Arabian Journal of Chemistry (2021) **14**, 103055

King Saud University

Arabian Journal of Chemistry

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ORIGINAL ARTICLE

Quantification of macro, micro and trace elements, and antimicrobial activity of medicinal herbs and their products

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Received 2 December 2020; accepted 24 January 2021
Available online 4 February 2021

KEYWORDS

Herbs;
Pakistan;
Trace elements;
Inductively coupled plasma-optical emission spectroscopy;
Antimicrobial

Abstract The study describes the content of macro, micro, and trace essential and toxic elements in thirteen medicinal herbs and their products including *Acorus calamus*, *Blepharis edulis*, *Caesalpinia bonducella*, *Curculigo orchioides*, *Helicteres isora*, *Holarrhena pubescens*, *Pastinaca sativa*, *Pistacia integerrima*, *Quercus infectoria*, *Rauwolfia serpentina*, *Saussurea lappa*, *Teucrium stocksianum*, and *Xanthium strumarium* available in the local markets of Pakistan. The elemental content were analyzed with the techniques of inductively coupled plasma (ICP) optical emission spectroscopy (OES) and ICP-mass spectrometry (MS). Furthermore, their antibacterial and antifungal activities were evaluated against the selected microbial pathogens including *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Escherichia coli*, *Candida albicans*, *Candida krusei*, *Aspergillus flavus*, and *Trichophyton mentagrophytes*. Among macro elements, K and Ca showed the highest content, micro elements were in the order of Rb^{85}/ Sr^{87} > Zn^{64}/Cu^{63} > Ni^{60}, and among essential trace elements, the content of Cr^{52}/Cr^{53} and Co^{59} were high. The content of the analyzed toxic elements were lower than the permissible standard values. The antimicrobial activities against the subject strains were

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.arabjc.2021.103055

significant with inhibition zones of 7.0-19.0 mm in disc diffusion procedure, and 62.5–1000 μg/mL in minimum inhibitory concentration method. Hence, the presence of nutritional elements at appreciable concentrations, toxic elements within permissible ranges, and significant antimicrobial potential assume the subject herbs as promising nutritional and therapeutic remedies.

1. Introduction

Plants, either herbs, fruits or vegetables in the form of food and supplement are the main source of bioactive components and inorganic elements, and traditionally used to cure different diseases (Dall'Acqua et al., 2009; Billah et al., 2013; Peng et al., 2014; Jamila et al., 2016). In today's market, a large number of medicinal herbs and their formulations are available to treat various diseases. For example, the species; *Acorus calamus*, *Blepharis edulis*, *Caesalpinia Bonducella*, *Cuculigo orchioides*, *Helicteres isora, Holarrhena pubescens*, *Pastinaca sativa*, *Pistacia integrima, Quercus infectoria*, *Rauwolfia serpentina*, *Sassurea lappa*, *Teucrium stocksianum*, and *Xantium stumarium* and their products are found in the Pansaar/Hakeems' shops throughout Pakistan, which are used by the local people for the treatment of ulcer, diarrhea, digestive disorders, asthma, cough, diabetes, cardiovascular and as tonic (Table S1, supplementary material). Medicinal herbs are cost effective and easily accessible, and therefore a large number of Pakistani population specifically local segments, rely on these medicinal herbs. *A. calamus* is an aromatic plant playing significant role in the nervous system disorders, removal of fat, hemorrhoids, nasal problems, colic pain, diabetes, bronchitis, and skin diseases having anti-inflammatory, antidiabetic, and antimicrobial properties (Kim et al., 2009; Wu et al., 2009). *B. edulis* distributed throughout Pakistan, Iran, Afghanistan, and India is sold in markets as diuretic, expectorant, and wounds healing agent, showing significant antimicrobial, anticancer, and antiplatelet aggregation activities (Mahboubi et al., 2013). *C. bonducella* is utilized to cure tumors, inflammation, and liver disorders possessing antidiarrheal, antimicrobial, antidiabetic, antitumor, anti-inflammatory, and antioxidant properties (Billah et al., 2013). *C. orchioides* is locally used for jaundice, asthma, male sex dysfunction, bleeding, injuries, and to regulate menstrual cycle and possesses hepatoprotective, cytotoxic, and anticonvulsive properties (Wu et al., 2005; Dall'Acqua et al., 2009; Nie et al., 2013). *H. isora* is used to treat diarrhea, snake biting, and constipation (Kumar and Singh, 2014). *H. pubescens* is useful in amoebic dysentery, spleen and chest infections (Tuntiwachwuttikul et al., 2007; Yang et al., 2012). *P. sativa* is known as tonic, anti-inflammatory, and carminative (Waksmundzka-Hajnos et al., 2004). *P. integerrima* has been reported to reduce inflammation and gastrointestinal disorder having anticancer, anti-inflammatory, and leishmanicidal activities (Ahmad et al., 2008; Rauf et al., 2014). *Q. infectoria* is used as wound healing, in digestive disorders, kidneys, dental, and vaginal tightening in Asia (Fan et al., 2014; Kaur et al., 2004). *R. serpentina* is widely utilized as antihypertensive and tranquilizing agent to cure schizophrenia, cholera, colic, and snakebite (Bharti et al., 2017). *S. lappa* is used to cure skin allergies, asthma, carminative, and anthelmintic (Robinson et al., 2008; Julianti et al., 2011). *T. stocksianum* has been used in stomachic, diarrhea, diabetes, gastrointestinal ailments, and sore throat having anti-inflammatory activity (Bakhtiari and Asgarpanah, 2015; Shah and Shah, 2015). *X. strumariam* is taken in asthma, nasal sinusitis, and headache (Han et al., 2007; Peng et al., 2014). The detailed phytochemical and pharmacological profile of these medicinal herbs is given in Table S1, supplementary material.

Plants are the main source of several elements, which are essential for human beings. The elements when present in large quantity (100 mg/g) are known as macronutrients/major elements, those in small amounts (>1 μg/g) referred as micronutrients or minor elements or those present in trace amount (<1 μg/g) are called trace elements. These elements have major functions in metabolic cycles and inhibition of diseases within their recommended and permissible ranges. Therefore, to fulfil the nutritional requirement, plants and herbs should be used in the daily diet. In addition, consumers have increased interest to choose the diet having high nutrient levels, preferably from natural sources (Asioli et al., 2017). However, nowadays, the use of fertilizers and pesticides have been considerably contributed to the increased level of non-biodegradable toxic metals such as arsenic, cadmium, lead, and mercury in the soil and crops, which are accumulated by vegetables, fruits and medicinal plants/herbs (Chen et al., 2014; Bloise et al., 2016, 2020). In products formulation and synthesis, various processes including extraction, dehydration, refrigeration, preservation, packaging and storage can cause contamination of the products with toxic metals (Abbasi et al., 2020). For example, several Indian and Chinese herbal medicines contained lead, arsenic, and mercury more than the permissible limits, which caused poisoning to human (Saeed et al., 2011; Shen et al., 2012). The excessive accumulation of these metals in food and herbal medicine may pose serious health risks to humans upon consumption. Hence, besides the market quality attributes, the determination of content levels (permissible/impermissible) of toxic metals, and safety of the medicinal herbs and their products need to be investigated.

Different analytical techniques such as atomic absorption spectrometry, inductively coupled plasma optical emission spectroscopy (ICP-OES), and inductively coupled plasma mass spectrometry (ICP-MS) have been applied for content determination of trace essential and toxic elements in environmental and biological samples (Huang et al., 2007; Ivanova-Petropulos et al., 2015; Park et al., 2018). Among these methods, ICP-MS and ICP-OES are efficient and accurate techniques with low detection limits and wider linear dynamic range (Park et al., 2018; Jamila et al., 2019, 2020).

Considering the scientific and consumers' demand predominantly the toxicity associated with heavy metals, the present study aimed to analyze medicinal herbs including *A. calamus*,

Table 1 Mean concentration (μg/g, dry weight basis) of macro, micro, trace essential, nontoxic, and toxic elements in the medicinal herbs used in Pakistan.

