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Establishment of a standardized ethanolic extract of *Avicennia officinalis* leaves in Vietnam

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Abstract. The mangrove plant *Avicennia officinalis* is widespread throughout the subtropical coast and is known for its many pharmacological properties, such as antibacterial, antiulcer, anti-inflammatory, anti-cancer, and antioxidant. This study determined the physicochemical parameters of the standardized extract, qualitative by phytochemical screening and thin layer chromatography (TLC), qualitative total phenolic content, and the content of the marker compound by high-pressure liquid chromatography (HPLC). Compounds such as *p*-coumaric acid and cinnamic acid have been found to be major phenolic acids in the leaves of this plant. These results provided useful information for the evaluation of the quality of *Avicennia officinalis* raw materials and its commercial products. The parameters of the ethanol extract of *Avicennia officinalis* leaves were obtained as follows: the loss of dry weight (10.9±2.09%), total ash content (22.31%), microbiological limits achieved to meet the requirements of medicinal standards, and total heavy metal content not detected. The content of total phenolics determined amounted to 81.83 mg GAE/g extract. The quantification method by HPLC was validated according to AOAC, and the content of two markers in the extract was *p*-coumaric acid (1.19 mg/g) and cinnamic acid (3.82 mg/g). The outputs of this research will be standardized extracts that's the source of medicinal plants in Vietnam.

Keywords: *Avicennia officinalis, standardized extract, phenolic acid, HPLC, and TLC.*

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1. Introduction

The *Avicenniaceae* family contains the important mangrove genus *Avicennia*, which has eight to ten species that have been identified worldwide. *Avicennia officinalis*, a medium-sized mangrove plant, is widespread throughout the subtropical coast and thrives in brackish water (Thirunavukkarasu et al., 2011; Assaw et al., 2020). This plant's stem or bark is used traditionally as diuretic, antiulcer, therapy for snakebite, rheumatism, skin conditions, hepatitis, antitumor, antibacterial, gastroprotective, boils, and abscesses. Pharmacological studies have demonstrated the antibacterial, antiulcer, anti-inflammatory, anti-cancer, and antioxidant activities of several *Avicennia officinalis* leaf and bark extracts (Bhimba et al., 2010; Kumar et al., 2017; Ngoc-Van et al., 2022). The majority of this plant's distribution is in Malaysia, Bangladesh, India, and Indonesia. This species may be found in abundance in Vietnam's mangrove regions, particularly in the coastal provinces of Ca Mau, Kien Giang, and Tra Vinh (Thu et al., 2019; Duong et al., 2022) (**Figure 1**).

In developed nations, there has been a huge increase in the demand for items made from plants in recent years. These goods are being sought after more and more as pharmaceuticals, nutraceuticals, and cosmetics (Kunle et al., 2012). It has become crucial to establish dependable, precise, and sensitive quality control systems employing a combination of traditional and contemporary instrumental ways of analysis in order to have good coordination between the quality of raw materials, process materials, and the final products. For the purpose of guaranteeing the quality control of herbal medications,

standardization is a crucial measurement. The process of prescribing a set of standards or intrinsic qualities, consistent parameters, and unambiguous qualitative and quantitative values that convey an assurance of quality, effectiveness, safety, and repeatability is known as the standardization of herbal medicines. It is the process of developing and agreeing upon technical standards (Garg et al., 2012). Standardization is defined by the American Herbal Product Association as "the body of knowledge and control required to produce material of adequate consistency." This was accomplished by reducing the natural variations in product composition by applying quality assurance procedures to the manufacturing and farming operations (Gaedcke et al., 2004). All actions that are conducted during the production process and quality control that result in reproducible quality are referred to as "standardization" in this context. A standardized extract is a form of extract obtained after the extraction process from batches of medicinal herbs to achieve uniformity with a high bioactive compound content. Bioactive compounds in herbal extracts are enriched during the extraction process and standardized with a number of uniform criteria in all batches.

