

Article

Comparison of Antioxidant and Antibacterial Properties of Five Plants with Anti-Diabetes and Anti-Cancer Potential

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Abstract: Polyphenols and flavonoids are bioactive organic compounds extracted from medicinal plants. They exhibit significant antioxidant and antibacterial properties, which help fight several chronic diseases, such as diabetes and cancer. Numerous therapeutic effects and a broad spectrum of biological activities are exhibited by the following five medicinal plants traditionally utilized in medicine for the treatment of diabetes and cancer: *Ginger*, *ephedra alata*, *ajuga iva*, *nettle*, and *graviola (annona muricata)*. The objective of the present study is to examine ethanolic and aqueous extracts exhaustively obtained from these plants through decoction and maceration using ethanol, with particular emphasis on the content of total polyphenols and flavonoids, and to evaluate their in vitro antioxidant and antibacterial potential. The antibacterial effect was assessed on the strains *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pneumoniae*. The study was complemented by an FTIR analysis of the different extracts. The results indicate that for *ginger*, *graviola*, and *ajuga iva*, as opposed to *ephedra alata*, maceration appears to be the more efficacious technique compared to decoction. The highest yield (27.465%) was observed in the case of the ethanolic extract of ginger. Ethanolic extracts contain higher concentrations of polyphenols and flavonoids than aqueous extracts. The aqueous extracts of *ajuga iva* and *nettle* demonstrate the highest inhibition of *Staphylococcus aureus* bacteria.

Keywords: phytotherapy; medicinal plants; polyphenol; flavonoid; extraction; ethanolic extract



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1. Introduction

Since ancient times and throughout the world, plants have been used for therapeutic purposes [1,2]. Plant material contains a large number of interesting molecules, including alkaloids, polyphenols, tannins, and flavonoids, widely used in the food industry, cosmetology, pharmacy, and medicine [3–5]. Evaluation of antioxidant and antimicrobial properties remains a very interesting and useful task, especially for plants used by the population for the treatment of chronic diseases, such as diabetes and cancer [6,7].

As a popular spice and condiment, *ginger* is also one of the most widely used remedies in the world [8]. Since the earliest times, it has been highly prized in Asia [9]. *Ginger* relieves morning sickness and motion sickness and was reported to be very useful in treating circulatory disorders [10]. This plant has an anti-ulcer effect similar to that of the drug omeprazole [11]. Several recent studies have shown the effect of *ginger* in the treatment of diabetes [12,13], prostate cancer [14], colon cancer [15], and cervical cancer [16]. Species of the genus *ephedra alata* are among the oldest medicinal herbs known to humankind. *Ephedra alata* has traditionally been used in China to combat bronchial asthma, colds, fever, rhinitis, nasal congestion, headaches, and arthralgia and as a diaphoretic, antiallergic, and antitussive [17–19]. Currently, *ephedra alata* is used by people as an herbal tea to treat cancer. In traditional medicine, *ajuga iva* is used to treat diabetes and hypertension, as well as gastrointestinal disorders and stomach ulcers [20,21]. Several medicinal properties are attributed to *nettle*, including adaptogen, nutritive, astringent, tonic, antiasthmatic, stimulating, and depurative properties. Currently, people use nettle as an herbal tea to treat diabetes [22,23]. All parts of *graviola* are used to treat malaria, stomach ailments, parasitic infections, diabetes, cancer, and skin diseases [24–26].

This work aims to investigate the impact of the plant extracts' nature and the extraction method on their antioxidant and antibacterial potential. This comparative study between extracts of five important medicinal plants (*ginger*, *graviola*, *ephedra alata*, *ajuga Iva*, and *nettle*) obtained through two conventional methods, maceration and decoction, uses in one part an evaluation of the polyphenol and flavonoid content as an indicator of their antioxidant potential and, in another part, their antibacterial capacity. These properties involve the therapeutic effects of these medicinal plants. Characterization carried out with UV-visible spectrometry and the FTIR of the whole of the extracts is used to deepen the comprehension of the differences recorded in these two potential effects.

2. Materials and Methods

2.1. Plants

The various plants were purchased in Algiers. For *ajuga iva*, *graviola*, *ephedra*, and *nettle*, the leaves were used for extraction, while for *ginger* the rhizome was used.

2.2. Reagents

The following reagents were purchased from Eden Lab, Feralco, Scharlau, and Biochem, respectively: ethanol (C_2H_5OH 97%), aluminum chloride ($AlCl_3$), Folin Ciocâlteu reagent (mixture of $H_3PW_{12}O_{40}$ and $H_3PMo_{12}O_{40}$), and gallic acid ($C_7H_6O_5$). Meanwhile, sodium carbonate (Na_2CO_3) and quercetin ($C_{15}O_7H_{10}$) were obtained from Sigma-Aldrich. St. Louis, MO, USA

2.3. Extraction Process

Extraction with ethanol as the solvent was carried out according to the work of Al-Bayar [27] using maceration and decoction, with some modifications described in the following sections.