Elements	*A. calamus*	*B. edulis*	*C. Bonducella*	*C. orchioides*	*H. isora*	*H. pubescens*	*P. integerrima*	*P. sativa*	*Q. infectoria*	*R. serpentina*	*S. lappa*	*T. stocksianum*	*X. strumarium*
Macro elements													
Al	308 ± 21.3	33.1 ± 6.26	24.8 ± 6.39	3188.5 ± 152.7	2.5 ± 0.68	853.6 ± 197.6	114.8 ± 18.6	728.6 ± 24.6	32.4 ± 8.7	785.7 ± 46.0	174.5 ± 24.3	2712.3 ± 448.3	893.6 ± 211.1
Ca	4634.9 ± 136.2	155.2 ± 11.6	1181.0 ± 44.6	16293.6 ± 293.3	141.9 ± 1.44	9409.0 ± 221.6	1918.5 ± 37.0	6306.3 ± 46.0	1426.0 ± 12.4	6515.8 ± 562.3	14984.7 ± 765.8	12569.3 ± 741.6	6012.8 ± 284.8
Fe	758.3 ± 34.5	29.9 ± 2.16	56.2 ± 1.97	225.3 ± 126.2	2.85 ± 0.408	1063.5 ± 230.2	39.8 ± 5.39	776.6 ± 17.3	16.6 ± 2.45	630.7 ± 37.3	234.9 ± 16.2	3067.2 ± 554.0	733.8 ± 76.4
K	29748.0 ± 970.5	400.4 ± 1.48	15887.6 ± 244.0	10249.1 ± 384.1	523.6 ± 13.7	10336.5 ± 233.2	24556.5 ± 616.2	8180.8 ± 193.7	6821.6 ± 76.5	10256.4 ± 492.1	7591.0 ± 283.8	15509.6 ± 735.2	7341.1 ± 143.6
Mg	1291.0 ± 26.5	68.2 ± 1.45	1015.9 ± 32.0	2728.8 ± 84.9	104.5 ± 6.87	3048.7 ± 31.6	727.8 ± 50.4	1549.8 ± 145.7	411.9 ± 2.29	1157.1 ± 33.6	2771.3 ± 189.6	3143.4 ± 207.8	1382.3 ± 26.5
Na	2439.6 ± 105.8	8.96 ± 0.770	14.0 ± 3.62	428.3 ± 36.5	2.99 ± 0.586	94.3 ± 12.4	79.2 ± 25.8	76.5 ± 8.2	73.5 ± 0.403	196.3 ± 23.0	29.9 ± 7.08	1515.9 ± 84.9	177.6 ± 82.5
P	6372.6 ± 77.9	114.7 ± 4.45	2653.5 ± 91.5	1105.9 ± 26.7	67.5 ± 6.58	5586.3 ± 233.2	2737.7 ± 57.9	999.7 ± 40.9	565.4 ± 12.4	2131.2 ± 159.0	1495.4 ± 49.2	2045.6 ± 83.7	725.6 ± 5.85
S	1385.9 ± 35.5	14.8 ± 0.415	362.7 ± 7,03	282.5 ± 9.70	11.0 ± 0.841	1111.2 ± 12.1	188.2 ± 5.70	242.7 ± 20.2	80.9 ± 4.75	573.4 ± 26.3	1754.5 ± 102.4	616.8 ± 32.9	178.3 ± 0.858
Micro elements													
Ni^{60}	1.30 ± 0.052	0.095 ± 0.016	2.01 ± 0.129	7.17 ± 0.171	0.080 ± 0.019	8.21 ± 0.760	0.8500.061 ±	4.94 ± 0.382	3.01 ± 0.278	7.09 ± 0.042	4.61 ± 0.246	6.36 ± 0.613	5.93 ± 0.507
Cu^{63}	8.21 ± 0.137	0.554 ± 0.045	9.30 ± 0.305	6.90 ± 0.230	0.206 ± 0.022	10.5 ± 0.764	2.60 ± 0.088	4.78 ± 0.127	4.36 ± 0.194	9.48 ± 0.442	0.824 ± 0.007	8.03 ± 0.092	9.59 ± 0.213
Zn^{64}	17.1 ± 0.968	0.856 ± 0.024	23.2 ± 0.813	50.8 ± 0.830	0.861 ± 0.086	41.2 ± 1.30	7.91 ± 2.89	23.2 ± 1.89	3.91 ± 1.45	20.23 ± 0.779	9.53 ± 0.453	31.2 ± 0.524	26.9 ± 0.676
Sr^{87}	162.8 ± 7.23	0.649 ± 0.043	122.9 ± 2.05	416.1 ± 10.7	8.68 ± 0.342	180.0 ± 15.0	184.2 ± 11.2	62.4 ± 25.4	113.7 ± 5.74	164.5 ± 7.79	417.1 ± 15.1	140.6 ± 2.45	192.1 ± 8.43
Rb^{85}	83.7 ± 4.75	1.45 ± 0.309	137.5 ± 2.75	284.0 ± 5.56	8.18 ± 0.290	145.7 ± 13.6	215.9 ± 11.9	6.79 ± 0.186	124.1 ± 7.37	13.1 ± 7.77	80.3 ± 1.63	67.4 ± 2.18	106.5 ± 5.55
Trace essential elements													
V^{51}	0.95 ± 0.03	0.10 ± 0.002	0.09 ± 0.001	5.78 ± 0.386	0.01 ± 0.0009	2.84 ± 0.482	0.10 ± 0.003	2.57 ± 0.16	0.12 ± 0.002	1.73 ± 0.089	0.722 ± 0.119	5.25 ± 0.880	2.54 ± 0.31
Cr^{52}	1.21 ± 0.090	0.295 ± 0.108	3.70 ± 0.529	7.957 ± 0.365	0.040 ± 0.004	89.2 ± 15.3	0.777 ± 0.144	5.62 ± 0.037	0.526 ± 0.127	33.7 ± 6.97	1.60 ± 0.120	8.08 ± 0.857	3.76 ± 0.430
Cr^{53}	1.13 ± 0.092	0.214 ± 0.004	2.40 ± 0.552	8.66 ± 0.412	0.032 ± 0.004	21.0 ± 3.70	0.506 ± 0.130.6	5.83 ± 0.395	0.445 ± 0.137	10.7 ± 0.772	1.45 ± 0.108	7.87 ± 0.926	3.27 ± 0.404
Co^{59}	0.697 ± 0.015	0.011 ± 0.001	0.418 ± 0.004	1.359 ± 0.084	0.003 ± 0.0003	0.496 ± 0.039	0.025 ± 0.0004	0.461 ± 0.028	0.087 ± 0.021	0.506 ± 0.042	0.171 ± 0.049	0.736 ± 0.128	0.771 ± 0.033
Se^{82}	0.033 ± 0.010	0.001 ± 0.0002	0.051 ± 0.003	1.44 ± 0.194	0.0006 ± 0.0001	0.052 ± 0.008	0.010 ± 0.001	0.190 ± 0.041	0.051 ± 0.011	0.178 ± 0.037	0.088 ± 0.018	0.833 ± 0.224	0.016 ± 0.009
Trace non toxic elements													
Li^{7}	0.196 ± 0.012	0.051 ± 0.003	0.167 ± 0.008	3.20 ± 0.224	0.006 ± 0.0006	0.702 ± 0.151	0.070 ± 0.002	3.61 ± 0.211	4.45 ± 0.424	2.20 ± 0.144	0.427 ± 0.101	7.12 ± 1.12	1.74 ± 0.281
Ga^{69}	0.738 ± 0.049	0.013 ± 0.002	0.117 ± 0.009	2.63 ± 0.041	0.005 ± 0.0003	0.626 ± 0.030	0.110 ± 0.023	1.79 ± 0.045	0.273 ± 0.067	0.698 ± 0.048	0.889 ± 0.058	0.756 ± 0.136	2.86 ± 0.207
Trace toxic elements													
As^{75}	0.212 ± 0.044	0.008 ± 0.002	0.026 ± 0.002	0.189 ± 0.012	0.001 ± 0.00008	0.091 ± 0.005	0.018 ± 0.002	0.221 ± 0.017	0.040 ± 0.008	0.168 ± 0.020	0.063 ± 0.015	0.899 ± 0.127	0.159 ± 0.042
Cd^{111}	0.021 ± 0.001	0.0004 ± 0.00004	0.014 ± 0.001	0.028 ± 0.008	0.0008 ± 0.000004	0.019 ± 0.002	0.002 ± 0.0009	0.025 ± 0.003	0.022 ± 0.003	0.021 ± 0.001	0.041 ± 0.009	0.088 ± 0.0002	0.052 ± 0.002
In^{115}	0.009 ± 0.004	0.0002 ± 0.00004	0.0001 ± 0.00001	0.004 ± 0.0001	0.0003 ± 0.000001	0.011 ± 0.0001	0.012 ± 0.00002	0.002 ± 0.0003	0.071 ± 0.006	0.008 ± 0.002	0.054 ± 0.011	0.003 ± 0.0007	0.010 ± 0.0002
Cs^{133}	0.019 ± 0.001	0.008 ± 0.00003	0.027 ± 0.0002	0.526 ± 0.021	0.005 ± 0.0003	0.182 ± 0.057	0.355 ± 0.131	0.236 ± 0.011	0.367 ± 0.020	0.295 ± 0.018	0.171 ± 0.0008	0.506 ± 0.088	266.8 ± 0.035
Ba^{138}	418.8 ± 9.72	3.25 ± 0.085	5.35 ± 0.315	1086.0 ± 187.9	3.83 ± 0.166	375.7 ± 44.2	5.18 ± 1.02	772.3 ± 34.2	102.0 ± 5.60	205.4 ± 15.3	680.5 ± 23.1	182.5 ± 22.0	1829.9 ± 67.8
Be^{9}	0.003 ± 0.0006	0.0004 ± 0.00007	0.0009 ± 0.0002	0.044 ± 0.002	0.00005 ± 0.000008	0.016 ± 0.002	0.00004 ± 0.00001	0.018 ± 0.001	0.025 ± 0.003	0.016 ± 0.0009	0.017 ± 0.003	0.038 ± 0.0007	0.016 ± 0.004
Tl^{205}	0.028 ± 0.008	0.0003 ± 0.00004	0.005 ± 0.0002	0.043 ± 0.001	0.0006 ± 0.0001	0.007 ± 0.001	0.007 ± 0.0001	0.015 ± 0.0004	0.080 ± 0.016	0.033 ± 0.002	0.057 ± 0.090	0.020 ± 0.003	0.024 ± 0.001
Pb^{206}	2.99 ± 0.260	0.051 ± 0.006	4.58 ± 0.465	1.24 ± 0.069	0.040 ± 0.022	0.872 ± 0.068	0.429 ± 0.060	1.98 ± 0.168	0.917 ± 0.242	0.638 ± 0.064	2.15 ± 0.061	4.51 ± 0.937	1.84 ± 0.190
Pb^{208}	3.03 ± 0.274	0.051 ± 0.005	4.86 ± 0.461	1.28 ± 0.067	0.039 ± 0.023	0.833 ± 0.077	0.419 ± 0.063	2.04 ± 0.189	0.933 ± 0.241	0.638 ± 0.060	2.14 ± 0.051	4.80 ± 1.12	1.82 ± 0.199
U^{238}	0.038 ± 0.004	0.002 ± 0.00008	0.002 ± 0.0001	0.047 ± 0.002	0.000009 ± 0.000009	0.181 ± 0.015	0.005 ± 0.0003	0.059 ± 0.007	0.0001 ± 0.00003	0.091 ± 0.008	0.002 ± 0.0009	0.004 ± 0.001	0.038 ± 0.009

B. edulis, *C. Bonducella*, *C. orchioides*, *H. isora, H. pubescens*, *P. sativa*, *P. integerrima, Q. infectoria*, *R. serpentina*, *S. lappa*, *T. stocksianum*, *X. strumarium*, and their products available in the local markets of Khyber Pakhtunkhwa, Pakistan. The content of macronutrients (Al, Ca, Fe, K, Mg, Na, P, S), micronutrients (Cu, Ni, Rb, Sr, Zn), trace essential nutrients (Co, Cr, Se, V), trace non-toxic (Ga, Li), and toxic elements (As, Ba, Be, Cd, Cs, In, Pb, Tl, U) were determined. The applied ICP-OES and ICP-MS techniques were validated by quality parameters; limits of detection (LOD), limits of quantification (LOQ), precision (%RSD), spiking recovery tests, analyzing the certified reference materials (NIST-1570a, spinach leaves), and by participation in accredited laboratory proficiency test (inter laboratories calibrations) organized by Food Analysis Performance Assessment Scheme (FAPAS). Plants are the potential sources of antimicrobial drugs against microbial pathogens (Shin et al., 2018; Hammerbacher et al., 2019). Therefore, the current study in addition to the elemental content determination, reports the antibacterial and antifungal activities of the subject medicinal herbs ad their related products.

