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Phytochemical Content, Inorganic Composition, Mineral Profile, and Evaluation of Antioxidant Activity of Some Common Medicinal Plants

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Abstract

The current research focuses on the antioxidant activity, phytochemical, and inorganic content of five common medicinal plants: Asian pigeonwings, lemon balm, lemongrass, ginger, and turmeric. According to the phytochemical screening, flavonoids were present in all plants; tannins were present in lemon balm and lemongrass, saponins were present in all plants except lemongrass, and anthraquinone glycosides were detected only in turmeric. The aqueous extracts from lemon balm, Asian pigeonwings, and the methanolic extract from turmeric had the highest phenolic content, respectively. The inorganic composition of plants was determined using energy dispersive X-ray fluorescence. Asian pigeonwings, ginger and turmeric are mainly composed of K, while lemon balm and lemongrass are mainly composed of Ca. The mineral profile of each herb was determined by inductively coupled plasma-optical emission spectrometry analysis. The antioxidant activity of each herb was determined by the 2,2-diphenyl-1-picrylhydrazyl radical scavenging activity assay. Among the extracts, the aqueous extract from ginger had the highest antioxidant activity with a percentage of 88.05 ±0.31 %.

Keywords: phytochemical content of plants; inorganic composition of plants; mineral profile of plants; antioxidant activity

دراسة مكونات بعض النباتات الطبية من حيث المنتجات الطبيعية والمعادن وقياس نشاطها كمضاد للأكسدة

فضيلة السلمان, علي علي رضا, زينب عقيل, زهراء علي
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الخلاصة

البحث يسلط الضوء على خمس نباتات طبية تستخدم في البحرين كأدوية شعبية (الشاي الازرق ، بلسم الليمون ،عشبة الليمون، الزنجبيل ، والكركم). تمت دراسة النباتات من حيث محتواها من المنتجات الطبيعية والمعادن وقدرتها على مقاومة الأكسدة. وفقاً للتحليل النوعي للمنتجات الطبيعية تم اثبات وجود الفلافونيدات في الخمس نباتات بدون استثناء ووجود التانين في بلسم الليمون وعشبة الليمون. الصابونين تم الاستدلال عليه في جميع النباتات ما عدا عشبة الليمون بينما جليكوسيدات الأنثركوينون تم الاستدلال عليها في الكركم فقط. وتبين ان الخلاصة المائية لبلسم الليمون والشاي الازرق وكذلك الخلاصة الكحولية للكركم تحتوي على اعلى

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مستوى من المواد الفينولية. تم تقييم نشاط النباتات قيد الدراسة من حيث مقاومة الأكسدة باستخدام مقايسة نشاط الكسح الجذري 2،2-ثنائي فينيل -1-بيكريل هيدرازيل . تبين ان الخلاصة المائية للزنجبيل تمتلك اعلى نشاط لمقاومة الأكسدة بنسبة مئوية مقدارها %0.31± 88.05.

1. Introduction

Medicinal plants have shown various pharmacological properties and have been considered a significant source of medicines since the start of human development [1]. Most research focusing on exploring medicinal plants and their components mainly focuses on the organic composition of the plants and the organic compounds present in the plants. Nevertheless, understanding the inorganic composition and mineral profile of medicinal plants can also contribute to the field of functional food and nutraceutical development. Some trace elements have significant effects on improving metabolic disorders [2]. They also facilitate several biochemical reactions by acting as co-factors or catalysts for enzymes [3]. However, trace elements can become harmful and toxic to the human body at high concentrations [4]. Thus, it is essential to determine and monitor the concentrations of heavy metals in nutraceuticals [5].

This study focuses on five common plants; Asian pigeonwings (*Clitoria ternatea*), lemon balm (*Melissa officinalis*), lemongrass (*Cymbopogon schoenanthus*), ginger (*Zingiber officinale*), and turmeric (*Curcuma longa*) which are used across Asia. The primary aim of this study is to explore the inorganic content of those plants and report possible future pharmacological and nutraceutical investigations based on their inorganic composition and mineral content. The five respective plants were chosen due to the limited information and findings available regarding their inorganic content, especially Asian pigeonwings, lemon balm, and lemongrass. This study also involves phytochemical investigations (phytochemical screening and estimation of phenolic content) and explores the antioxidant potential of the respected plants.