2.3.1. Extraction with Ethanol Maceration

First, 20 g of dry plant powder (*graviola*, *ajuga Iva*, *ginger*, *nettle*, *ephedra alata*) was introduced into 100 mL of ethanol at ambient temperature for 24 h, with magnetic stirring at 150 rpm. After filtration, the obtained filtrate was kept, and 100 mL of ethanol was added to the residue under magnetic stirring for 24 h. This operation was repeated two times. The two filtrates were combined, and the mixture was concentrated with a rotavapor at 40–45 °C (Figure 1).

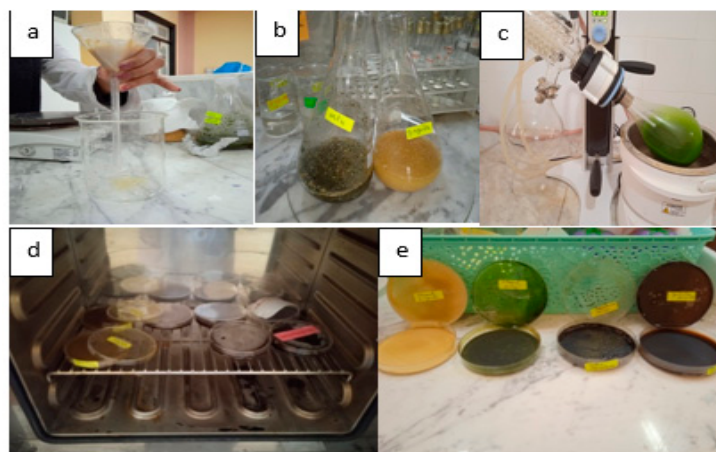


Figure 1. Plant extract preparation: (a) filtration of macerated plant. (b) Ethanol and aqueous plant extract before solvent elimination. (c) Solvent evaporation using rotavapor. (d) Plant extract drying. (e) Dried plants' extracts.

Another sample was prepared by extracting 3 times with a total volume of 300 mL of ethanol using 20 g of the plant and a volume of 100 mL for each extraction.

A third sample was prepared by extracting 2 times with a total volume of 300 mL of ethanol using 20 g of the plant and a volume of 150 mL for each extraction. Finally, the extract obtained was oven-dried at 40 °C and weighed (Figure 1) to determine the yield.

2.3.2. Extraction Using Decoction

First, 20 g of plant powder (*graviola*, *ajuga lva*, *ginger*, *nettle*, *ephedra alata*) was introduced into boiling distilled water (200 mL, 300 mL). The mixture was kept at 60 °C in a closed Enlenmeyer under magnetic agitation of 150 rpm for 45 min. The mixture was then filtered, and then the obtained filtrate was concentrated in a rotavapor (65 °C to 68 °C). The aqueous extract obtained was dried in an oven at 40 °C.

The yield of the isolated extracts using maceration or decoction was calculated based on the weight of the leaves, as given in the following equation:

$$R\% = \frac{M_e}{M_v} \cdot 100 \quad (1)$$

$R\%$: yield in %.

M_e : dry extract mass.

M_v : mass of plant used for extraction.

2.4. Characterization

2.4.1. Fourier Transform Infrared Analysis FTIR

The infrared spectra were obtained using a SHIMADZU brand FTIR-8400 spectrophotometer from anhydrous KBr pellets containing approximately 1% by mass of extract sample powders. First, the sample was prepared in pellet form. The powdered sample was incorporated into a potassium bromide KBr support. The pellet was placed in the spectrum measurement compartment. The signal recorded by the detector gives the IR spectrum of the sample.

2.4.2. Determination of Total Polyphenols

Total polyphenols in the various extracts were determined through the Folin–Ciocalteu method [28]. First, 1 g of dry extract was dissolved in 100 mL of distilled water or ethanol. Then, 0.3 mL of this stock solution was mixed with 1.5 mL of 10% (v/v) Folin–Ciocâlțeu

reagent and 0.2 mL of 7.5% (m/V) sodium carbonate (Na_2CO_3). The mixture was stirred for 10 s and kept in the dark for 1 h 30 min. The total polyphenol concentrations of the extracts were calculated by referring to the calibration line obtained using gallic acid as a standard at different concentrations under the same conditions as the samples. A UV-visible spectrometer was used to measure the concentrations. The results are expressed in mg of gallic acid equivalent/g of dry extract by applying the following formula [29]:

$$C_p = \frac{C_g \cdot F \cdot V}{m} \quad (2)$$

C_p : total polyphenol content (mg of gallic acid/g of dry extract).

C_g : gallic acid concentration (mg/mL).

F : dilution factor (6.76 in our case).

V : ethanolic or aqueous extract volume (mL).

m : dry extract weight (g).

A blank solution was formed as a mixture of ethanol or water with Folin–Ciocâlteu reagent and sodium carbonate.