2. Materials and methods

2.1. Samples collection

A. calamus (root), *B. edulis* (seeds), *C. Bonducella* (seeds), *C. orchioides* (root), *H. isora* (fruit), *H. pubescens* (seeds), *P. sativa* (root), *P. integerrima* (fruit), *Q. infectoria* (galls), *R. serpentina* (root), *S. lappa* (root), *T. stocksianum* (aerial part), and *X. strumarium* (leaves) and their products (P1-P39) in triplicate, were collected and procured from three different markets of Peshawar, Khyber Pakhtunkhwa, during January-June 2019. The samples were identified by a Taxonomist from Kohat University of Science and Technology, Kohat, and the voucher specimen are deposited at the Herbarium of Department of Botany.

Table 2 Mean concentration (μg/g, dry weight basis) of macro elements of the analyzed herbal products available in local markets of Pakistan.

Herb	Product	Al	Ca	K	Fe	Mg	Na	P	S
A. calamus	P1	18.7 ± 1.44	196.7 ± 6.08	219.7 ± 8.19	116.8 ± 1.01	186.3 ± 1.17	179.5 ± 3.03	39.3 ± 0.173	138.9 ± 2.78
	P2	93.7 ± 3.17	119.4 ± 2.09	210.0 ± 1.43	104.9 ± 0.91	389.2 ± 11.9	215.8 ± 6.49	59.8 ± 1.60	219.7 ± 10.5
	P3	29.5 ± 3.02	194.3 ± 11.6	307.1 ± 7.91	148.4 ± 5.79	201.8 ± 5.19	172.4 ± 3.82	48.7 ± 1.03	176.0 ± 5.29
B. edulis	P4	28.3 ± 1.16	186.4 ± 7.18	227.9 ± 5.05	167.2 ± 5.19	56.9 ± 0.91	17.6 ± 0.59	38.9 ± 1.00	85.3 ± 1.03
	P5	50.5 ± 2.15	169.2 ± 3.19	257.7 ± 4.41	94.7 ± 1.04	79.0 ± 3.17	37.6 ± 1.49	50.3 ± 1.03	106.8 ± 4.19
	P6	38.3 ± 0.916	119.4 ± 3.00	301.2 ± 11.0	108.9 ± 1.38	84.0 ± 2.11	28.9 ± 1.11	41.7 ± 1.16	95.0 ± 1.93
C. bonducella	P7	101.8 ± 1.59	69.6 ± 3.10	206.7 ± 11.9	17.9 ± 0.917	191.4 ± 8.29	95.7 ± 5.82	71.5 ± 1.72	18.5 ± 0.617
	P8	104.6 ± 7.13	58.4 ± 1.53	147.1 ± 11.3	42.8 ± 4.19	115.8 ± 4.72	100.6 ± 5.16	25.9 ± 2.91	32.6 ± 1.88
	P9	131.7 ± 1.34	37.2 ± 1.10	151.5 ± 5.07	15.0 ± 1.93	93.7 ± 4.03	69.3 ± 3.07	17.5 ± 1.03	11.3 ± 1.08
C. orchioides	P10	73.2 ± 1.88	232.2 ± 7.93	531.7 ± 16.1	104.9 ± 6.17	128.4 ± 11.8	39.0 ± 5.09	48.1 ± 5.08	101.6 ± 11.0
	P11	91.9 ± 1.13	159.6 ± 4.45	363.8 ± 12.9	248.4 ± 15.9	164.4 ± 16.9	61.8 ± 3.81	79.9 ± 7.16	90.0 ± 4.79
	P12	58.6 ± 6.10	139.7 ± 13.7	293.1 ± 9.15	293.2 ± 21.7	175 ± 11.9	64.4 ± 5.23	91.9 ± 7.86	106.9 ± 12.2
H. isora	P13	2.10 ± 5.43	347.2 ± 15.3	229.5 ± 14.1	115.9 ± 15.8	188.3 ± 29.0	371.7 ± 28.3	19.0 ± 1.15	97.6 ± 19.5
	P14	3.27 ± 0.941	323.1 ± 16.0	199.2 ± 7.88	110.8 ± 12.3	200.7 ± 10.7	314.3 ± 21.6	26.3 ± 1.63	99.3 ± 16.0
	P15	5.51 ± 1.16	286.9 ± 13.9	217.3 ± 2.05	91.7 ± 7.65	214.6 ± 3.76	165.2 ± 12.6	23.2 ± 1.83	114.7 ± 11.7
H. pubescens	P16	5.99 ± 1.34	84.9 ± 1.41	277.8 ± 5.49	127.6 ± 9.51	381.1 ± 4.79	193.7 ± 10.2	181.3 ± 8.45	93.7 ± 9.89
	P17	4.29 ± 1.61	94.8 ± 0.858	191.8 ± 11.3	113.5 ± 7.32	279.3 ± 13.4	178.3 ± 4.99	172.8 ± 11.7	113.5 ± 11.9
	P18	9.17 ± 2.70	110.1 ± 14.3	266.9 ± 18.9	98.9 ± 9.94	237.3 ± 12.6	160.5 ± 12.9	235.2 ± 27.3	87.5 ± 8.39
P. integerrima	P19	11.7 ± 2.46	212.4 ± 9.99	391.2 ± 12.1	194.2 ± 9.18	254.5 ± 11.2	211.2 ± 3.99	283.8 ± 20.7	71.9 ± 10.5
	P20	21.2 ± 2.46	194.7 ± 11.0	423.7 ± 16.6	236.8 ± 15.0	326.1 ± 13.7	143.9 ± 12.6	307.5 ± 19.0	94.9 ± 7.91
	P21	32.8 ± 4.86	159.5 ± 14.4	352.8 ± 20.3	147.9 ± 16.7	366.4 ± 9.59	217.3 ± 10.5	123.6 ± 13.5	108.6 ± 10.1
P. sativa	P22	53.8 ± 2.95	71.5 ± 4.02	472.4 ± 13.6	79.6 ± 8.17	243.5 ± 10.4	154.1 ± 12.8	73.0 ± 6.29	51.9 ± 3.06
	P23	63.4 ± 6.15	97.8 ± 7.51	630.5 ± 27.8	49.0 ± 4.52	193.6 ± 12.6	119.4 ± 9.69	105.4 ± 10.2	39.8 ± 3.17
	P24	48.7 ± 3.16	104.7 ± 14.3	428.6 ± 28.9	103.7 ± 11.3	236.4 ± 9.93	138.2 ± 12.9	127.2 ± 16.8	51.3 ± 9.74
Q. infectoria	P25	58.2 ± 7.98	285.7 ± 13.8	908.7 ± 73.5	197.9 ± 14.5	178.1 ± 15.5	205.6 ± 11.2	155.6 ± 15.3	98.1 ± 9.26
	P26	61.9 ± 7.39	158.5 ± 12.9	715.4 ± 71.3	254.1 ± 10.7	213.5 ± 12.3	241.5 ± 28.1	194.2 ± 13.8	139.0 ± 19.4
	P27	28.0 ± 2.95	170.3 ± 13.8	594.4 ± 41.8	295.7 ± 21.3	22.9^{e} ± 1.39	50.8^{d} ± 3.98	117.8 ± 9.01	172.6 ± 20.8
R. serpentina	P28	104.7 ± 23.9	481.5 ± 63.7	140.5 ± 11.8	194.8 ± 12.6	103.7 ± 9.78	37.7 ± 4.98	59.0 ± 6.44	84.2 ± 10.3
	P29	92.3 ± 9.6	355.0 ± 25.0	112.6 ± 19.3	238.9 ± 17.8	117.5 ± 12.7	39.5 ± 3.13	71.3 ± 7.99	67.2 ± 5.87
	P30	99.3 ± 7.36	265.6 ± 14.7	116.6 ± 15.8	211.4 ± 16.9	138.3 ± 13.8	45.9 ± 6.70	63.8 ± 8.15	59.9 ± 7.16
S. lappa	P31	16.8 ± 0.916	56.8 ± 5.8	271.3 ± 57.3	98.4 ± 11.7	112.2 ± 1.96	51.9 ± 3.73	31.8 ± 1.03	52.9 ± 2.85
	P32	37.8 ± 6.82	49.5 ± 2.96	272.5 ± 15.6	127.9 ± 13.8	206.7 ± 26.6	79.9 ± 5.24	27.4 ± 1.58	63.7 ± 7.47
	P33	20.6 ± 2.50	91.5 ± 7.85	167.6 ± 17.0	100.5 ± 8.04	203.4 ± 17.6	71.8 ± 5.78	36.9 ± 4.19	50.5 ± 2.03
T. stocksianum	P34	105.5 ± 9.33	80.6 ± 6.02	941.9 ± 57.3	73.2 ± 5.81	531.6 ± 65.4	401.9 ± 43.5	137.8 ± 14.6	118.6 ± 11.4
	P35	99.6 ± 7.62	104.7 ± 11.3	888.5 ± 94.9	58.3 ± 6.14	396.1 ± 21.8	430.8 ± 37.4	103.7 ± 11.9	144.7 ± 12.9
	P36	87.9 ± 9.18	118.3 ± 15.8	885.1 ± 78.6	63.7 ± 7.72	305.3 ± 35.9	413.4 ± 26.9	99.6 ± 9.36	102.7 ± 11.1
X. strumarium	P37	116.8 ± 8.26	482.3 ± 86.2	407.6 ± 31.9	79.5 ± 8.16	378.8 ± 27.5	693.7 ± 56.0	72.4 ± 8.13	47.3 ± 4.11
	P38	91.7 ± 7.31	493.4 ± 71.9	469.4 ± 87.8	65.8 ± 8.29	329.9 ± 22.69	562.0 ± 35.6	65.0 ± 7.14	39.6 ± 5.11
	P39	77.3 ± 6.94	406.4 ± 31.9	519.4 ± 57.8	53.8 ± 7.35	311.6 ± 38.6	650.8 ± 60.2	55.7 ± 6.71	44.8 ± 3.19

2.2. Chemicals and instrumentation

Chemicals for the samples digestion and elemental analysis; nitric acid (HNO_3), hydrogen peroxide (H_2O_2), ultra-pure deionized water (>18.0 MΩ.cm), multi-element standards (1000 mg/L and 10 mg/L), and standard reference material (SRM-1570a, spinach leaves) were purchased from Dongwoo, Fine-Chem (Korea), Millipore (Bedford, MA, USA), Perkin Elmer (CT, USA), and National Institute of Standards and Technology, NIST (Gaithersburg, MD, USA). For samples digestion and elemental profile, a microwave reaction system (Analytik Jena Topwave 3000, Austria), an ICP-OES (Optima 8000), and ICP-MS (300D) were used under the conditions established by our laboratory (Park et al., 2018; Jamila et al., 2019). Microbial strains; *Staphylococcus aureus* (ATCC 29213), *Bacillus subtilis* (ATCC 19659), *Pseudomonas aeruginosa* (ATCC 17588), *Escherichia coli* (ATCC 25922), *Candida albicans* (ATCC90029), *Candida krusei* (ATCC6258), *Aspergillus flavus* (ATCC9807), and *Trichophyton mentagrophytes* (ATCC40004), nutrient agar (Muller Hinton Broth), *p*-iodonitrotetrazolium chloride (INT), vancomycin, streptomycin, fluconazole, and amphotericin were purchased from Oxoid (England) and Sigma-Aldrich (USA).