2. Materials and methods

Sample preparation

Dried Asian pigeonwings, fresh lemon balm, fresh lemongrass, dried ginger, and dried turmeric were bought from the local market. The fresh samples were washed with tap water, followed by distilled water, and dried at room temperature. A fine powder of each plant material was prepared using a commercial grinder. Samples were stored in dark glass bottles at room temperature until use.

Phytochemical screening

Phytochemical screening involved tests of the presence of flavonoids, tannins, saponins, anthraquinone glycosides, terpenes, sterols, and alkaloids, according to Wall *et al.* [6] and the modified version of Hassan *et al.* [7]. Aqueous extract, methanolic extract and acidic extract of each type of plant were prepared. For the aqueous extract, 1.0 g of each sample was added to 50 mL of water and boiled for 20 min at 90-100°C, followed by gravity filtration. For the methanolic extract, 1.0 g of each sample was added to 50 mL of methanol and magnetically stirred for 30 minutes at room temperature, followed by gravity filtration. For the acid extract, 0.5 g of each sample was added to 4.0 mL of dilute hydrochloric acid (4.0 M) and maintained at room temperature for 30 minutes, followed by gravity filtration.

To test for the presence of tannins, a few drops of ferric chloride (5%) were added to the aqueous extract (2.0 mL). The formation of a blue or green colour indicates a positive result, suggesting the presence of tannins. The formation of a blue colour suggests the presence of hydrolysable tannins, while the green colour suggests the presence of condensed tannins.

To test for the presence of flavonoids, two tests were conducted; (a) on 1.0 mL of each acidic extract, sodium hydroxide (4.0 M) solution was added until the pH reached 10. The formation of yellow colour indicates a positive result, suggesting the presence of flavonoids, (b) to 1.0 mL of each acidic extract, a few magnesium turnings were added and allowed to stand for 3

minutes at room temperature. The formation of a magenta-red colour indicates a positive result, suggesting the presence of flavonoids.
To test for the presence of saponins, each aqueous extract (2.0 mL) was shaken vigorously in a test tube and allowed to stand for 20 minutes; the formation of froth indicates a positive result, suggesting the presence of saponins.
To test for the presence of anthraquinone glycosides, to 2.0 mL of each methanolic extract of the plant being studied, concentrated ammonia (1.0 mL) was added. The formation of a red-rose colour indicates a positive result, suggesting the presence of anthraquinone glycosides.
Two tests were conducted to detect terpenes and sterols; (a) The Salkowski test in which concentrated sulphuric acid (5 drops) was added to methanolic extract (2 mL) of each sample. The formation of a red-brown colour at the bottom of the test tube indicates a positive result, suggesting the presence of terpenes and/or sterols, (b) The Liebermann-Burchard (5-10 drops) reagent [prepared by mixing concentrated sulphuric acid (19 mL) and acetic acid (1 mL)] was added slowly to the wall of each test tube that contained methanolic extract (2 mL). The formation of a red-violet colour indicates a positive result, suggesting the presence of triterpenes, while the formation of a red-violet colour indicates a positive result, suggesting the presence of steroidal compounds.
To test for the presence of alkaloids, Mayer reagent (5-10 drops) was added to each methanolic extract (2.0 mL). The formation of a white precipitate indicates a positive result, suggesting the presence of alkaloids.

Estimation of phenolic content

The phenolic content of the methanolic and aqueous extracts of each sample prepared in the "Phytochemical screening" section was estimated. A volume of 0.5 mL of each extract was mixed with a diluted Folin-Ciocalteu reagent (2.5 mL) (prepared by diluting the concentrated reagent with a dilution factor of 10) and shaken. After standing for 5 minutes at room temperature, sodium carbonate solution (2.0 mL, 75 g/L) was added to each test sample and vortexed [8,9]. After standing for an hour at room temperature, the absorbance of the solutions was measured at 760 nm using an ORION AQUAMATE 8000 UV/VIS spectrophotometer. Three replicates were conducted for each extract. The concentration of phenolic content was calculated by preparing a standard curve of gallic acid [a solution of gallic acid (10 mg) in 100 mL of distilled water was prepared (as a 100 μg/mL standard], which was used to prepare standard solutions of 5, 10, 20, 40 and 80 μg/mL by serial dilution]. The phenolic content of each extract was reported as a gallic acid equivalent (GAE).