2.4.3. Dosage of Flavonoids

The flavonoid contents of the extracts were measured using a colorimetric method using aluminum chloride. A mixture of 1 mL of each extract solution (prepared through dissolution of 0.1 g of dry extract in an appropriate volume of water and ethanol in the case of decoction and maceration, respectively) at different concentrations was mixed with 1 mL of aluminum chloride (2%) and then incubated at room temperature for 10 min. Absorbances of the prepared solutions were measured using a UV-visible spectrometer at 430 nm.

The results are expressed in equivalent mg of quercetin per gram of dry extract by referring to the calibration curve of quercetin and then applying the following formula [29]

$$C_F = \frac{C_q \cdot F \cdot V}{m} \quad (3)$$

C_F : the flavonoid content (mg of quercetin/g of dry extract).

c_q : quercetin concentration (mg/mL).

F : dilution factor ($F = 2$ in our case).

V : ethanolic or aqueous extract volume (mL).

m : dry extract weight (g).

A blank solution was formed as a mixture of ethanol or water with aluminum chloride.

2.4.4. Evaluation of Antibacterial Activity

The antibacterial activity of (*graviola*, *ajuga Iva*, *ginger*, *nettle*, *ephedra alata*) leaf extracts was assessed invitro on the following pathogenic bacteria: Gram-negative *Escherichia coli* and *Pseudomonas aeruginosa* and Gram-positive *Staphylococcus aureus* and *Streptococcus pneumoniae*. The antibacterial activity was ascertained using the disc diffusion method with standard Mueller Hunton Agar media. Sterile filter paper discs (9 mm in diameter) were impregnated with 5 μL of pure isolated extracts and then placed on inoculated Petri plates. The plates were then incubated at 37 °C for 24 h, and the zone of inhibition around the disc was measured. The extract was prepared in the concentration range of 25, 50, 75, and 100 mg/mL.

2.5. Statistical Analysis

The data are represented as the mean $\pm$ standard deviation (SD) of three replicates. One-way analysis of variance (ANOVA) was performed to assess the statistical significance

of differences among treatments, followed by Tukey's test at a 5% significance level to compare the means. These analyses were performed using IBM SPSS statistical software (ver.22.0, SPSS Inc., Chicago, IL, USA).

3. Results and Discussion

3.1. Extraction Yield

Table 1 depicts the yield of the isolated extracts using both decoction and ethanolic maceration from the five medicinal plants investigated in this study.

Table 1. Yield values obtained for various plants tested under different extraction methods.

Plants		<i>Nettle</i>	<i>Ajuga iva</i>	<i>Graviola</i>	<i>Ginger</i>	<i>Ephedra Alata</i>
Yield %	Decoction m/v = 0.1	7.205	15.27	14.985	25.14	15.27
	Decoction m/v = 0.067	11.805	20.995	12.02	25.305	16.455
	Maceration (2-fold) m/v = 0.1	12.65	6.565	3.62	23.795	18.22
	Maceration (2-fold) m/v = 0.067	7.92	9.6	6.61	16.39	26.32
	Maceration (3-fold) m/v = 0.067	6.765	8.755	3.06	27.465	21.07

The highest rate (27.465%) was found in the ethanolic extract of *ginger* produced through three-fold maceration, followed by the ethanolic extract (three-fold maceration) of *ephedra*, with a yield of 26.32%. The lowest rate (3.06%) was noted in the ethanolic extract of *graviola* for the same ratio of mass/volume of 0.067.

The extraction yield depends on the extraction method as well as the type of plant studied. Ethanol is often used as a solvent for maceration due to its effectiveness in extracting polyphenols. In a study on *Kaempferia parviflora* (black *ginger*), maceration with 70% ethanol for 48 h yielded the highest total flavonoid content of 10.6161 ± 0.0768 mg EQ/g [30]. For *Zingiber officinale*, maceration with ethanol at room temperature for 8 h generally showed better total phenolic content, 6-gingerol content, and antioxidant activity compared to other solvents and methods [31].

According to our findings, two-fold maceration gave higher yields than three-fold maceration, except for *ginger*. This unexpected superiority may be due to the impact of the m/v ratio rather than the number of repeated extractions, as, for each extraction, the m/v was higher than the total m/v ratio. As with maceration, the reduction of the m/v ratio had a favorable effect on the extract yields in the case of decoction. This was not the case for *nettle*, *ginger*, or *graviola* when using maceration or decoction.

Our results show that for *ginger*, *graviola*, and *ajuga iva*, as opposed to *ephedra alata*, maceration seems to be the more effective technique compared to decoction. However, by modifying the mass-to-volume ratio for *nettle*, the yield's monotonic behavior is altered, with the degree of change depending on the extraction method employed. This could be due to the influence of the nature of the compounds contained in each plant and their interaction with the polarity of the solvents used (such as solubility and affinity) on the one hand, and, on the other hand, the impact of the heating used in the case of decoction (60 °C). The solvent's nature plays a key role in the extraction process, as solubility is the most important parameter [31].