2.3. Samples preparation for elemental analysis and antimicrobial activity

For elemental analysis, the dried powdered samples were digested using microwave digestion system and decomposition procedures (Park et al., 2018; Jamila et al., 2019) in which 0.5 g of the samples were mixed with 1.0 mL H_2O_2 (30%, v/v) and 7.0 mL HNO_3 (70%) in microwave polytetrafluoroethylene digestion vessels. The system was operated at 1000 W and different temperature as; 80 °C, 5 min; 120 °C, 5 min; 150 °C, 5 min; 180 °C, 20 min; and cooling at 40 °C. After decomposition, the combusted samples after dilution with deionized water up to 20.0 g were filtered and subjected to elemental analysis. For antimicrobial activity,

Table 3 Mean concentration (μg/g, dry weight basis) of micro elements of the analyzed herbal products available in local markets of Pakistan.

Herb	Product	Ni^{60}	Cu^{63}	Zn^{64}	Sr^{87}	Rb^{85}
A. calamus	P1	1.69 ± 0.111	1.11 ± 0.219	11.3 ± 1.95	59.3 ± 3.87	63.4 ± 4.69
	P2	2.00 ± 0.107	1.49 ± 0.404	19.1 ± 1.18	50.1 ± 4.01	68.9 ± 6.19
	P3	1.59 ± 0.405	0.915 ± 0.010	12.7 ± 1.67	61.2 ± 11.1	72.4 ± 3.07
B. edulis	P4	4.18 ± 0.311	0.518 ± 0.071	3.39 ± 0.017	2.17 ± 0.051	3.06 ± 0.215
	P5	3.78 ± 0.401	0.383 ± 0.091	4.59 ± 1.02	3.14 ± 0.072	2.27 ± 0.081
	P6	4.97 ± 0.917	0.69 ± 0.039	2.17 ± 0.729	2.73 ± 0.052	2.79 ± 0.471
C. bonducella	P7	1.60 ± 0.591	0.911 ± 0.029	9.18 ± 1.83	25.6 ± 1.21	95.8 ± 3.17
	P8	2.01 ± 0.715	0.893 ± 0.091	12.8 ± 2.30	29.3 ± 2.17	104.8 ± 9.17
	P9	2.06 ± 0.619	1.06 ± 0.431	10.4 ± 1.06	23.4 ± 1.73	110.6 ± 13.8
C. orchioides	P10	1.85 ± 0.092	0.557 ± 0.048	21.1 ± 2.79	74.3 ± 7.17	196.9 ± 11.0
	P11	1.71 ± 0.094	0.618 ± 0.098	18.2 ± 3.50	79.5 ± 5.18	173.8 ± 10.4
	P12	2.10 ± 0.097	1.00 ± 0.059	15.9 ± 1.16	80.3 ± 9.03	205.7 ± 15.6
H. isora	P13	0.454 ± 0.016	1.50 ± 0.261	1.75 ± 0.704	10.2 ± 1.00	10.1 ± 0.981
	P14	0.706 ± 0.031	1.39 ± 0.906	2.60 ± 0.946	9.44 ± 0.973	9.63 ± 1.03
	P15	0.517 ± 0.068	1.10 ± 0.291	3.38 ± 1.19	11.3 ± 2.03	11.7 ± 1.81
H. pubescens	P16	0.963 ± 0.031	1.32 ± 0.604	31.3 ± 3.16	90.3 ± 9.01	123.5 ± 9.24
	P17	1.18 ± 0.059	1.49 ± 0.439	26.4 ± 2.01	101.1 ± 10.1	134.6 ± 8.24
	P18	0.946 ± 0.051	1.68 ± 0.379	34.3 ± 4.15	87.1 ± 7.18	157.9 ± 14.0
P. integerrima	P19	2.09 ± 0.712	0.330 ± 0.005	2.50 ± 0.701	130.6 ± 8.83	169.9 ± 12.1
	P20	2.01 ± 0.503	0.519 ± 0.073	2.18 ± 0.513	159.9 ± 10.8	193.7 ± 14.9
	P21	1.87 ± 0.701	0.364 ± 0.014	3.11 ± 0.241	129.0 ± 6.78	207.7 ± 10.7
P. sativa	P22	4.03 ± 1.01	2.21 ± 0.037	4.57 ± 0.735	194.6 ± 14.7	11.5 ± 1.17
	P23	4.72 ± 0.872	2.27 ± 0.031	4.44 ± 0.719	117.9 ± 10.0	11.0 ± 0.741
	P24	4.43 ± 0.819	1.92 ± 0.043	4.92 ± 0.536	160.7 ± 13.8	9.47 ± 1.10
Q. infectoria	P25	2.61 ± 0.771	0.536 ± 0.091	0.971 ± 0.517	52.8 ± 7.93	117.8 ± 14.0
	P26	3.01 ± 0.913	0.626 ± 0.017	0.986 ± 0.083	60.1 ± 6.09	126.1 ± 10.1
	P27	2.89 ± 0.952	0.654 ± 0.080	1.36 ± 0.075	73.5 ± 9.15	109.6 ± 9.93
R. serpentina	P28	1.14 ± 0.089	0.357 ± 0.017	29.3 ± 4.10	103.6 ± 9.00	9.94 ± 1.06
	P29	1.25 ± 0.348	0.328 ± 0.041	31.6 ± 2.27	97.6 ± 10.2	12.6 ± 1.59
	P30	0.998 ± 0.063	0.295 ± 0.058	27.8 ± 2.96	113.1 ± 15.9	14.8 ± 1.51
S. lappa	P31	0.108 ± 0.021	0.009 ± 0.001	7.89 ± 1.01	180.1 ± 20.6	71.4 ± 7.91
	P32	0.095 ± 0.009	0.016 ± 0.003	6.94 ± 1.00	203.7 ± 17.1	87.1 ± 11.0
	P33	0.122 ± 0.030	0.019 ± 0.002	8.73 ± 0.994	317.9 ± 19.2	76.2 ± 7.38
T. stocksianum	P34	5.21 ± 1.26	1.01 ± 0.003	1.59 ± 0.310	103.2 ± 9.03	56.7 ± 12.3
	P35	4.57 ± 0.991	1.43 ± 0.053	1.48 ± 0.073	129.8 ± 11.0	49.5 ± 4.67
	P36	4.25 ± 1.02	0.937 ± 0.051	1.77 ± 0.094	119.3 ± 9.09	55.4 ± 5.19
X. strumarium	P37	1.13 ± 0.039	5.00 ± 0.901	19.9 ± 1.37	100.5 ± 7.11	101.7 ± 9.51
	P38	0.986 ± 0.051	4.63 ± 0.994	25.8 ± 1.04	148.0 ± 12.4	92.7 ± 9.90
	P39	0.902 ± 0.039	5.31 ± 1.05	20.9 ± 2.03	126.3 ± 11.2	87.2 ± 5.08

the ethanolic extracts of the subject samples were prepared by extracting dried grinded samples (10 g) in Soxhlet extractor with ethanol (100 mL) and evaporation through rotary evaporator.

2.4. Elemental analysis and validation of ICP-OES and ICP-MS techniques

Macronutrients; Al, Ca, Fe, K, Mg, Na, P and S were analyzed by ICP-OES whereas micro (Cu, Ni, Rb, Sr, Zn), trace essential (Co, Cr, Se, V), trace non-toxic (Li, Ga) and trace toxic (As, Ba, Be, Cd, Cs, In, Os, Pb, Tl, U) elements were determined by ICP-MS technique. The methods were validated through linearity, limits of detection and quantification (LOD, LOQ), precision, accuracy, and analysis of certified reference material (Park et al., 2018; Jamila et al., 2019). The LOD and LOQ were calculated as three and ten times standard deviations (3xSD and 10xSD) from ten replicates of blank per slope of the calibration curve, respectively.

2.5. Antimicrobial activity assessments of herbs and their products

Antimicrobial activity was determined using disc diffusion (DD) and micro-dilution (MD) methods against *S. aureus*, *B. subtilis*, *P. aeruginosa*, *E, coli*, *C, albicans*, *C. krusei*, *A. flavus*, and *T. mentagrophytes* using Muller-Hinton agar (MHA) and broth (MHB) micro-dilution methods (Jamila et al., 2020). In DD assay, an inoculum of 100 µL was streaked on the Mueller–Hinton agar surface using a sterile cotton swab. Then, sterile paper disc impregnated with 20 µL of 2000 µg/mL (2 mg extract per 1 mL of ethanol) extracts of each herb and product sample, and the standards including streptomycin, vancomycin, fluconazole, and amphotericin were kept on inoculated agar. The samples were incubated at 37 °C overnight, and after that, the diameters of inhibition zone (mm) were measured. In MD assay, a concentration range of 1000–31.2 5 µg/mL using sterile flat-bottom 96-well plate was performed. The bacterial strains were *Bacillus subtilis*, *Staphylococcus aur-*

Table 4 Mean concentration (µg/g, dry weight basis) of trace essential elements of the analyzed herbal products available in local markets of Pakistan.