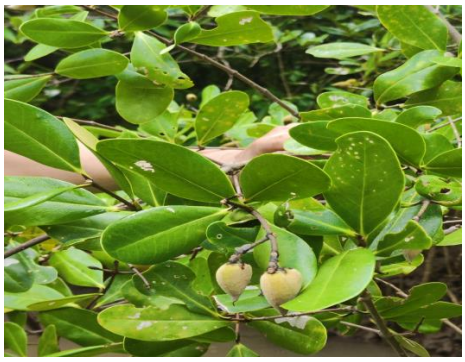

Figure 1. Leaves of *Avicennia officinalis* L. collected in Ca Mau Province, Vietnam

The leaves of *Avicennia officinalis* are now being considered as future herbal resources. Although there are numerous reports on the therapeutic benefits of *Avicennia officinalis* extract, there have only been a few studies to establish standards for this medicinal herb. To the best of our knowledge, there have been no studies conducted in Vietnam to evaluate the standardized extract of *Avicennia officinalis*. This study was conducted to determine the loss of dry weight, total ash content, acid-insoluble ash content, total heavy metal content (Pb), microbial contamination, and the content of marker compounds in the ethanol leaf extract of *Avicennia officinalis*, creating the basis for the use of effective medicinal herbs and contributing to the standard development and quality management of herbal extracts.

2. Materials and Methods

2.1. Chemicals and solvents

Two phenolic acids (*p*-coumaric acid and cinnamic acid) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Ethanol and n-hexane were acquired from Vietnam. Acetonitrile, methanol, and HPLC-grade water were acquired from Honeywell (North Carolina, USA). Ammonium acetate and formic acid were purchased from Merck (Darmstadt, Germany). Folin-Ciocalteu (FC) reagent, sodium carbonate anhydrous, and gallic acid were purchased from Sigma-Aldrich GmbH (Merck, Germany).

2.2. Plant materials

Leaves of fresh *Avicennia officinalis* L. were collected in Ca Mau Province, Vietnam. The leaves were washed with water to remove soil and dirt. The moisture content of the dried sample was lower than 13%. All samples were stored in black glass containers and kept at room temperature.

2.3. Plant extracts

Reflux heating in 800 mL of 80% ethanol at 70 °C extracted the 100-gram leaf sample powder. The sample extract was extracted again in triplicate. Ethanol solvent is recovered by a rotary evaporator. The standardized extract was recovered and stored at 4 °C for use in experiments later.

2.4. Physicochemical parameters

2.4.1. Determination of loss on drying

For 30 minutes, the extract (1 g) was cooked in an oven at 105 °C until the weight was steady. When weighing, make sure the bottle is closed and has had time to reach room temperature in a desiccators (Ministry of Health Vietnam, 2018).

2.4.2. Determination of total ash content

The extract (2 g) was properly weighed before being added to the crucible. The crucible was then gradually lit until the charcoal was removed, cooled, and had a steady weight. Levels of acid-insoluble ash were determined by boiling the ash and calculating the total ash content for 5 minutes in 25 mL of diluted sulfuric acid. The acid-insoluble portion was gathered, filtered through ash-free filter paper, rinsed in hot water, and then ignited to maintain weight. Calculated using the dried material's ash content, which does not dissolve in acid (Ministry of Health Vietnam, 2018).

2.4.3. Determination of total lead (Pb) content

Atomic absorption spectrophotometry was used to determine the amount of lead in the extract. Wet destruction was used to prepare the extract from the material. HNO3 with 10 mL was added to the sample, which was then left for one day. The sample was then heated until white smoke started to emerge, and five drops of $HClO_4$ were then gently added. After filtering the sample solution through filter paper and allowing it to cool, it was diluted with distilled water and put into a 50-mL volumetric flask. Then, a spectrophotometer was used to measure the absorbance (AOAC, 2005).

2.4.4. Microbial contamination

Test the level of contamination with aerobic bacteria and viable molds and detect medical indicator bacteria in the product based on the microbial limit test method. Count the total number of bacteria and molds on agar plates and check for the presence of *E. coli, Staphylococcus aureus, and Pseudomonas aeruginosa* (Ministry of Health Vietnam, 2018).

2.5. Quantitative of phytochemicals in standardized extracts

2.5.1. Phytochemical screening of the extracts

Phytochemical constituents such as flavonoids, alkaloids, saponins, tannins, glycosides, coumarins, anthraquinon, tanins, reducing sugars, carotenoids, and phyosterols in the extracts were performed using standard analytical procedures with slight modifications (Rao et al., 2023).