Inorganic composition

The inorganic composition of each plant sample was determined using Epsilon 1 Meso energy dispersive X-ray fluorescence (ED-XRF) (PANalytical, Netherlands). Non-destructive measurement of the inorganic composition was accomplished with 15 W, 50 kV, and 1500 μA. The instrument contained a silver X-ray tube and was equipped with a small spot camera with a 1 mm^2 spot size to select the best spot for targeting. The analysis was performed with the help of Omnian qualitative software for analysing the X-ray spectra. Prior to analysis, the dry samples were grinded and then pressed onto a tablet by a compressor, and the analysis was repeated three times for each plant sample.

Mineral profile

The amounts of Ag, Al, As, B, Ba, Cd, Cu, Cr, Fe, Mn, Ni, P, Pb, Sb, Se, Zn, Ca, K, Mg and Na in each sample were determined using Agilent Technologies 700 Series inductively coupled plasma-optical emission spectrometry (ICP-OES). First, using a 1000 ppm multi-element stock solution containing Ag, Al, As, B, Ba, Cd, Cu, Cr, Fe, Mn, Ni, P, Pb, Sb, Se, Zn, Ca, K, Mg and Na, a 20 ppm elemental mother solution was prepared, from which a series of standard solutions with concentrations of 400, 200, 100 and 50 ppb were prepared for calibration. Then, a 500 mg amount of each herb sample was boiled in distilled water (25 mL) for 20 minutes. The samples were filtered and acidified with concentrated nitric acid (1.0 mL), and then diluted with a dilution factor of 100 using distilled water before analysis. The mineral profile of the plant samples of interest was determined with the aid of ICP-OES. Three replicates were conducted for each extract.

Determination of antioxidant activity

The *In vitro* antioxidant activity of each plant sample was determined by a 2,2-diphenyl-1-picrylhydrazyl (DPPH) inhibition assay [10,11]. A volume of 50 µL of aqueous or methanolic extract of each plant (prepared in the phytochemical screening section) was added to the DPPH solution (2.95 mL), which was prepared by dissolving DPPH (11.3 mg) in methanol (250 mL) and allowed to stand for 30 minutes at room temperature after being mixed. The absorbance of the fresh DPPH sample and each reaction mixture was measured at 517 nm using an ORION AQUAMATE 8000 UV/VIS spectrophotometer. Three replicates were conducted for each extract. Finally, the antioxidant potential for each extract was determined by calculating the percentage potential for scavenging DPPH radical according to the following equation:

$$\%\ Antioxidant\ activity = \left(\frac{Absorbance\ of\ fresh\ DPPH\ solution - Absorbance\ of\ test\ sample}{Absorbance\ of\ fresh\ DPPH\ solution}\right) \times 100\%$$

3. Results and discussion

Phytochemical screening

Qualitative tests were conducted to identify the phytochemicals in the plants of interest. According to the phytochemical screening, flavonoids were detected in all plants. Lemon balm and lemon grass extracts gave positive results for the test of tannins. Saponins were present in all the plants except for lemongrass, whereas anthraquinone glycosides were identified only in the turmeric extract. Terpenes, sterols, and alkaloids were not detected in any plant. Phytochemical screening results are outlined in Table 1.

Tannins are phenolic compounds and have been reported as antioxidants, being antimicrobial and anticancer, and having cardio-protective potential [12]. Thus, lemon balm and lemongrass could be evaluated for those biological activities. Flavonoids have similar pharmacological properties to tannins. Moreover, flavonoids have also been reported for their antiviral, anti-allergic, vasodilatory, and hepato-protective potential [13]. As stated, flavonoids were detected in the plants, and thus, the plants of interest can be evaluated for the outlined pharmacological activities based on their flavonoid content. Due to the presence of saponins in Asian pigeonwings, lemon balm, ginger and turmeric, they can be investigated for their anti-inflammation, anti-tumor, antiviral, antidiabetic, anti-inflammatory, antifungal, antiparasitic and antibacterial potential [14]. Additionally, turmeric can be evaluated for phytoestrogen, immunosuppressive, vaso-relaxing, laxative, cathartic, diuretic and anticancer activities in favour of anthraquinone glycosides [15].