Herb	Product	V^{51}	Cr^{52}	Cr^{53}	Co^{59}	Se^{82}
A. calamus	P1	0.882 ± 0.116	1.17 ± 0.092	0.991 ± 0.011	0.658 ± 0.096	0.028 ± 0.006
	P2	0.952 ± 0.081	1.19 ± 0.061	1.020 ± 0.089	0.662 ± 0.054	0.019 ± 0.005
	P3	1.06 ± 0.045	1.57 ± 0.369	0.967 ± 0.027	0.599 ± 0.046	0.022 ± 0.006
B. edulis	P4	0.108 ± 0.017	0.291 ± 0.501	0.024 ± 0.006	0.012 ± 0.004	0.002 ± 0.0003
	P5	0.119 ± 0.027	0.310 ± 0.059	0.245 ± 0.004	0.017 ± 0.005	0.001 ± 0.0002
	P6	0.959 ± 0.045	0.295 ± 0.061	0.211 ± 0.003	0.019 ± 0.006	0.001 ± 0.0001
C. bonducella	P7	1.07 ± 0.034	3.35 ± 0.039	2.01 ± 0.092	0.399 ± 0.098	0.049 ± 0.007
	P8	1.01 ± 0.085	3.11 ± 0.062	1.93 ± 0.219	0.364 ± 0.012	0.051 ± 0.006
	P9	1.12 ± 0.117	2.69 ± 0.075	2.04 ± 0.322	0.410 ± 0.045	0.041 ± 0.003
C. orchioides	P10	4.43 ± 0.749	6.852 ± 0.812	7.75 ± 1.00	0.312 ± 0.008	1.04 ± 0.009
	P11	4.347 ± 0.618	7.27 ± 0.948	6.91 ± 0.908	0.336 ± 0.046	1.08 ± 0.009
	P12	3.87 ± 0.459	9.13 ± 1.09	7.14 ± 0.993	0.369 ± 0.092	1.12 ± 0.008
H. isora	P13	0.073 ± 0.001	0.057 ± 0.007	0.029 ± 0.009	0.004 ± 0.001	0.003 ± 0.0002
	P14	0.021 ± 0.004	0.046 ± 0.002	0.020 ± 0.008	0.005 ± 0.001	0.002 ± 0.0003
	P15	0.024 ± 0.004	0.039 ± 0.004	0.024 ± 0.004	0.006 ± 0.002	0.001 ± 0.0001
H. pubescens	P16	2.18 ± 0.761	82.2 ± 4.51	20.05 ± 1.06	0.427 ± 0.069	0.051 ± 0.011
	P17	2.21 ± 0.839	76.9 ± 3.93	18.6 ± 1.10	0.367 ± 0.057	0.049 ± 0.010
	P18	1.95 ± 0.617	81.4 ± 6.31	19.0 ± 1.03	0.479 ± 0.061	0.047 ± 0.009
P. integerrima	P19	0.172 ± 0.031	0.700 ± 0.039	0.514 ± 0.005	0.019 ± 0.189	0.014 ± 0.006
	P20	0.131 ± 0.007	0.753 ± 0.028	0.465 ± 0.039	0.024 ± 0.005	0.009 ± 0.0008
	P21	0.127 ± 0.007	0.631 ± 0.044	0.531 ± 0.089	0.019 ± 0.005	0.009 ± 0.003
P. sativa	P22	2.18 ± 0.743	4.471 ± 0.615	5.06 ± 0.992	0.496 ± 0.006	0.170 ± 0.006
	P23	2.37 ± 0.962	5.49 ± 0.918	4.85 ± 0.836	0.411 ± 0.038	0.182 ± 0.009
	P24	3.17 ± 0.949	6.01 ± 0.913	4.14 ± 0.937	0.392 ± 0.096	0.146 ± 0.007
Q. infectoria	P25	0.217 ± 0.017	0.473 ± 0.009	0.372 ± 0.011	0.084 ± 0.013	0.058 ± 0.006
	P26	0.259 ± 0.089	0.398 ± 0.051	0.420 ± 0.087	0.079 ± 0.018	0.048 ± 0.007
	P27	0.214 ± 0.045	0.513 ± 0.041	0.464 ± 0.054	0.076 ± 0.009	0.046 ± 0.009
R. serpentina	P28	2.01 ± 0.094	29.5 ± 2.38	9.05 ± 1.03	0.502 ± 0.069	0.179 ± 0.017
	P29	2.21 ± 0.072	23.6 ± 1.96	7.94 ± 0.910	0.567 ± 0.009	0.164 ± 0.030
	P30	2.59 ± 0.419	27.1 ± 1.14	8.01 ± 0.995	0.479 ± 0.039	0.170 ± 0.089
S. lappa	P31	0.792 ± 0.134	1.56 ± 0.077	1.01 ± 0.042	0.119 ± 0.018	0.082 ± 0.006
	P32	0.801 ± 0.089	2.03 ± 0.600	0.930 ± 0.092	0.114 ± 0.011	0.075 ± 0.009
	P33	0.726 ± 0.074	2.11 ± 0.716	1.19 ± 0.752	0.120 ± 0.009	0.074 ± 0.011
T. stocksianum	P34	6.11 ± 1.00	6.44 ± 1.15	6.38 ± 0.905	0.712 ± 0.911	0.740 ± 0.006
	P35	5.87 ± 0.762	7.26 ± 1.09	5.84 ± 0.939	0.701 ± 0.717	0.789 ± 0.009
	P36	5.07 ± 0.718	8.16 ± 1.05	5.14 ± 0.973	0.699 ± 0.428	0.764 ± 0.007
X. strumarium	P37	1.79 ± 0.471	3.75 ± 1.17	3.07 ± 0.719	0.841 ± 0.011	0.015 ± 0.006
	P38	2.28 ± 0.051	2.97 ± 0.961	2.78 ± 0.908	0.769 ± 0.095	0.016 ± 0.005
	P39	2.41 ± 0.089	1.99 ± 0.847	3.41 ± 0.991	0.696 ± 0.066	0.011 ± 0.004

eus, *Escherichia coli* and *Pseudomonas aeruginosa* whereas yeasts included *Candida albicans*, *Candida krusei*, *Aspergillus flavus*, and *Trichophyton mentagrophytes*.

2.6. Statistical analysis

The obtained results are reported as means ± standard deviations ($n = 3$). The mean significant differences (represented by superscript letters) between the obtained values were analyzed using ANOVA along with Tukey's HSD test in SPSS, version 20.0 (SPSS Inc., Chicago, USA). Data are expressed with a significance level of $p < 0.05$.

3. Results and discussion

3.1. Validation of ICP-OES and ICP-MS techniques

The validation results obtained are given in Table S2, supplementary material. The values of relative standard deviation (% RSD) for precision were below 3%, and spike recovery (%) obtained was ranging from 94 to 104%. The recoveries of reference material; NIST SRM-1573a, tomato leaves analysis for accuracy are given in Table S3, supplementary material. In the inter laboratories calibrations by FAPAS, the results of the proficiency test were successfully achieved within 0.5 Z-score. Hence, the applied techniques for the elemental analysis of the subject herbs fulfill the required standards of Association of Official Analytical Chemists (AOAC, 2012).

3.2. Elemental content analysis of herbs and their products

A large segment of the developing countries still relies on the traditional medicine due to their efficacy and accessibility. However, contamination of medicinal herbs and their products with heavy metals, which ultimately affect their safety and quality, is one of the most pressing threats to human health as well as the pharmaceutical industries (Asgari et al., 2017). The results (mean ± standard deviation, μg/g) of macro, micro and essential trace nutrients are enlisted as Tables 1–6. From the result of the elemental analysis, it was found that the content of the macronutrients are in the order of: K > P > Ca > Na > S > Mg > Fe > Al (*A. calamus*), K > Ca > P > Mg > Al > Fe > S > Na (*B. edulis*), K > P > Ca > Mg > S > Fe > Al > Na (*C. bonducella*), Ca > K > Al > Mg > P > Na > S > Fe (*C. orchioides*), K > Ca > Mg > P > S > Na > Fe > Al (*H. isora*), K > Ca > P > Mg > S > Fe > Al > Na (*H. pubescens*), K > P > Ca > Mg > S > Al > Na > Fe (*P. integerrima*), K > Ca > Mg > P > Fe > Al > S > Na (*P. sativa*), K > Ca > P > Mg > S > Na > Al > Fe (*Q. infectoria*), K > Ca > P > Mg > Al > Fe > S > Na (*R. serpentina*), Ca > K > Mg > S > P > Fe > Al > Na (*S. lappa*), K > Ca > Mg > Fe > Al > P > Na > S (*T. stocksianum*), and K > Ca > Mg > Al > Fe > P > S > Na (*X. strumarium*). All the herbs except *C. orchioides* and *S. lappa* showed the highest content of K followed by Ca. Overall, the analyzed herbs were preferably the rich sources of K, Ca, P, and Mg. An inconsistency and variations were found in the content of macronutrients, when compared to the literature, which might be attributed to geographical variations. For example, in *Q. infectoria* of Turkish origin, the content of K and Fe analyzed by atomic absorption spectroscopy, were present as 8326 μg/g and 19 μg/g, respectively. In *A. calamus*, the content of Ca, K, Al, and Fe were 2658 μg/g, 11447 μg/g, 979 μg/g, and 707 μg/g, respectively (Özcan and Bayçu, 2005; Özcan and Akbulut, 2008). Regarding the macro elements content, on the whole, Ca, K, Mg and Na were the dominating macronutrients in almost all the products whereas S and P were present at lower concentrations (Table 2).

Most herbs contain several micro and essential elements such as Co, Cr, Cu, Ni, Rb, Se, V, Sr, and Zn in therapeutic concentrations. However, the low and excessive daily intake could lead to deficiency or overload and ultimately severe consequences. From the results (Table 1), it was observed that among micronutrients, Rb^{85} (1.45, *B. edulis* to 284 μg/g, *C. orchioides*) and Sr^{87} (0.649, *B. edulis* to 417 μg/g, *S. lappa*) had the higher content in all the analyzed herbs. The amount of Zn, Cu, and Ni was found to be in similar increasing order

Table 5 Mean concentration (μg/g, dry weight basis) of trace non-toxic elements of the analyzed herbal products available in local markets of Pakistan.