While both decoction and maceration with ethanol can be used to extract *ginger* polyphenols, maceration with ethanol generally yields good results. The optimal maceration time and ethanol concentration may vary, but studies suggest that 48 h with 70%

ethanol or 8 h with pure ethanol can be effective. However, advanced techniques, like MAE or UAE, may offer improved efficiency and yields compared to traditional methods [32].

Also, according to previous research on *ephedra alata*, maceration using different solvents has proven effective for extracting polyphenols, with aqueous and ethanolic extracts showing promising results. However, the optimal extraction conditions may vary depending on the specific compounds of interest and the desired application [33].

Differences in solvent polarity and extraction temperature between maceration and decoction methods may influence the efficiency of phytochemical extraction.

Decoction, which involves higher temperatures, likely facilitated the extraction of heat-stable polar compounds, such as certain polyphenols and flavonoid glycosides, leading to higher antioxidant and antibacterial activity in some fractions.

Maceration, performed at room temperature, may have better preserved thermolabile compounds and, depending on the solvent used, selectively extracted non-polar or moderately polar metabolites.

These factors may explain the qualitative and quantitative differences observed in both photochemical profiles and biological activity between extracts derived from each method. This addition enhances the mechanistic understanding of how extraction conditions affect compound recovery and bioactivity

3.2. Total Phenolic Content

The concentrations (C) of total polyphenols in mg EGA/g of dried extract contained in the extracts are calculated by referring to the following calibration lines obtained using gallic acid as a standard at different concentrations:

In ethanol:

$$A = 0.132 C \quad (4)$$

In distilled water:

$$A = 0.216 C \quad (5)$$

A: absorbance in UV-visible spectrometer.

According to the results summarized in Table 2, it appears clearly that the five plants are rich in phenolic compounds, with high levels recorded for *ephedra alata*. This underlines its use by the population for the treatment of cancer, as a plant that is rich in antioxidants (polyphenols, flavonoids) has a more effective effect on hard diseases [4,28]. A study by Zaater et al. [34] assessed the phytochemical content and antioxidant effects of *ephedra alata* leaf extract and found that the total phenolic content and total flavonoid content were $48.7 \pm 0.9 \text{ mgg}^{-1}$ and $1.7 \pm 0.4 \text{ mgg}^{-1}$, respectively. In addition, ethanolic extracts contain more polyphenols than aqueous extracts. This finding is in agreement with the work of Muanda et al. [35] on *Daniella oliveri* and *Ficus capensis* plants, where they found that the aqueous extracts were the least rich in total polyphenols. So, a hydroalcoholic mixture of 30 and 70% (v/v) methanol/water was used to improve the PPT rate, and the best result was obtained with 70% methanol. In another study performed by Do et al. [36], the highest amounts of total polyphenols were obtained when extracting with 100% ethanol.

Previous studies have reported total polyphenol contents in *ephedra alata* ranging from 53.3 mg EGA/g to 291.45 mg EGA/g [37–40]. Bragic et al. [37] found that the polyphenol content in *ephedra alata* from Bosnia was $53.3 \pm 0.1 \text{ mg EAG/g}$. Al-Rimawi et al. [38] found that the amount in Palestinian *ephedra alata* was $101.2 \pm 0.9 \text{ mg EAG/g}$. Kebili, who used extraction with methanol [39], found $291.45 \pm 4.37 \text{ mg EAG/g}$ of total polyphenol content in south Algerian *ephedra alata*. Our observed value of 356.76 mg EGA/g suggests higher polyphenol content, potentially due to differences in species, geographical origin, or extraction methods. Environmental stresses, such as drought, nutrient deficiency, and

strong sunlight, can contribute to increased levels of phenolic compound production in some plants. This may explain the richness of *ephedra alata* from the Saharan region in phenolic compounds compared to other *ephedra* species worldwide [40]. Most *Ephedra* species are adapted to arid and desert conditions and widely used in folk medicine to treat several disorders [41]. The high levels of antioxidant in Algerian *ephedra alata* explain these therapeutic effects on certain chronic diseases [42]. *Ephedra alata* could provide a potential source of new compounds that have pro-apoptotic effects to treat breast cancer [43]. Furthermore, we acknowledge the limitations of our study, including the lack of in vivo validation and the need to assess the clinical relevance of the tested concentrations.

Table 2. Total polyphenol content in dry extracts.

Extract	Polyphenol Concentration (mg EGA/g DE)				
	Aqueous Extract (Decoction)		Ethanollic Extract (Maceration)		
	m/v				
	0.1	0.067	0.1 (2 times)	0.067 (2 times)	0.067 (3 times)
<i>Ginger</i>	14.73 ± 0.17	31.33 ± 0.58	41.10 ± 0.27	101.6 ± 0.55	103.92 ± 0.08
<i>Ajuga iva</i>	39.92 ± 0.03	36.14 ± 0.16	57.94 ± 0.12	87.15 ± 0.06	88.61 ± 0.14
<i>Graviola</i>	54.76 ± 0.79	51.18 ± 0.16	56.48 ± 0.50	82.21 ± 0.72	124.0 ± 0.27
<i>Ephedra</i>	237.33 ± 0.13	134.95 ± 0.05	308.68 ± 0.59	137.28 ± 0.14	356.76 ± 0.57
<i>Nettle</i>	50.41 ± 0.08	41.10 ± 0.11	101.6 ± 0.53	133.40 ± 0.42	126.41 ± 0.26

mg EGA/g DE: mg equivalent of gallic acid per gram of dry extract.