2.5.2. Thin Layer Chromatography (TLC)

Thin layer chromatography was performed on chromatography silica gel 60 F254 (Merck, Germany), with a 5 µL sample obtained in the thin layer chromatography using chloroform, methanol, and formic acid (46:1.5:2.5 v/v/v) as the mobile phase. Samples were run, dried, and observed under a UV lamp with a wavelength of 254 nm.

2.6. Qualitative of phytochemicals in standardized extracts

2.6.1. Determination of total phenolic content

The determination of the overall phenolic content was conducted by employing the Folin-Ciocalteu reagent. A measurement of absorption was conducted at a wavelength of 760 nm using a UV-VIS spectrophotometer (V-550 model, Jasco, Tokyo, Japan). The obtained values were then compared to the calibration curve of gallic acid. These findings were quantified in milligrams of gallic acid equivalents (GAE) per gram of extract. This study was conducted three times for each assay.

2.6.2. RP-HPLC

Sample preparation. A precisely measured quantity of Avicennia officinalis extract (0.5 g) was added to a 20-mL volumetric flask. The sample underwent sonication, and the volume was modified using MeOH (w/v). The entire set of sample solutions underwent filtration using a 0.45 µm membrane filter and was then transferred into sample vials. Prior to HPLC analysis, the samples were degassed.

Construction of the standard calibration curves. Accurate measurements of cinnamic acid and p-coumaric acid were dissolved in MeOH to create standard stock solutions with a final concentration of 1000 µg/mL. The stock solutions were diluted

in a series to get final concentration ranges of 0.05-100 µg/mL for cinnamic acid and *p*-coumaric acid. Calibration curves were created using five different concentrations as a reference. The experiment was conducted using three replicates for each sample. The chemical structure of cinnamic acid and *p*-coumaric acid is shown in **Figure 2**.

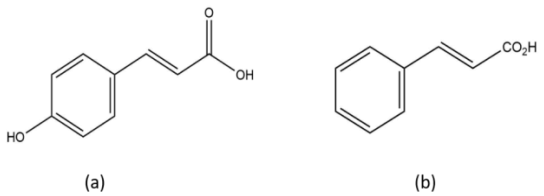

Figure 2. Chemical structures of two markers in *Avicennia officinalis* leaves
(a: *p*-coumaric acid; b: cinnamic acid)

2.6.3. HPLC analysis

HPLC analysis was performed on an UFLC Shimadzu (LC-20AD) instrument with an injection volume of 20 µL and a diode diode array and multiple wavelength detector (DAD SPD-M20A), as well as a reverse phase Luna C18 (5 µm, 4.6 x 250 mm) Phenomenex.

The mobile phase consisted of MeCN (Solvent A), MeOH (Solvent B), and ammonium acetate (0.2%), acid formic (0.1%/H_2O) (Solvent C). The gradient elution program began with 5% B and 95% C for 5 min, 15% B and 75% C for 15 min, 10% B and 75% C for 5 min, 21% B and 68% C for 10 min, and then B was reduced to 5% and C was increased by 95% again for 3 min. The total run time was 38 minutes. The flow rate was 1 mL/min. The detection was made at 280 nm.

2.6.4. Validation of the quantification method

Linearity. The linearity of the calibration curves generated from HPLC analysis was assessed at five concentrations ranging from 0.05 to 100 µg/mL for cinnamic acid and *p*-coumaric acid. Both correlation coefficients were computed using the linear regression technique.

Precision. A specimen was generated using the previously outlined procedure and examined six times on the same day to assess the variability within the day. Additionally, it was studied on three successive days to evaluate the variability between days, resulting in a total of nine measurements. The precision was quantified using the relative standard deviation (RSD %).

Accuracy. The accuracy was tested by separate spiking with a known amount of the standard cinnamic acid (20 µg/mL) and p-coumaric acid (10 µg/mL) for 100%. The accuracy was carried out with three levels of concentration (50%, 100%, and 150%) and analyzed in triplicate. The average recovery was calculated according to the formula:

Recovery (%) = [(net measured amount original amount)/spiked amount] x 100.