Table 1 - Phytochemical screening of plants

Phytochemicals		**Asian pigeonwings**	**Lemon balm**	**Lemongrass**	**Ginger**	**Turmeric**
Tannins		–	+	+	–	–
Flavonoids	Test A	–	+	+	+	+
	Test B	+	–	–	+	–
Saponins		+	+	–	+	+

Anthraquinone glycosides		–	–	–	–	+
Terpenes and sterols	Test A	–	–	–	–	–
	Test B	–	–	–	–	–
Alkaloids		–	–	–	–	–

Phenolic content

The phenolic content of the respected plants was estimated by the Folin-Ciocalteu method and is illustrated in Figure 1. Water was more efficient in extracting the phenolic compounds in Asian pigeonwings and lemon balm compared to methanol. However, methanol was more efficient in extracting the phenolic compounds of turmeric, compared to water. The aqueous extracts from lemon balm, the aqueous extract of Asian pigeonwings, and the methanolic extract of turmeric had the highest phenolic content, respectively. Thus, those extracts could be evaluated for their pharmacological properties, such as their antioxidant activity.

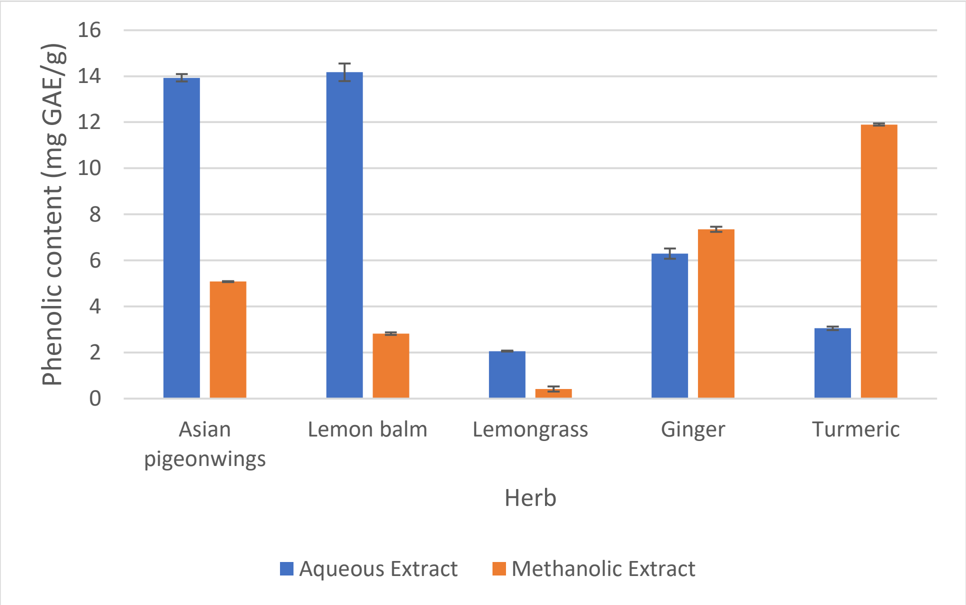

Figure 1 - Phenolic content of aqueous and methanolic extracts of plants. The values are reported as a mean of three replicates, with error bars representing the standard deviation

Inorganic composition

The inorganic composition of plants, in a solid state, was determined using ED-XRF and is outlined in Table 2. Asian pigeonwings, ginger and turmeric were mainly composed of K. On the other hand, lemon balm and lemongrass mainly comprised Ca. Both K and Ca are considered to be major elements [2]. Based on the respective results, Asian pigeonwings, ginger, and turmeric, can be explored for treating diabetes, reduction of fracture risks, and osteoporosis due to their high Ca content [16]. In fact, those plants can also be explored for preventing hypertensive disorders during pregnancy [17]. However, lemon balm and lemongrass could be investigated for regulating vascular calcification and arterial stiffness due to their high K content [18]. Additionally, those two plants can also be investigated for decreasing blood pressure and lowering the risk of chronic kidney disease due to their high K content [19].

Table 2 - Plant sample elemental composition. The values are reported as a mean of three replicates ± standard deviation