3.3. Total Flavonoid Content

Figure 2 represents the results of flavonoid content in the extracts isolated from the five studied plants, which were expressed in equivalent mg of quercetin per g of dry extract, referring to the quercetin calibration curve:

$$A = 0.0281 \cdot CF + 0.0102 \quad (6)$$

with CF in $\mu\text{g/mL}$

The ethanollic extracts are found to be richer in flavonoids than the aqueous extracts. Compared to water, ethanol enables the extraction of a greater range of compounds, including those with varying polarities. Our results show that *ajuga Iva* and *graviola* contain more flavonoids than the other plants tested, and this fact remains clearly visible in both ratios tested.

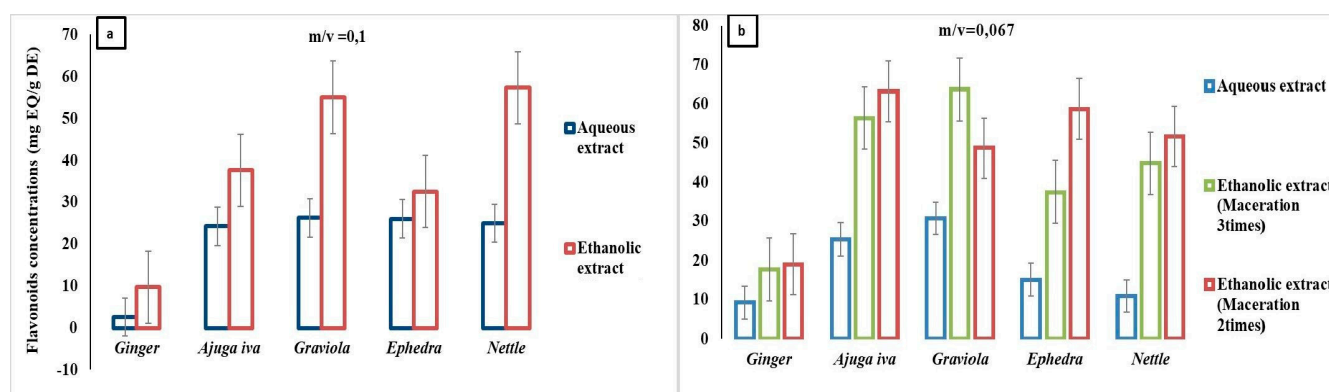


Figure 2. Total flavonoid content in ethanollic and aqueous plant extracts in equivalent mg of quercetin per g of dried extract under two m/v ratios. (a). m/v ratio 0.1; (b) m/v ratio 0.067.

Polyphenols, flavonoid, isocoumarins, and their glycosylated derivatives are well-known for their antioxidant, anti-inflammatory, and anti-diabetic activities [44]. To fight

against multifaceted diseases, such as inflammation or cancer, many natural products rich in antioxidants are all identified to be acting on multiple targets [44,45].

3.4. Antibacterial Analysis

To evaluate the in vitro antibacterial potency of our extracts, the inhibition diameter was taken as an indicator. Figure 3 shows that the diameter of the inhibition zone changes with the bacteria strain type and the extract's nature. In fact, all of the tested extracts exhibited an inhibitory effect more significantly against *Staphylococcus aureus*. In the case of this last strain, the aqueous extracts of *ajuga iva* and *nettle* have the highest inhibition diameters compared to other plants (Figure 4). The variations in the antimicrobial activity of samples explain variations in their chemical composition.