Limit of detection (LOD) and limit of quantitation (LOQ). According to the International Conference on Harmonization Guidelines, the limit of detection (LOD) and limit of quantitation (LOQ) were calculated using the standard deviation of the response (r) and the slope of the calibration curve (S). LOQ = 10 * r/S and LOD = 3.3 * r/S (AOAC, 2013).

2.7. Statistical analysis

All of the collected data were entered and analyzed using Microsoft Excel 2016 and SPSS 23.0 software.

3. Results and Discussion

3.1. Physicochemical parameters

The ethanol extract of *Avicennia officinalis* leaf extract was obtained in the form of thick and sticky, dark green, aromatic odor, and bitter taste. The results of the physicochemical parameters of *Avicennia officinalis* leaf extract are shown in Table 1. The standardized extract of *Avicennia officinalis* leaves has averaged 10.9% percent drying, a total ash content of 22.31%, and undetected acid insoluble ash content. Total lead (Pb) content was not detected in *Avicennia officinalis* extract. The microbial contamination levels of *E. coli*, *S. aureus*, *and P. aeruginosa* were lower than 10 CFU/g. The ethanol extract of Moringa leaves averaged 21.18% percent drying, a total ash content of 1.70%, and an acid-insoluble ash content of 0.28% (Mun'im et al., 2016). The difference between these parameters is due to the difference in tree species between the two studies. *Avicennia officinalis* in

this research is a mangrove tree; the leaves are thick and dry, and the amount of water in the leaves is low, leading to a high content of inorganic substances in the leaves and a total ash content that is higher than Moringa leaf extract many times.

Table 1. Physicochemical parameters of *Avicennia officinalis* leaves extract

Parameters	**Results**
The loss of dry weight (%)	10.9 ± 2.09
Total ash content (%)	22.31
Acid-insoluble ash content (%)	Not detected
Heavy metal content (Pb)	Not detected
Escherichia coli (E. coli) (CFU/g)	< 10
Staphylococcus aureus (CFU/g)	< 10
Pseudomonas aeruginosa (CFU/g)	< 10

3.2. Quantitative of phytochemicals in a standardized extract

3.2.1. Phytochemical screening of the extracts

The chemical make-up of the *Avicennia officinalis* leaf extract was screened and found to include flavonoids, alkaloids, saponins, tannins, glycosides, coumarins, anthraquinon, tanins, reducing sugars, carotenoids, and phyosterols (Table 2). The *A. officinalis* leaves standardized extract contained alkaloids, flavonoids, and tannins with strong positive rerults. The saponin and reducing sugars in *Avicennia officinalis* leaf standardized extract had positive results. This result is similar to the previous study by Ganesh and Vennila (2011) analysis of phytochemicals in the methanol extract of *Avicennia officinalis* collected in India.

Table 2. Phytochemical screening of *Avicennia officinalis* leaves extract

Compounds	**Results**
Alkaloids	+++
Glycosides	-
Saponins	+
Flavonoids	+++
Coumarin	-
Anthraquinons	-
Tanins	+++
Reducing sugars	++
Carotenoids	-
Phytosterols	-
Notes: (+): Positive, (++): Clearly positive, (+++): Strong positive, (-): Negative	

3.2.2. Quantitative by TLC

TLC chromatographic conditions are optimized to best separate phenolic acid group compounds. The mobile phase contains a mixture of chloroform, methanol, and formic acid (46:1.5:2.5 v/v/v) that separates the phenolic acid compounds. These compounds were observed under 254 nm UV light for the upper fluorescence bands on the TLC plate. Figure 3 shows the results of the cinnamic acid compound in test and standard samples for light-off chromatographic lines with an Rf value of 0.78.