Element	Percentage Elemental Composition (%)				
	Asian pigeonwings	**Lemon balm**	**Lemongrass**	**Ginger**	**Turmeric**
Si	1.152 ±0.053	6.785 ±0.020	14.072 ±0.085	1.331 ±0.042	1.303 ±0.033
P	5.615 ±0.084	1.694 ±0.063	1.916 ±0.048	3.043 ±0.057	2.276 ±0.031
S	4.415 ±0.091	2.412 ±0.098	2.382 ±0.056	11.229 ±0.104	1.345 ±0.022
Cl	2.541 ±0.072	4.030 ±0.087	5.170 ±0.102	2.192 ±0.034	6.179 ±0.094
K	83.872 ±2.914	18.053 ±0.783	18.417 ±0.451	76.659 ±0.505	86.754 ±1.102
Ca	-	65.944 ±1.831	50.731 ±1.074	-	-
Mn	0.295 ±0.013	0.188 ±0.014	-	3.402 ±0.039	-
Ti	-	-	0.355 ±0.021	-	0.188 ±0.037
Fe	1.372 ±0.042	0.757 ±0.031	1.906 ±0.089	1.905 ±0.026	1.734 ±0.044
Zn	0.514 ±0.012	0.079 ±0.005	0.240 ±0.026	0.240 ±0.014	0.067 ±0.002
Br	0.142 ±0.010	-	4.120 ±0.077	-	0.037 ±0.013
Rb	0.083 ±0.018	-	-	-	0.039 ±0.000
Sr	-	-	0.357 ±0.028	-	0.079 ±0.005
Zr	-	0.059 ±0.008	-	-	-
Co	-	-	0.335 ±0.036	-	-

Mineral profile

The amounts of Ag, Al, As, B, Ba, Cd, Cu, Cr, Fe, Mn, Ni, P, Pb, Sb, Se, Zn, Ca, K, Mg, and Na in plants were determined using ICP-OES as outlined in Table 3. The elements detected and quantified can be grouped into two groups; (a) elements present in concentrations of approximately 0-200 ppm, which include Ag, Al, As, B, Ba, Cd, Cu, Cr, Fe, Mn, Ni, Pb, Sb, Se, and Zn. (b) elements present in concentrations of above 500 ppm, which include P, Ca, K, Mg, and Na. The most significant findings in this section include high concentrations of K in Asian pigeonwings, lemon balm, ginger, and turmeric, high concentrations of Ca and Mg in lemon balm, high concentrations of Se, Al, and Sb in turmeric, and high concentrations of Se in Asian pigeonwings. The results of the mineral profile support the findings illustrated in Table 2 in terms of K and Ca content in different plants. Based on the mineral profile and the presence of high amounts of Mg in lemon balm, lemon balm can be investigated for its potential in reducing the risk of stroke, heart failure, migraine headaches, Alzheimer's disease, insulin resistance and diabetes [20,21]. Due to the high concentrations of Se in Asian pigeonwings and turmeric, these two plants can be evaluated in favour of treating hyperlipidaemia, hyperglycaemia, and hyperphenylalaninemia [22]. Sb was detected only in turmeric, with a concentration of 198.873 ± 2.229 ppm, which is considered very high and hazardous to the human body. The concentration of Sb in rivers and lakes is usually less than 5 ppb, and according to a U.S. geological survey, the mean concentration of Sb in soil was 0.48 ppm [23]. In fact, the concentration of Al in turmeric and Asian pigeonwings was very high (139.970 ±1.209 and 41.628 ±1.041 ppm, respectively). While the allowed aluminum level in drinking water is 0.2 ppm according to the U.S. Food and Drug Administration [24].

Table 3 - Mineral profile of plant samples. The values are reported as a mean of three replicates ± standard deviation