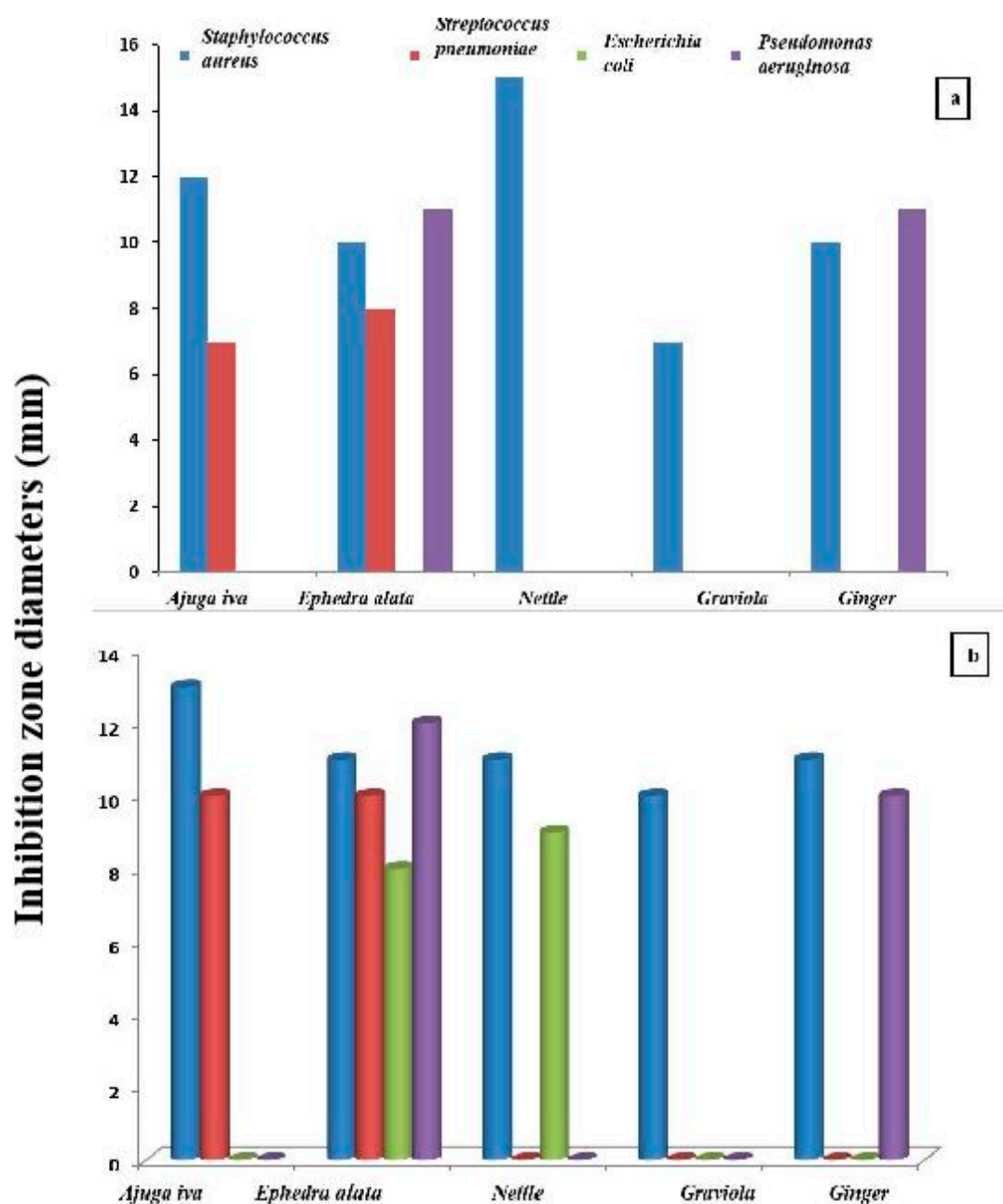


Figure 3. In vitro antibacterial activity against various bacterial strains of different tested extracts. (a) Aqueous extract and (b) ethanolic extract.