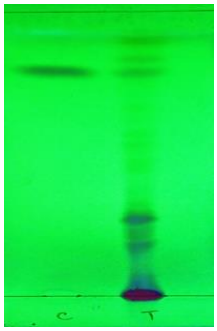

Figure 3. TLC of ethanol leave extract of *Avicennia officinalis*
(C: Cinnamic acid standard sample, T: *Avicennia officinalis* extract sample)

3.3. Qualitative of phytochemicals in standardized extracts

3.3.1. Determination of total phenolic content

The ethanolic extract of *Avicennia officinalis* leaves had 81.83 mg GAE/g extract of total phenolics, which was found using a colorimetric method. The findings of this study's total phenolic content analysis are greater than those of Febriani et al. (2020), who found 23.024 mg GAE/g for leaves, 49.119 mg GAE/g for fruits, and 33.738 mg GAE/g for stem barks isolated from *Avicennia marina* using ethanol. The total phenolic content of *Avicennia rumphiana* was found to be 1.436 mg GAE/g for the ethyl acetate extract of leaves, 1.2431 mg GAE/g for the ethyl acetate extract of bark, and 0.8761 mg GAE/g for the ethanol extract of leaves, according to Sulmartiwi et al. (2018). The ethanol extract of *Avicennia officinalis* collected in India was 257 mg GAE/g (Thirunavukkarasu et al., 2011), which was higher than the total phenolic content in this research.

3.3.2. Validation of the quantification method

System suitability. The stability of the system was evaluated (Rs) by executing six duplicate injections of a mixed standard solution (50 g/mL) and computing the theoretical plate number (N) and resolution. For the peak area and RT, all analytes exhibited RSD values that were less than 2.0%. The recommended strategy therefore met this requirement (Table 3).

Table 3. System suitability of the HPLC-PDA method

		t_R	S	As	Rs	N
***p*-coumaric acid**	Mean	13.15	5801267	1.1	--	35659
	RSD%	1.63	0.77	0.5	--	1.3
Cinnamic acid	Mean	29.46	8699742	1.1	24.9	61410
	RSD%	1.30	1.87	0.8	0.8	1.7

Specificity. The specificity was put to the test by employing the UFLC method to look at *Avicennia officinalis* leaf-standardized extract. It was determined by comparing the peaks of each component's RT and UV absorption spectra in reference solutions to those observed following extract analysis. The UFLC method may be used to extract *p*-coumaric acid and cinnamic acid from other components of the leaf matrix, as shown in Figure 4. The peak purity of the two compounds was >99.9%, as shown by the spectrum overlaid on the three-point purity detection plots.

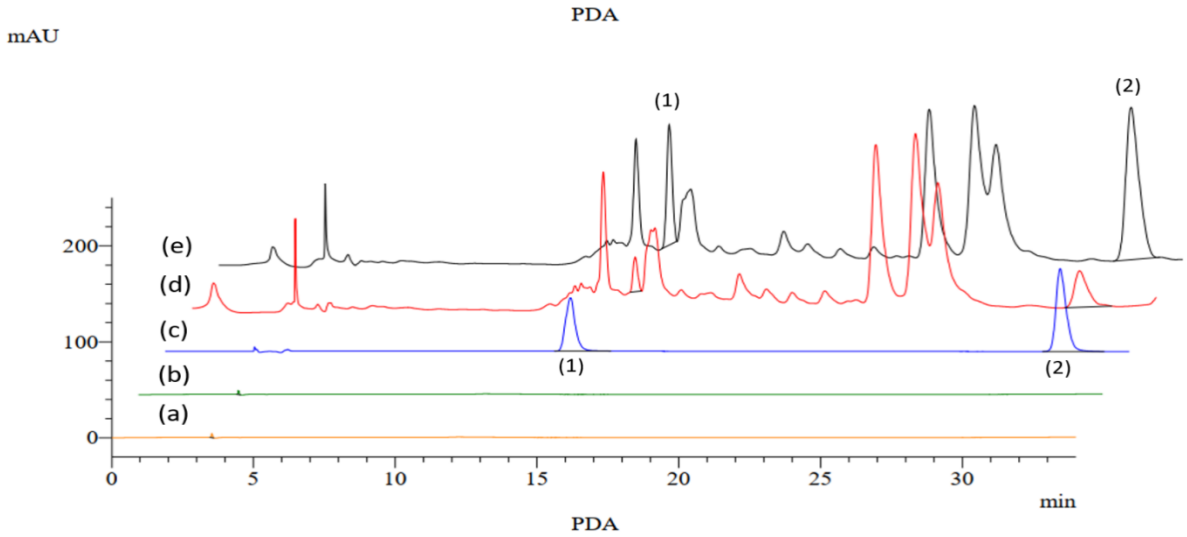

Figure 4. HPLC chromatograms for specificity: Peak (1): *p*-coumaric acid; Peak (2): cinnamic acid (a: Mobile phase solvent; b: Extraction solvent; c: Mixed standard solution of *p*-coumaric acid and cinnamic acid; d: Leaf sample standardized extract; e: Leaf sample extract added standard)