Herb	Product	Li^7	Ga^{69}
A. calamus	P1	0.186 ± 0.009	0.614 ± 0.009
	P2	0.167 ± 0.099	0.662 ± 0.073
	P3	0.177 ± 0.083	0.602 ± 0.015
B. edulis	P4	0.058 ± 0.017	0.014 ± 0.005
	P5	0.048 ± 0.012	0.011 ± 0.004
	P6	0.052 ± 0.015	0.011 ± 0.003
C. bonducella	P7	0.177 ± 0.031	0.101 ± 0.042
	P8	0.169 ± 0.048	0.126 ± 0.047
	P9	0.182 ± 0.061	0.104 ± 0.011
C. orchioides	P10	2.91 ± 0.815	1.68 ± 0.130
	P11	3.30 ± 0.727	1.83 ± 0.471
	P12	3.47 ± 0.618	2.14 ± 0.536
H. isora	P13	0.003 ± 0.001	0.006 ± 0.001
	P14	0.005 ± 0.001	0.004 ± 0.001
	P15	0.004 ± 0.001	0.007 ± 0.002
H. pubescens	P16	0.718 ± 0.061	0.602 ± 0.071
	P17	0.721 ± 0.093	0.763 ± 0.191
	P18	0.698 ± 0.097	0.700 ± 0.041
P. integerrima	P19	0.072 ± 0.022	0.114 ± 0.007
	P20	0.069 ± 0.007	0.127 ± 0.009
	P21	0.072 ± 0.007	0.119 ± 0.035
P. sativa	P22	3.11 ± 0.618	1.81 ± 0.717
	P23	2.61 ± 0.467	1.89 ± 0.992
	P24	2.19 ± 0.816	1.94 ± 0.990
Q. infectoria	P25	4.26 ± 0.051	0.272 ± 0.015
	P26	3.99 ± 0.817	0.269 ± 0.091
	P27	3.82 ± 0.749	0.264 ± 0.040
R. serpentina	P28	2.11 ± 0.911	0.608 ± 0.073
	P29	1.93 ± 0.718	0.799 ± 0.093
	P30	2.09 ± 0.614	0.707 ± 0.071
S. lappa	P31	0.399 ± 0.091	0.801 ± 0.057
	P32	0.382 ± 0.071	0.831 ± 0.078
	P33	0.429 ± 0.080	0.819 ± 0.693
T. stocksianum	P34	7.01 ± 0.901	0.739 ± 0.080
	P35	6.19 ± 0.619	0.802 ± 0.201
	P36	5.81 ± 0.415	0.700 ± 0.064
X. strumarium	P37	1.66 ± 0.417	2.09 ± 0.667
	P38	1.69 ± 0.475	2.29 ± 0.887
	P39	1.77 ± 0.999	2.11 ± 0.718

Table 6 Mean concentration (μg/g) of toxic elements of the analyzed herbal products available in local markets of Pakistan.

Herb	Product	As^{75}	Cd^{111}	In^{115}	Cs^{133}	Ba^{138}	Be^{9}	Tl^{205}	Pb^{206}	Pb^{208}	U^{238}
A. calamus	P1	0.200 ± 0.005	0.020 ± 0.004	0.008 ± 0.0003	0.017 ± 0.002	121.5 ± 9.05	0.004 ± 0.0001	0.023 ± 0.003	2.82 ± 0.072	2.51 ± 0.042	0.0001 ± 0.00002
	P2	0.199 ± 0.016	0.019 ± 0.005	0.008 ± 0.0003	0.015 ± 0.004	148.3 ± 8.10	0.004 ± 0.0001	0.021 ± 0.003	2.75 ± 0.063	2.59 ± 0.091	0.0001 ± 0.00002
	P3	0.183 ± 0.082	0.024 ± 0.008	0.006 ± 0.0004	0.013 ± 0.005	137.5 ± 8.09	0.002 ± 0.0001	0.019 ± 0.004	2.68 ± 0.075	2.48 ± 0.593	0.0001 ± 0.00001
B. edulis	P4	0.009 ± 0.003	0.0005 ± 0.003	0.0002 ± 0.00004	0.007 ± 0.0003	3.20 ± 0.808	0.0005 ± 0.00003	0.001 ± 0.0001	0.053 ± 0.006	0.059 ± 0.007	0.00002 ± 0.000005
	P5	0.008 ± 0.001	0.005 ± 0.001	0.0002 ± 0.00003	0.008 ± 0.0002	3.09 ± 0.091	0.0006 ± 0.00003	0.001 ± 0.0005	0.050 ± 0.007	0.055 ± 0.007	0.00002 ± 0.000003
	P6	0.008 ± 0.001	0.0004 ± 0.001	0.0003 ± 0.00003	0.008 ± 0.0007	3.23 ± 0.098	0.0004 ± 0.00002	0.005 ± 0.001	0.046 ± 0.007	0.054 ± 0.007	0.00002 ± 0.000003
C. bonducella	P7	0.020 ± 0.004	0.012 ± 0.006	0.004 ± 0.0003	0.0002 ± 0.00007	6.23 ± 1.01	0.001 ± 0.0002	0.006 ± 0.0003	3.74 ± 0.058	3.38 ± 0.059	0.0003 ± 0.0001
	P8	0.019 ± 0.006	0.011 ± 0.007	0.004 ± 0.0004	0.022 ± 0.008	5.71 ± 0.974	0.0009 ± 0.0002	0.006 ± 0.00002	3.06 ± 0.576	3.42 ± 0.511	0.0003 ± 0.00002
	P9	0.019 ± 0.005	0.015 ± 0.s001	0.005 ± 0.0003	0.029 ± 0.003	4.02 ± 0.902	0.001 ± 0.0002	0.005 ± 0.0001	2.52 ± 0.873	3.27 ± 0.517	0.0002 ± 0.0001
C. orchioides	P10	0.193 ± 0.083	0.028 ± 0.001	0.0003 ± 0.0001	0.453 ± 0.004	104.0 ± 10.7	0.049 ± 0.011	0.041 ± 0.016	1.17 ± 0.508	1.10 ± 0.300	0.0003 ± 0.00002
	P11	0.203 ± 0.095	0.029 ± 0.001	0.0003 ± 0.0001	0.496 ± 0.043	120.3 ± 9.08	0.058 ± 0.009	0.049 ± 0.012	0.999 ± 0.095	1.09 ± 0.038	0.0002 ± 0.00003
	P12	0.200 ± 0.046	0.025 ± 0.003	0.0003 ± 0.0001	0.513 ± 0.068	157.2 ± 10.9	0.041 ± 0.005	0.053 ± 0.009	1.01 ± 0.082	1.13 ± 0.047	0.0001 ± 0.00003
H. isora	P13	0.002 ± 0.0004	0.0008 ± 0.0003	0.018 ± 0.0002	0.003 ± 0.0002	3.01 ± 0.065	0.0001 ± 0.00004	0.0006 ± 0.0001	0.035 ± 0.002	0.039 ± 0.007	0.0004 ± 0.0001
	P14	0.001 ± 0.0004	0.0008 ± 0.0002	0.013 ± 0.004	0.005 ± 0.001	3.32 ± 0.904	0.0007 ± 0.00001	0.0007 ± 0.0001	0.039 ± 0.003	0.043 ± 0.006	0.0005 ± 0.00009
	P15	0.002 ± 0.0005	0.0009 ± 0.0002	0.014 ± 0.004	0.003 ± 0.0005	3.51 ± 0.095	0.0005 ± 0.0001	0.0007 ± 0.0001	0.027 ± 0.002	0.036 ± 0.006	0.0003 ± 0.00001
H. pubescens	P16	0.085 ± 0.004	0.0205 ± 0.003	0.013 ± 0.001	0.164 ± 0.057	217.2 ± 11.8	0.019 ± 0.009	0.007 ± 0.0001	0.617 ± 0.076	0.626 ± 0.085	0.0003 ± 0.00002
	P17	0.099 ± 0.006	0.019 ± 0.001	0.010 ± 0.002	0.183 ± 0.092	194.9 ± 10.3	0.023 ± 0.007	0.008 ± 0.0005	0.559 ± 0.007	0.619 ± 0.068	0.0002 ± 0.00001
	P18	0.088 ± 0.007	0.0 ± 0.001	0.022 ± 0.003	0.173 ± 0.078	199.2 ± 9.19	0.021 ± 0.007	0.008 ± 0.0003	0.643 ± 0.005	0.661 ± 0.069	0.0003 ± 0.00001
P. integerrima	P19	0.185 ± 0.005	0.005 ± 0.0006	0.002 ± 0.0004	0.326 ± 0.027	4.23 ± 0.777	0.00015 ± 0.00009	0.008 ± 0.0003	0.385 ± 0.049	0.328 ± 0.027	0.0004 ± 0.00001
	P20	0.191 ± 0.067	0.006 ± 0.0003	0.003 ± 0.0004	0.311 ± 0.017	4.16 ± 0.099	0.0003 ± 0.00002	0.008 ± 0.0001	0.318 ± 0.053	0.337 ± 0.032	0.0003 ± 0.00001
	P21	0.176 ± 0.054	0.005 ± 0.00001	0.003 ± 0.0004	0.349 ± 0.013	4.05 ± 0.914	0.0001 ± 0.00004	0.006 ± 0.0001	0.399 ± 0.094	0.348 ± 0.037	0.0002 ± 0.0001
P. sativa	P22	0.231 ± 0.087	0.024 ± 0.001	0.061 ± 0.002	0.279 ± 0.046	123.4 ± 8.91	0.011 ± 0.003	0.012 ± 0.005	0.993 ± 0.098	0.817 ± 0.059	0.0003 ± 0.000001
	P23	0.215 ± 0.096	0.026 ± 0.001	0.074 ± 0.007	0.294 ± 0.053	115.4 ± 7.92	0.009 ± 0.002	0.011 ± 0.002	1.00 ± 0.175	1.37 ± 0.149	0.0002 ± 0.00001
	P24	0.207 ± 0.009	0.025 ± 0.003	0.078 ± 0.003	0.271 ± 0.071	121.6 ± 7.19	0.009 ± 0.003	0.013 ± 0.002	1.17 ± 0.047	1.21 ± 0.038	0.0001 ± 0.00001
Q. infectoria	P25	0.041 ± 0.008	0.022 ± 0.0003	0.008 ± 0.0004	0.161 ± 0.023	43.4 ± 3.24	0.011 ± 0.004	0.083 ± 0.003	0.785 ± 0.062	0.737 ± 0.039	0.0004 ± 0.00003
	P26	0.026 ± 0.009	0.022 ± 0.004	0.008 ± 0.0005	0.174 ± 0.054	39.6 ± 3.13	0.017 ± 0.003	0.089 ± 0.004	0.659 ± 0.033	0.690 ± 0.027	0.0005 ± 0.00002
	P27	0.043 ± 0.009	0.022 ± 0.007	0.007 ± 0.001	0.170 ± 0.057	47.6 ± 3.06	0.009 ± 0.002	0.080 ± 0.003	0.584 ± 0.11	0.683 ± 0.091	0.0004 ± 0.0001
R. serpentina	P28	0.177 ± 0.083	0.020 ± 0.003	0.057 ± 0.003	0.001 ± 0.0003	116.2 ± 9.10	0.009 ± 0.001	0.031 ± 0.002	0.063 ± 0.006	0.066 ± 0.016	0.0002 ± 0.0001
	P29	0.170 ± 0.057	0.009 ± 0.001	0.052 ± 0.003	0.001 ± 0.0002	127.8 ± 7.71	0.010 ± 0.001	0.029 ± 0.003	0.059 ± 0.007	0.069 ± 0.012	0.0002 ± 0.00001
	P30	0.188 ± 0.074	0.024 ± 0.001	0.053 ± 0.006	0.0003 ± 0.00007	139.5 ± 7.93	0.011 ± 0.001	0.020 ± 0.003	0.057 ± 0.004	0.077 ± 0.013	0.0002 ± 0.00003
S. lappa	P31	0.060 ± 0.016	0.022 ± 0.006	0.003 ± 0.004	0.072 ± 0.007	178.2 ± 11.7	0.012 ± 0.004	0.051 ± 0.009	1.04 ± 0.004	1.17 ± 0.281	0.0004 ± 0.00001
	P32	0.067 ± 0.011	0.011 ± 0.007	0.005 ± 0.0003	0.089 ± 0.008	201.0 ± 10.4	0.016 ± 0.004	0.049 ± 0.006	1.59 ± 0.005	1.48 ± 0.351	0.0003 ± 0.00002
	P33	0.061 ± 0.009	0.0005 ± 0.00001	0.002 ± 0.0005	0.091 ± 0.003	198.3 ± 10.8	0.014 ± 0.005	0.055 ± 0.005	1.78 ± 0.009	1.71 ± 0.082	0.0004 ± 0.00001
T. stocksianum	P34	0.785 ± 0.098	0.0004 ± 0.0001	0.531 ± 0.132	0.009 ± 0.0004	217.6 ± 7.81	0.031 ± 0.007	0.021 ± 0.005	3.31 ± 0.726	3.27 ± 0.648	0.0001 ± 0.00002
	P35	0.814 ± 0.095	0.0006 ± 0.00001	0.552 ± 0.094	0.009 ± 0.0003	273.6 ± 9.10	0.039 ± 0.009	0.019 ± 0.002	3.00 ± 0.575	3.16 ± 0.514	0.0001 ± 0.00001
	P36	0.807 ± 0.074	0.005 ± 0.0003	0.494 ± 0.093	0.008 ± 0.0006	199.7 ± 10.9	0.029 ± 0.005	0.030 ± 0.005	3.58 ± 0.602	3.16 ± 0.591	0.0001 ± 0.00002
X. strumarium	P37	0.148 ± 0.051	0.049 ± 0.003	0.013 ± 0.003	0.221 ± 0.092	316.8 ± 13.7	0.019 ± 0.007	0.023 ± 0.003	1.65 ± 0.715	1.69 ± 0.614	0.0002 ± 0.00001
	P38	0.136 ± 0.033	0.055 ± 0.003	0.018 ± 0.003	0.271 ± 0.004	328.3 ± 14.3	0.009 ± 0.001	0.021 ± 0.003	1.78 ± 0.816	1.71 ± 0.375	0.0002 ± 0.00001
	P39	0.155 ± 0.087	0.052 ± 0.003	0.011 ± 0.003	0.287 ± 0.056	298.5 ± 11.4	0.013 ± 0.002	0.018 ± 0.003	1.59 ± 0.781	1.53 ± 0.418	0.0002 ± 0.00001