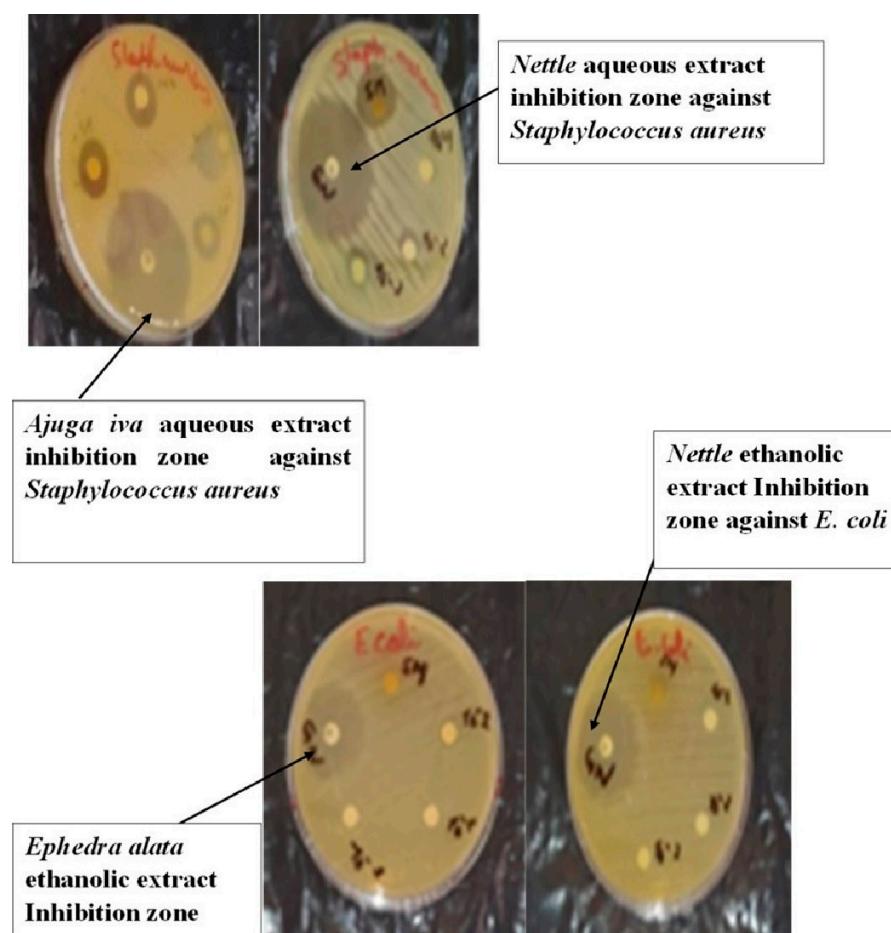


Figure 4. Inhibition zone of ethanolic and aqueous extracts against *Staphylococcus aureus* and *Escherichia coli*.

In previous studies, a rather concentrated solution was used to observe inhibition. As shown in the work of Albayati et al. [46] on ginger extracted through maceration with ethanol, the relationship showed an antibacterial effect on *Escherichia coli* for concentrations above 50 mg/mL and no inhibition for a concentration of 25 mg/mL. The diameter of inhibition increases with concentration from 15.33 mm at 50 mg/mL to 23 mm at 100 mg/mL.

According to our results, *ephedra alata* and *ajuga iva* are recorded as inhibitors of *streptococcus pneumoniae* bacteria, while the other plants have not demonstrated any inhibitory effects on these bacteria.

The antibacterial action is more pronounced for the ethanolic extract compared to the aqueous one, which may be due to the lack of a plant extract fraction that may contain important antibacterial and antioxidant elements.

The occurrence of flavonoids, as summarized in Figure 2, may be responsible for good antibacterial activity and is further related to the medicinal value index. This result is in agreement with the literature [46,47].

The antibacterial effect is also affected by the extraction method, as seen in the work of [47] on ginger, where larger inhibition diameters were obtained with ethanol maceration followed by decoction and cold-water extraction.

3.5. FTIR Analysis

The aim of infrared spectrometry is to identify the chemical functions present in a product. From the results of infrared analysis of the various extract fractions illustrated in Figures 5 and 6, it emerges that the five plants are characterized by several variable-function

bonds. The broad bands around 3400 cm^{-1} indicate O–H stretching vibrations of hydroxyl groups, suggesting the presence of alcohols and phenols [48,49], which may contribute to the bioactivity observed in our study. Considering the intensity of this peak in both extracts, it can be seen that there is a superiority of the presence of this kind of molecule in the case of ethanolic extract, and this is also confirmed by the result obtained by the dosage (Table 1). The peak at 1388 cm^{-1} corresponds to the in-plane deformation of the O–H bond in phenolic compounds [48,49].

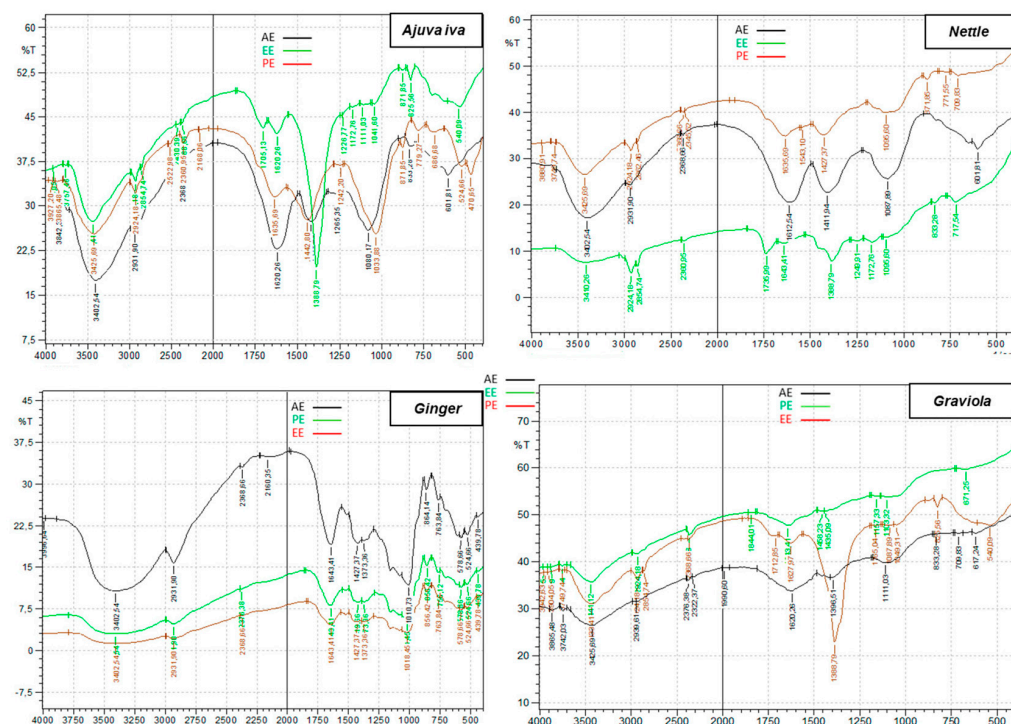


Figure 5. IR spectra of *ajuva iva*, *nettle*, *ginger*, and *graviola*. (AE: aqueous extract; EE: ethanolic extract; PE: plant before extraction).