Linearity, the limit of detection, and the limit of quantification. Five different *p*-coumaric acid and cinnamic acid concentrations, ranging from 0.05 to 100 g/mL, were used to dilute and mix the stock solutions. In order to evaluate the linearity, three duplicates of each mixed standard sample were injected into the UFLC system. Calibration curves were then made by

plotting the average of the peak area responses vs. concentration for each sample. For p-coumaric acid and cinnamic acid, the square correlation coefficients (R2) were more than 0.999 (Table 4). The LOD and LOQ were, respectively, 0.025 mg/mL and 0.05 mg/mL.

Precision. The correctness of the approach was verified by comparing the intra-day and inter-day precisions. The relative standard deviation (%RSD) was employed as an accuracy indicator. Each medicine had six samples evaluated to establish intra-day precision and three days of daily analysis to establish inter-day accuracy. The overall intra- and inter-day variations (%RSD) were less than 6%, in accordance with AOAC standards, as shown by the accuracy findings in Table 4.

Accuracy. The accuracy of the approach was examined by recovery trials. Low (50%), medium (100%), and high (150%) amounts of reference substances were applied to the blank samples. Then, following the aforementioned methods, the spiked samples were added and measured. The results demonstrated the devised method's high degree of accuracy, with a total recovery falling within an acceptable range of 80.4% to 92.4% (Table 4). Based on the results of the recovery test, it was believed that the approach was precise.

Table 4. Precision, recovery, calibration parameters

Substance	Calibration curve		Precision		Recovery (%)		
	Regression equation	R^2	Intra-day RSD (%) (n=6)	Inter-day RSD (%) (n=18)	Low-level	Mid-level	High-level
p-coumaric acid	y =146014x-29020	0.9995	1.961	2.383	84.5	87.3	90.0
Cinnamic acid	y = 241660x-151237	0.9993	4.626	4.474	80.4	83.7	92.4

Application of the quantification method in the ethanol leaf extract. The contents of *p*-coumaric acid and cinnamic acid are shown in Table 5. The content analytes were *p*-coumaric acid (1.19 mg/g) and cinnamic acid (3.82 mg/g). When comparing the results with the study of Huang et al. quantification of phenolic acids and flavonoids in the aqueous extract of *Avicennia marina* in Taiwan, the *p*-coumaric acid content (0.0058 mg/g) is much lower than the leaf sample extract (1.19 mg/g) in Vietnam (Huang et al., 2016). Plants of the same species can differ significantly in their content of secondary compounds under different environmental conditions. This is due to the fact that the quality and amount of bioactive compounds in medicinal plants are largely affected by environmental conditions. In some cases, the accumulation of secondary metabolites is influenced by geography and local biological conditions such as precipitation, humidity, temperature, water changes, and exposure to soil microorganisms, changes in soil pH, and nutrients. Furthermore, environmental factors also interact with genetic factors of plant transmission, which leads to a change in the concentration of secondary compounds and thus affects the substance quantity of medicinal plants (Sampaio et al., 2016).

Table 5. The content of *p*-coumaric acid and cinnamic acid in ethanol leave extract of *Avicennia officinalis*

Analyte	Content (mg/g)*
p-coumaric acid	1.19 ± 0.03
Cinnamic acid	3.82 ± 0.08

*Each value in the table is the mean of 3 replicates ± standard deviation (Mean ± SD), n=3.