after Rb^{85} and $Sr^{87}$and trend as Rb^{85}/ Sr^{87} > Zn^{64}/Cu^{63} > Ni^{60}. In the analyzed products, in micro elements content, the average levels were in subsequent order of Rb^{85} > Sr^{87} > Zn > Ni > Cu (Table 3). Furthermore, the measured levels of micro elements were well below the permissible limits, hence, the subject herbs can be considered safe for consumers' health.

Other elements; Co, Cr, Mn, Se, and V are essential trace elements, which have key role as cofactors in metabolic processes. Among these elements, all the herbs and products were rich in Cr^{52}/Cr^{53} followed by Co^{59}, V^{51}, and Se^{82} (Tables 1 and 4). Hence, these herbs contain appreciable macro and microelements to add on to the nutritional requirements as supplementary food. Some other elements such as Li^{7} and Ga^{69} present in the soil are also absorbed by plants and are non-toxic. Among the samples analyzed, the content of Li^{7} and Ga^{69} were found high (Tables 1 and 5).

Table 7 Antimicrobial activity by disc diffusion (DD) and minimum inhibitory concentration (MIC) methods of the ethanolic extract of selected herbs.

Samples	Disk diffusion (mm)				Minimum inhibitory concentration method (μg/mL)			
	S. aureus	*B. subtilis*	*P. aeruginosa*	*E. coli*	*S. aureus*	*B. subtilis*	*P. aeruginosa*	*E. coli*
Acorus calamus	9.0 ± 0.45^{a}	11.0 ± 0.52^{c}	7.0 ± 0.35^{a}	6.0 ± 0.10^{a}	500^{e}	500^{e}	1000^{c}	1000^{c}
Blepharis edulis	$11.0^{c} \pm 0.26^{c}$	13.0 ± 0.42^{e}	9.0 ± 0.32^{c}	8.0 ± 0.41^{c}	250^{d}	250^{d}	500^{b}	500^{b}
Caesalpinia bonducella	10.0 ± 0.22^{b}	11.0 ± 0.24^{c}	9.0 ± 0.30^{c}	9.0 ± 0.38^{d}	250^{d}	250^{d}	500^{b}	500^{b}
Curculigo orchioides	13.0 ± 0.62^{e}	12.0 ± 0.42^{d}	8.0 ± 0.28^{b}	8.0 ± 0.27^{c}	250^{d}	250^{d}	500^{b}	500^{b}
Helicteres isora	14.0 ± 0.51^{f}	15.0 ± 0.40^{g}	8.0 ± 0.33^{b}	6.0 ± 0.20^{a}	125^{c}	62.5^{b}	500^{b}	1000^{c}
Holarrhena pubescens	11.0 ± 0.62^{c}	11.0 ± 0.38^{c}	10.0 ± 0.41^{d}	8.0 ± 0.18^{c}	250^{d}	250^{d}	500^{b}	500^{b}
Pastinaca sativa	13.0 ± 0.10^{e}	10.0 ± 0.21^{b}	8.0 ± 0.32^{b}	7.0 ± 0.14^{b}	250^{d}	250^{d}	500^{b}	1000^{c}
Pistacia integerrima	14.0 ± 0.36^{f}	12.0 ± 0.44^{d}	9.0 ± 0.40^{c}	9.0 ± 0.52^{d}	125^{c}	250^{d}	500^{b}	500^{b}
Quercus infectoria	12.0 ± 0.61^{d}	14.0 ± 0.35^{f}	7.0 ± 0.09^{a}	6.0 ± 0.06^{a}	250^{d}	125^{c}	1000^{c}	1000^{c}
Rauwolfia serpentina	10.0 ± 0.11^{b}	9.0 ± 0.39^{a}	8.0 ± 0.51^{b}	7.0 ± 0.18^{b}	250^{d}	250^{d}	500^{b}	1000^{c}
Saussurea lappa	16.0 ± 0.47^{g}	17.0 ± 0.22^{h}	10.0 ± 0.16^{d}	10.0 ± 0.33^{e}	62.5^{b}	62.5^{b}	250^{a}	250^{a}
Teucrium stocksianum	10.0 ± 0.10^{b}	12.0 ± 0.23^{d}	7.0 ± 0.15^{a}	7.0 ± 0.26^{b}	250^{d}	250^{d}	1000^{c}	1000^{c}
Xanthium strumarium	12.0 ± 0.21^{d}	10.0 ± 0.17^{b}	10.0 ± 0.22^{d}	10.0 ± 0.33^{e}	250^{d}	250^{d}	250^{a}	250^{a}
Vancomycin*	19.0 ± 0.28^{i}	27.0 ± 0.31^{i}	13.0 ± 0.10^{e}	13.0 ± 0.15^{f}	31.25^{a}	31.25^{a}	500^{b}	250^{a}
Streptomycin*	17.0 ± 0.20^{h}	27.0 ± 0.35^{i}	16.0 ± 0.16^{f}	17.0 ± 0.19^{g}	31.25^{a}	31.25^{a}	500^{b}	500^{b}
Antifungal activity								
Samples	Disk diffusion (mm)				Minimum inhibitory concentration method (μg/mL)			
	C. albicans	*C. krusei*	*A. flavus*	*T. mentagrophytes*	*C. albicans*	*C. krusei*	*A. flavus*	*T. mentagrophyte*
Acorus calamus	11.0 ± 0.32^{d}	8.0 ± 0.21^{c}	10.0 ± 0.23^{d}	8.0 ± 0.42^{b}	250^{d}	1000^{f}	500^{c}	1000^{e}
Blepharis edulis	12.0 ± 0.14^{e}	14.0 ± 0.10^{f}	12.0 ± 0.13^{e}	15.0 ± 0.23^{g}	250^{d}	250^{d}	62.5^{a}	250^{c}
Caesalpinia bonducella	10.0 ± 0.17^{c}	9.0 ± 0.27^{d}	9.0 ± 0.25^{c}	10.0 ± 0.26^{d}	250^{d}	500^{e}	250^{b}	250^{c}
Curculigo orchioides	14.0 ± 0.31^{f}	12.0 ± 0.42^{e}	13.0 ± 0.33^{f}	13.0 ± 0.23^{f}	125^{c}	250	500^{c}	500^{d}
Helicteres isora	14.0 ± 0.11^{f}	16.0 ± 0.37^{g}	12.0 ± 0.62^{e}	9.0 ± 0.10^{c}	125^{c}	125^{c}	250^{b}	250^{c}
Holarrhena pubescens	9.0 ± 0.10^{b}	8.0 ± 0.24^{c}	10.0 ± 0.22^{d}	8.0 ± 0.31^{b}	500^{e}	250^{d}	250^{b}	500^{d}
Pastinaca sativa	9.0 ± 0.27^{b}	7.0 ± 0.16^{b}	8.0 ± 0.33^{b}	10.0 ± 0.21^{d}	500^{e}	500^{e}	500^{c}	250^{c}
Pistacia integerrima	9.0 ± 0.13^{b}	9.0 ± 0.34^{b}	7.0 ± 0.20^{a}	7.0 ± 0.16^{a}	500^{e}	500^{e}	500^{c}	500^{d}
Quercus infectoria	11.0 ± 0.41^{d}	12.0 ± 0.55^{e}	12.0 ± 0.25^{e}	9.0 ± 0.13^{c}	250^{d}	250^{d}	250^{b}	250^{c}
Rauwolfia serpentina	8.0 ± 0.20^{a}	7.0 ± 0.19^{b}	8.0 ± 0.38^{b}	8.0 ± 0.32^{b}	500^{e}	500^{e}	500^{c}	500^{d}
Saussurea lappa	16.0 ± 0.20^{g}	16.0 ± 0.29^{g}	17.0 ± 0.34^{g}	12.0 ± 0.17^{e}	125^{c}	125^{c}	62.5^{a}	250^{c}
Teucrium stocksianum	8.0 ± 0.17^{a}	6.0 ± 0.24^{a}	7.0 ± 0.36^{a}	8.0 ± 0.18^{b}	1000^{f}	1000^{f}	1000	1000^{e}
Xanthium strumarium	17.0 ± 0.26^{h}	17.0 ± 0.16^{h}	18.0 ± 0.23^{h}	19.0 ± 0.19^{h}	62.5^{b}	62.5^{b}	62.5^{a}	62.5^{b}
Fluconazole*	22.0 ± 0.31^{i}	24.0 ± 0.19^{j}	27.0 ± 0.35^{j}	29.0 ± 0.31^{j}	31.25^{a}	31.25^{a}	62.5^{a}	31.25^{a}
Amphotericin*	22.0 ± 0.24^{i}	21.0 ± 0.26^{i}	20.0 ± 0.38^{i}	24.0 ± 0.41^{i}	62.5^{b}	62.5^{b}	62.5^{a}	31.25^{a}