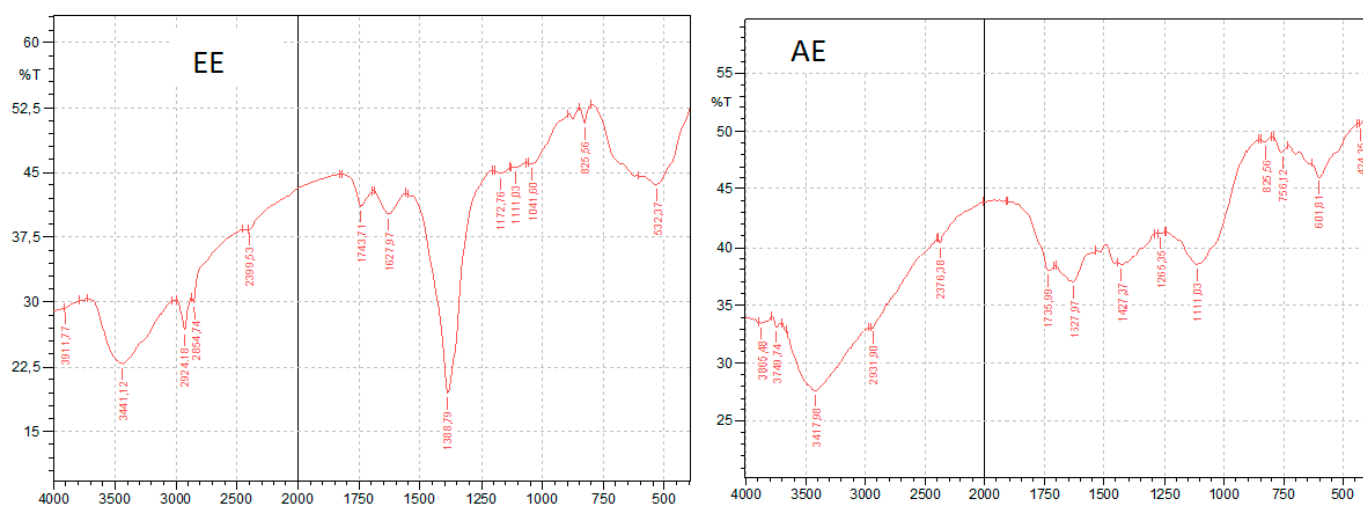


Figure 6. IR spectra of *ephedra alata* (AE: aqueous extract; EE: ethanolic extract).

The narrow bands around 2924 to 2926 cm^{-1} correspond to the valence vibration of the C-H bond (alkane function). Strong bands around 1630 to 1640 cm^{-1} characterize the double bond [47,48].

4. Conclusions

In this study, the antibacterial potency and total content of both flavonoids and polyphenols were investigated for five medicinal plants (*ginger*, *ajuga iva*, *graviola*, *ephedra alata*, and *nettle*) chosen on the basis of the frequency of their use by the local population for the treatment of diabetes and cancer. For both of the extraction methods used, ethanol maceration and decoction, our results showed that quantitative differences in extraction yield can be recorded as differences in the extraction method, the kind of plant, extraction solvent polarity, and the m/v ratio. All of the tested plants are rich in polyphenols with important content in ethanolic extracts, and the highest level is obtained in the case of *ephedra alata*. Compared to decoction, maceration using ethanol as the solvent can enhance the extraction of flavonoids.

Among all of the plants tested, *ajuga iva* and *graviola* were the richest ones in flavonoid content. All of the extracts have an inhibitory effect on *Staphylococcus aureus* bacteria and variable effects on other bacteria, with great improvement exerted by ethanolic extracts. Only *ephedra alata* and *nettle* can exhibit antibacterial activity on *Escherichia coli*, and only *ephedra alata* and *ajuga Iva* have an inhibitory effect against *Streptococcus pneumoniae* bacteria. For future purposes, we suggest using the green extraction method for the alkaloids class, which are highly effective in the treatment of cancer, and evaluating the anti-cancer activity of these plants.

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Abbreviations

m/v	Mass of the plant/volume of the extraction solvent
MAE	Microwave-Assisted Extraction
UAE	Ultrasonically Assisted Extraction
mg EGA/g DE	mg equivalent of gallic acid per gram of dry extract
mg EQ/g DE	mg equivalent of quercetin per gram of dry extract
PPT	Total polyphenols
<i>E coli</i>	<i>Escherichia coli</i>

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