Element	**Concentration of Element (ppm)**				
	Asian pigeonwings	**Lemon balm**	**Lemongrass**	**Ginger**	**Turmeric**
Ag	0.000 ±0.000	2.463 ±0.158	8.110 ±0.251	6.009 ±0.159	0.000 ±0.000
Al	41.628 ±1.041	0.000 ±0.000	0.000 ±0.000	4.215 ±0.055	139.970 ±1.209
As	23.626 ±0.831	24.780 ±0.787	68.256 ±0.985	51.424 ±0.472	82.665 ±0.781
B	86.856 ±1.022	114.301 ±2.640	93.194 ±1.127	112.027 ±1.078	79.155 ±1.873
Ba	9.943 ±0.211	12.651 ±0.585	2.378 ±0.277	2.585 ±0.028	2.618 ±0.101
Cd	17.060 ±0.852	1.836 ±0.060	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000
Cu	5.453 ±0.315	10.758 ±0.532	11.703 ±0.359	1.529 ±0.057	6.978 ±0.451
Cr	0.529 ±0.028	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000	2.751 ±0.213
Fe	87.431 ±1.181	19.777 ±0.634	52.927 ±2.100	27.440 ±0.304	12.469 ±0.300
Mn	8.464 ±0.134	7.857 ±0.358	2.640 ±0.354	99.030 ±0.472	4.523 ±0.054
Ni	1.718 ±0.026	23.548 ±0.681	2.491 ±0.261	9.382 ±0.086	15.802 ±0.205
P	639.026 ±4.241	404.341 ±3.061	368.346 ±2.247	892.461 ±4.297	155.308 ±2.081
Pb	53.624 ±0.742	48.135 ±1.372	71.217 ±2.081	84.924 ±1.875	52.148 ±3.102
Sb	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000	0.000 ±0.000	198.873 ±2.229
Se	160.921 ±2.010	17.473 ±0.350	32.215 ± 0.954	14.541 ±0.040	127.252 ±1.785
Zn	20.843 ±0.581	52.314 ±0.681	30.014 ±1.254	39.160 ±0.372	53.534 ±0.716
Ca	693.39 ±3.003	7714.6 ±7.602	1499.0 ±5.655	825.82 ±2.781	1094.5 ±4.552
K	8519.7 ±9.414	9003.2 ±9.318	2756.6 ±3.008	5581.6 ±4.684	9042.7 ±7.584
Mg	964.00 ±4.431	3243.7 ±6.014	284.98 ±2.587	1172.3 ±6.287	1010.3 ±5.282
Na	285.31 ±2.011	294.63 ±1.525	262.17 ±6.572	543.52 ±3.480	484.10 ±3.506

Antioxidant activity

The antioxidant potential of the five plants selected for this study was evaluated by determining the ability of the aqueous and methanolic extracts of each herb to scavenge DPPH radicals by reducing the radical. As shown in Figure 2, the aqueous extracts of all plants, except lemongrass, were more effective in reducing the DPPH radical compared with their methanolic extracts. Among all the extracts, the aqueous extract of ginger had the highest antioxidant activity with a percentage of 88.05 ± 0.31%.

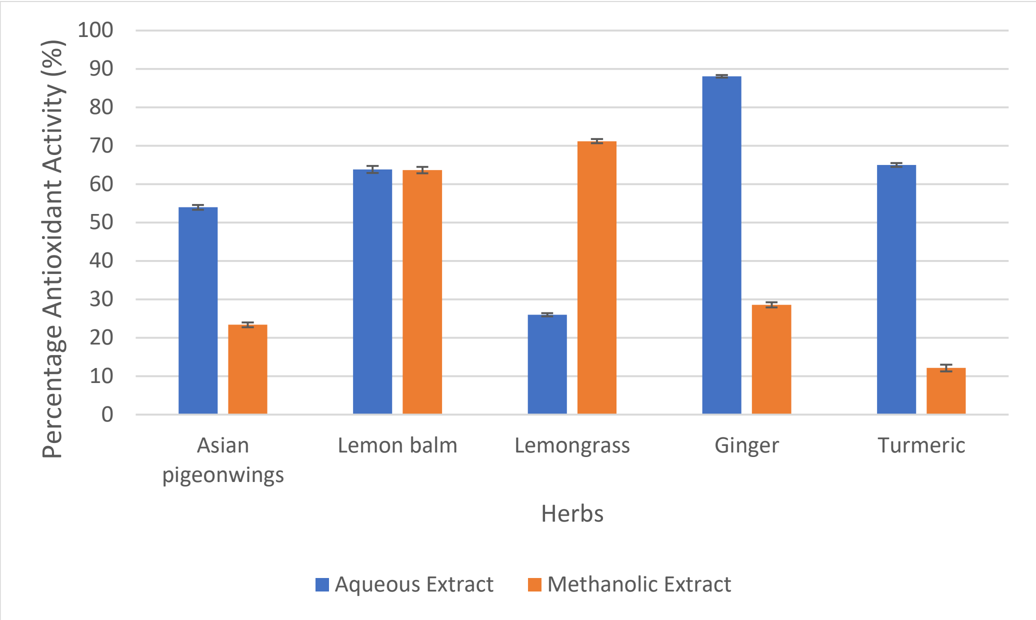

Figure 2 - Antioxidant activity of aqueous and methanolic extracts of plants. The values are reported as a mean of three replicates with error bars representing the standard deviation