3.4. Develop a standard for a standardized ethanolic extract

The results in Table 6 show that the ethanol leaf extract of *Avicennia officinalis* with the loss of dry weight, ash total, acid-insoluble ash, heavy metal content (Pb), microbial contamination level, qualitative by TLC, and quantitative by HPLC was used to determine the presence of marker compounds in the range of standard extract requirements for medicinal herbs according to the Vietnamese Pharmacopeia V. Therefore, this result contributes to providing data to be standardized and extracted from the leaves of *Avicennia officinalis*.

Table 6. Standard parameters of *Avicennia officinalis* leaves extract

Parameters	Method	Result	Quality level
The loss of dry weight (%)	Appendix 9.6, Vietnamese pharmacopeia V	10.9 ± 2,09	< 20%
Total ash content (%)	Appendix 9.8, Vietnamese pharmacopeia V	22.31	< 30%
Acid-insoluble ash content (%)	Appendix 9.7, Vietnamese pharmacopeia V	Not detected	< 3%
Heavy metal content (Pb)	AOAC 2013.06	Not detected	≤ 25 ppm
***Escherichia coli (E. coli)* (CFU/g)**	ISO 16649-2:2001	< 10	$< 10^2$
***Staphylococcus aureus* (CFU/g)**	ISO 13720:2010	< 10	$< 10^2$
***Pseudomonas aeruginosa* (CFU/g)**	ISO 6888-1:2021	< 10	$< 10^2$
Quantitation	Appendix 5.4, Vietnamese pharmacopeia V: Thin layer Chromatogram	The extract sample and cinnamic acid standard sample: R_f = 0.78	The chromatography of test sample with R_f of traces corresponds to the standard sample
	Appendix 5.3, Vietnamese pharmacopeia V:HPLC-PDA	The extract sample and cinnamic acid standard sample: The retention time t_R = 31.5 min	The peak of the test sample in chromatography has a retention time corresponds to the standard sample
Quantification	Appendix 5.3, Vietnamese pharmacopeia V:HPLC-PDA	*p*-coumaric acid: 0.119 % Cinnamic acid: 0.382 %	> 0.1 % (*p*-coumaric acid) > 0.3 % (cinnamic acid)

Standardization of traditional medicine involves all areas, including the procurement of essential components and the execution of rigorous treatment techniques. The effectiveness of traditional medication depends entirely on its chemical components. Therefore, rather than concentrating on a particular component of the medication, the phrases quality and purity embrace all aspects of the substance's attributes. Consequently, the standardization of traditional medicine requires the adoption of a thorough and varied methodology. This comprehensive approach should include the identification of the drug, including its name, botanical and geographic origins, sensory properties, physical characteristics, anatomical features, chemical composition, and biological effects (Wu et al., 2012). The World Health Organization (WHO) emphasizes the significance of utilizing both qualitative and quantitative approaches to characterize samples, evaluate biomarkers and/or chemical markers, and analyze fingerprint profiles. Quantification of a chemical is most rational when the principal active component is recognized. Standardization of botanical preparations is necessary to determine the active ingredients responsible for their medicinal effectiveness. A plant-specific marker component can be employed for analytical reasons in cases when the active ingredients remain undetermined (Lamichhane et al., 2023). The references provided in pharmacopoeia are used to verify the validity, excellence, and purity of herbal medications. These publications offer a basis for clinical practice by publishing conventional and traditional herbal medicinal applications. In monographs, the herb is described together with botanical details, laboratory findings, medicinal applications, and medication interactions. The pharmacopoeia establishes numerical values for the medications' structural, analytical, and physical standards (WHO, 2002). In this research, standardization was established for the ethanolic extract of *Avicennia officinalis* leaves in Vietnam, which is a potential plant with pharmacological effects to develop an herb in the future.

4. Conclusions

This study determined the parameters of the *Avicennia officinalis* leaf standardized extract, including the loss of dry weight, total ash content, acid-insoluble ash content, total heavy metal content (Pb), microbial contamination, qualitative by phytochemical screening and thin layer chromatography (TLC), qualitative total phenolic content, and the content of marker compounds (*p*-coumaric acid and cinnamic acid) by high-pressure liquid chromatography. This will contribute to providing data for other studies for the purpose of standardizing the source of this medicinal plant in Vietnam.

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Conflicts of interest. The authors mentioned that none of them have a conflict of interest when it comes to this article.

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