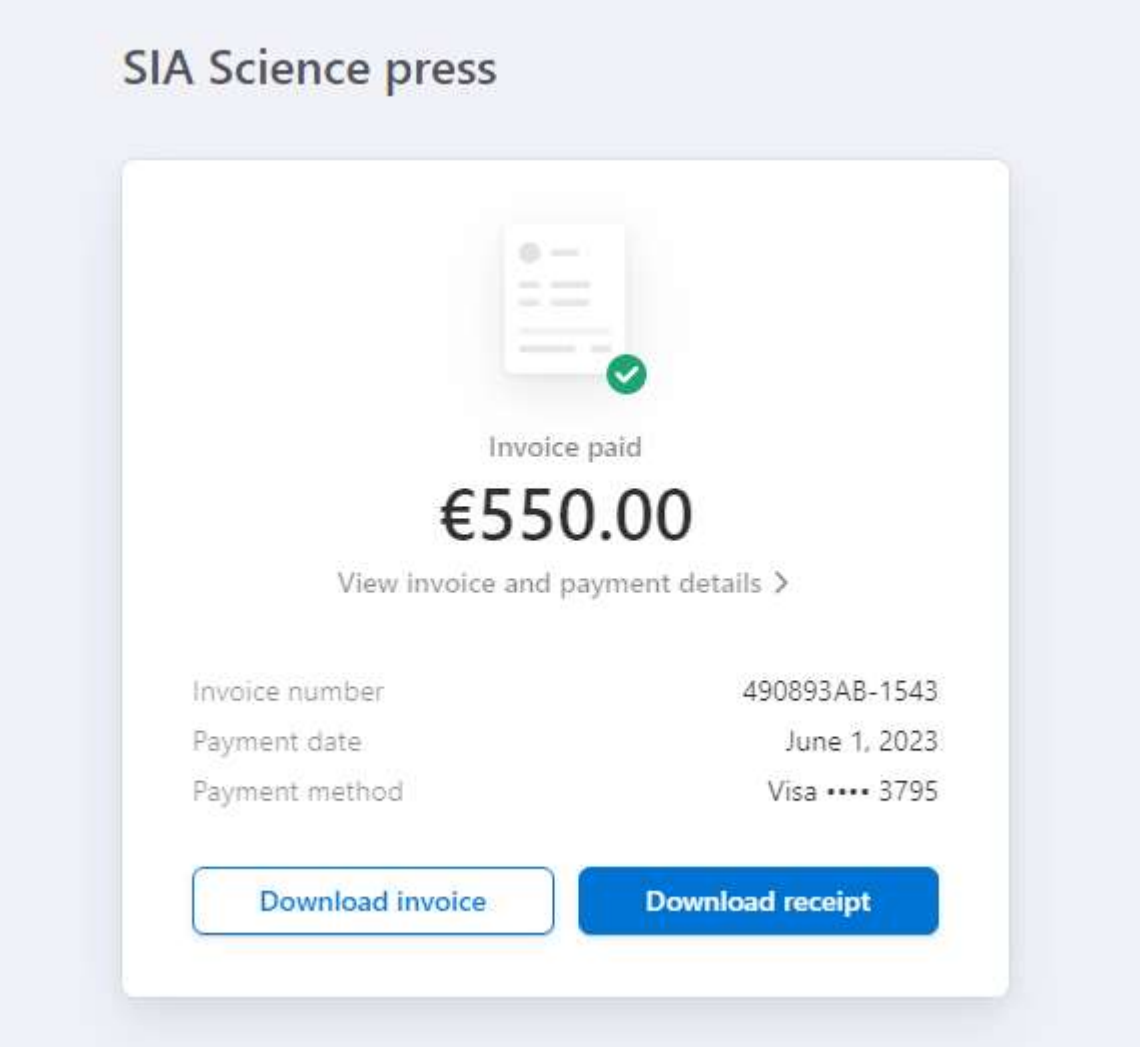

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