Conclusions

According to the phytochemical screening, flavonoids were present in all plants, tannins were present in lemon balm and lemongrass, saponins were present in all plants except lemongrass, and anthraquinone glycosides were detected only in turmeric. Regarding the phenolic content of the plant extracts, the aqueous extract of lemon balm, the aqueous extract of Asian pigeonwings, and the methanolic extract of turmeric had the highest phenolic content, respectively. According to the inorganic composition analysis of plants, Asian pigeonwings, ginger and turmeric were mainly comprised of K, while lemon balm and lemongrass were mainly comprised of Ca. According to the mineral profile, Asian pigeonwings, lemon balm, ginger, and turmeric contained high concentrations of K. High concentrations of Ca and Mg were found in lemon balm, high concentrations of Se, Al, and Sb were determined in turmeric, and high concentrations of Se were determined in Asian pigeonwings. The antioxidant activity of the aqueous and methanolic extracts of plants was determined. Among the extracts, the aqueous extract of ginger had the highest antioxidant activity with a percentage of 88.05 ±0.31 %

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References

[1] F. Al-Deen, 'Evolution of Antibacterial Activity of Various Solvents Extracts of Annona Squamosa Fruit', *Iraqi J. Sci.*, vol. 58, no. 4, pp. 2301-2308, 2018.

[2] M. Sium, P. Kareru, J. Keriko, B. Girmay, G. Medhanie, and S. Debretsion, 'Profile of Trace Elements in Selected Medicinal Plants Used for the Treatment of Diabetes in Eritrea', *Sci. World J.*, vol. 2016, pp. 1-7, 2016, doi: 10.1155/2016/2752836.

[3] R. Chitturi, V. R. Baddam, L. Prasad, L. Prashanth, and K. Kattapagari, 'A review on role of essential trace elements in health and disease', *J. Dr NTR Univ. Health Sci.*, vol. 4, no. 2, pp. 75-85, 2015, doi: 10.4103/2277-8632.158577.

[4] A. Mehri, 'Trace Elements in Human Nutrition (II) - An Update', *Int. J. Prev. Med.*, vol. 11, no. 2, pp. 1-17, 2020.

[5] S. I. Korfali, T. Hawi, and M. Mroueh, 'Evaluation of heavy metals content in dietary supplements in Lebanon', *Chem. Cent. J.*, vol. 7, no. 1, pp. 10-23, Dec. 2013, doi: 10.1186/1752-153X-7-10.

[6] M. E. Wall, M. M. Krider, C. F. Krewson, C. R. Eddy. J. J. Willaman, D. S. Corel, H S Gentry, 'Steroidal sapogenins. VII. Survey of plants for steroidal sapogenins and other constituents', *J. Am. Pharm. Assoc.*, vol. 53, no. 1, pp. 1-7, 1954.

[7] A. Hassan, F. Al-Salman, A. Ali Redha, M. Salem, and Z. Saeed, 'PHYTOCHEMICAL INVESTIGATIONS OF 10 EDIBLE PLANTS AND THEIR ANTIOXIDANT AND ANTIDIABETIC ACTIVITY', *Int. J. Res. Pharm. Chem.*, vol. 10, no. 3, pp. 260-272, 2020.

[8] A. A. Redha, A. M. Hasan, and Q. Mandeel, 'Phytochemical Determinations of Pomegranate (Punica granatum) Rind and Aril Extracts and their Antioxidant, Antidiabetic and Antibacterial Activity', *Nat. Prod. Chem. Res.*, vol. 06, no. 04, pp. 332-341, 2018, doi: 10.4172/2329-6836.1000332.

[9] A. A. Redha, 'Phytochemical Investigations of Nerium Oleander L. Leaves and Flowers', *Int. J. Sci. Res. Chem. Sci.*, vol. 7, no. 4, pp. 1-4, 2020.

[10] F. Al-Salman, A. Ali Redha, and Z. Al-Zaimoor, 'Inorganic Analysis and Antioxidant Activity of Shilajit', *Int. J. Sci. Res. Chem. Sci.*, vol. 7, no. 3, pp. 5-10, 2020.

[11] H. G. Bulama, D. Dahiru, and J. O. Madu, 'Investigation of the Anti-cataract and Anti-oxidant Activties of Cnidoscolus aconitifolius Leaves Extract In vitro', *Iraqi J. Sci.*, vol. 62, no. 1, pp. 28-38, 2021.