Values are mean ± standard deviations of three ($n = 3$) measurements.
The superscript letters in columns represent significantly different values ($p < 0.05$) by Tukey's and Duncan's multiple range tests.
* Represents standard drugs.

In developing countries, mostly the herbs and the products are directly sold in the market without their safety analysis, which may lead to lethality due to the presence of mycotoxins, pesticides or the presence of some elements such as arsenic, cadmium, lead, and mercury, which are harmful to the human body even when present in very small concentration. Therefore, from therapeutical and safety aspects, it is important to evaluate any herbs or products for toxic elements content. In this study, among the trace toxic elements (Tables 1 and 6), Ba^{138} and Pb^{206}/Pb^{208} were present in high levels. It is worth highlighting that the toxic elements content in the analyzed samples, were all well below the safety limits (50 μg/kg/day for As, 0.833 μg/kg/day for Cd, 0.63 μg/kg/day BMDL for adult for Pb) set by Joint Food and Agriculture Organization/World Health Organization Expert Committee on Food Additives, and European Food Safety Authority (JECFA, 2004, 2010; EFSA, 2010). On the basis of obtained trace and toxic elements data for the analyzed samples, the consumption of the analyzed herbs and their products apparently may not have health risks to the consumers.

3.3. Antimicrobial activity of the herbs and products

Antimicrobial activity of the ethanolic extracts was evaluated against selected microbial pathogens, and the zones of inhibition were compared with that of the standard antibiotics. The bacterial pathogens included *S. aureus*, *B. subtilis*, *P. aeruginosa*, *E. coli*, and the pathogenic fungi were; *C. albicans*, *C. krusei*, *A. flavus*, and *T. mentagrophytes*. Most of the herbal extracts significantly inhibited the analyzed microbes (Tables 7 and 8). In DD assay, the zones of inhibition ranged from 7.0 mm to 19.0 mm and MIC values of 62.5 to 1000 μg/mL. Among the analyzed samples, the ethanolic extracts of *S. lappa* and its products were the most active against both the Gram-positive and negative bacterial strains with inhibition zones 10.0–17.0 mm (herbs) and 9.0 to 18.0 mm (products), and MIC values of 62.5 to 250 μg/mL (herbs) and 62.5 to 500 μg/mL (products). This potent antibacterial activity could be due to the presence of bioactive constituents present in the samples. The detailed values of inhibition zone and MIC are given in Tables 7 and 8. Regarding the antifungal activity

Table 8 Antimicrobial activity by disc diffusion (DD) and minimum inhibitory concentration (MIC) methods of the ethanolic extract of selected products.

Samples	Disk diffusion (mm)				Minimum inhibitory concentration method (μg/mL)			
	S. aureus	*B. subtilis*	*P. aeruginosa*	*E. coli*	*S. aureus*	*B. subtilis*	*P. aeruginosa*	*E. coli*
P1-P3	7.0–10.0	9.0–11.0	6.0–8.0	6.0–8.0	250–500	250–500	500–1000	500–1000
P4-P6	7.0–12.0	11.0–14.0	8.0–9.0	6.0–9.0	125–250	125–250	250–500	500–1000
P7-P9	7.0–10.0	10.0–12.0	9.0–10.0	8.0–10.0	125–250[d]	250–500	250–500	500–1000
P10-P12	8.0–12.0	10.0–13.0	7.0–9.0	7.0–9.0	250–500	250–500	500–1000	500–1000
P13-P15	8.0–14.0	13.0–15.0	7.0–9.0	6.0–7.0	125–250	62.5–250	500–1000	500–1000
P16-P18	7.0–10.0	10.0–11.0	10.0–11.0	7.0–9.0	250–500	250–500	500–1000	500–1000
P19-P21	8.0–13.0	9.0–11.0	6.0–8.0	7.0–8.0	125–250	250–500	250–1000	500–1000
P22-P24	10.0–14.0	10.0–13.0	9.0–10.0	8.0–10.0	125–250	250–500	500–1000	250–500
P25-P27	10.0–13.0	11.0–15.0	7.0–9.0	6.0–7.0	250–500	125–500	500–1000	500–1000
P28-P30	7.0–10.0	8.0–10.0	8.0–9.0	6.0–8.0	250–500	250–500	500–1000	500–1000
P31-P33	12.0–16.0	14.0–18.0	10.0–12.0	9.0–11.0	62.5–250	62.5–125	125–250	125–500
P34-P36	7.0–10.0	10.0–13.0	7.0–9.0	7.0–8.0	250–500	250–500	500–1000	500–1000
P37-P39	10.0–13.0	9.0–11.0	10.0–12.0	9.0–11.0	250–500	250–500	125–500	125–250
Vancomycin*	19.0	27.0 ± 0.31	13.0 ± 0.10	13.0 ± 0.15	31.25	31.25	500	250
Streptomycin*	17.0	27.0 ± 0.35	16.0 ± 0.16	17.0 ± 0.19	31.25	31.25	500	500
Antifungal activity								
Samples	Disk diffusion (mm)				Minimum inhibitory concentration method (μg/mL)			
	C. albicans	*C. krusei*	*A. flavus*	*T. mentagrophytes*	*C. albicans*	*C. krusei*	*A. flavus*	*T. mentagrophyte*
P1-P3	10.0–12.0	7.0–8.0	10.0–11.0	6.0–8.0	250–500	1000	250–500	1000
P4-P6	19.01.0–13.0	13.0–14.0	12.0–14.0	14.0–15.0	125–250	125–250	125–250	125–250
P7-P9	9.0–10.0	8.0–10.0	8.0–9.0	9.0–10.0	250–500	500–1000	250–500	250–500
P10-P12	13.0–15.0	11.0–12.0	12.0–13.0	12.0–14.0	125–250	250–500	125–250	125–250
P13-P15	13.0–15.0	15.0–17.0	12.0–13.0	8.0–11.0	125–250	125–250	125–250	250–500
P16-P18	8.0–9.0	6.0–8.0	9.0–11.0	6.0–9.0	250–500	500–1000	250–500	500–1000
P19-P21	8.0–10.0	6.0–8.0	6.0–8.0	9.0–10.0	250–500	500–1000	500–1000	250–500
P22-P24	8.0–9.0	8.0–9.0	6.0–7.0	6.0–7.0	500–1000	500–1000	500–1000	1000
P25-P27	11.0–12.0	11.0–12.0	11.0–12.0	9.0–10.0	125–250	250–500	250–500	250–500
P28-P30	7.0–9.0	7.0–8.0	7.0–8.0	6.0–8.0	500–1000	500–1000	500–1000	500–1000
P31-P33	15.0–17.0	15.0–17.0	15.0–17.0	11.0–13.0	125–250	125–250	62.5–125	250–500
P34-P36	7.0–8.0	6.0–7.0	6.0–7.0	6.0–8.0	1000	1000	1000	1000
P37-P39	16.0–17.0	15.0–17.0	16.0–18.0	17.0–19.0	62.5–125	62.5–125	62.5–125	62.5–125
Fluconazole*	22.0 ± 0.31	24.0 ± 0.19	27.0 ± 0.35	29.0 ± 0.31	31.25	31.25	62.5	31.25
Amphotericin*	22.0 ± 0.24	21.0 ± 0.26	20.0 ± 0.38	24.0 ± 0.41	62.5	62.5	62.5	31.25

* Represents standard drugs.

described in Tables 7 and 8, *X. strumarium*, *S. lappa*, and *H. isora* and their products exhibited pronounced inhibitory potential against all fungal species.

4. Conclusions

The study investigated thirteen medicinal herbs and their products for elemental content and antimicrobial activity. The study revealed that the analyzed samples are the significant sources of K, Ca, Mg, P, S, Co, Ni, Se, V, and Zn. The content of potentially toxic elements are present below the provisional tolerable intake values. Hence, the analyzed herbs and products along with medicinal remedies could also be potential sources of mineral elements. This research further concludes that the evaluated herbs and products are potentially inhibiting the microbial pathogens. Overall, the analyzed samples are effective nutritional as well as safe source against various microbial pathogens. This is the first detailed report on the mineral and toxic elements content of the commonly used medicinal herbs and the products available in the local markets of Peshawar, Pakistan.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This research study was supported by the research grant; 8967/KPK/NRPU/R&D/HEC/2017. The authors are thankful to the Higher Education Commission (HEC) for awarding this project.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.arabjc.2021.103055.

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