[12] A. Smeriglio, D. Barreca, E. Bellocco, and D. Trombetta, 'Proanthocyanidins and hydrolysable tannins: occurrence, dietary intake and pharmacological effects: Pharmacological aspects of tannins', *Br. J. Pharmacol.*, vol. 174, no. 11, pp. 1244-1262, Jun. 2017, doi: 10.1111/bph.13630.

[13] K. S. S. Sangeetha, S. Umamaheswari, C. U. M. Reddy, and S. N. Kalkura, 'FLAVONOIDS: THERAPEUTIC POTENTIAL OF NATURAL PHARMACOLOGICAL AGENTS', *Int. J. Pharm. Sci. Res.*, vol. 7, no. 10, pp. 3924-3930.

[14] A. D. P. Barbosa, 'AN OVERVIEW ON THE BIOLOGICAL AND PHARMACOLOGICAL ACTIVITIES OF SAPONINS', vol. 6, no. 8, pp. 47-50, 2014.

[15] J. Duval, V. Pecher, M. Poujol, and E. Lesellier, 'Research advances for the extraction, analysis and uses of anthraquinones: A review', *Ind. Crops Prod.*, vol. 94, pp. 812-833, Dec. 2016, doi: 10.1016/j.indcrop.2016.09.056.

[16] J. A. Beto, 'The Role of Calcium in Human Aging', *Clin. Nutr. Res.*, vol. 4, no. 1, pp. 1-8, 2015, doi: 10.7762/cnr.2015.4.1.1.

[17] G. J. Hofmeyr, T. A. Lawrie, Á. N. Atallah, and M. R. Torloni, 'Calcium supplementation during pregnancy for preventing hypertensive disorders and related problems', *Cochrane Database Syst. Rev.*, vol. 10, pp. 1-123, Oct. 2018, doi: 10.1002/14651858.CD001059.pub5.

[18] Y. Sun, C. H. Byon, Y. Yang, W. E. Bradley, L. J. Dell'Italia, P. W. Sanders, A. Agarwal, H. Wu, and Y. Chen, 'Dietary potassium regulates vascular calcification and arterial stiffness', *JCI Insight*, vol. 2, no. 19, pp. e94920, Oct. 2017, doi: 10.1172/jci.insight.94920.

[19] K. H. Mun, G. I. Yu, B. Y. Choi, M. K. Kim, M.-H. Shin, and D. H. Shin, 'Association of Dietary Potassium Intake with the Development of Chronic Kidney Disease and Renal Function in Patients with Mildly Decreased Kidney Function: The Korean Multi-Rural Communities Cohort Study', *Med. Sci. Monit.*, vol. 25, pp. 1061-1070, Feb. 2019, doi: 10.12659/MSM.913504.

[20] X. Fang, K. Wang, D. Han, X. He, J. Wei, L. Zhao, M. U. Imam, Z. Ping, Y. Li, Y. Xu, J. Min, and F. Wang, 'Dietary magnesium intake and the risk of cardiovascular disease, type 2 diabetes, and all-cause mortality: a dose–response meta-analysis of prospective cohort studies', *BMC Med.*, vol. 14, no. 1, pp. 210-222, Dec. 2016, doi: 10.1186/s12916-016-0742-z.

[21] U. Gröber, J. Schmidt, and K. Kisters, 'Magnesium in Prevention and Therapy', *Nutrients*, vol. 7, no. 9, pp. 8199-8226, Sep. 2015, doi: 10.3390/nu7095388.

[22] N. Wang, H.-Y. Tan, S. Li, Y. Xu, W. Guo, and Y. Feng, 'Supplementation of Micronutrient Selenium in Metabolic Diseases: Its Role as an Antioxidant', *Oxid. Med. Cell. Longev.*, vol. 2017, pp. 1-13, 2017, doi: 10.1155/2017/7478523.

[23] S. Sundar and J. Chakravarty, 'Antimony Toxicity', *Int. J. Environ. Res. Public. Health*, vol. 7, no. 12, pp. 4267-4277, Dec. 2010, doi: 10.3390/ijerph7124267.

[24] C. Bakar, H. I. Ö. Karaman, A. Baba, and F. Şengünalp, 'Effect of High Aluminum Concentration in Water Resources on Human Health, Case Study: Biga Peninsula, Northwest Part of Turkey', *Arch. Environ. Contam. Toxicol.*, vol. 58, no. 4, pp. 935-944, May 2010, doi: 10.1007/s00244-009-9435-3.