

DETERMINATION OF ESSENTIAL ELEMENTS IN HERBAL EXTRACTS BY NEUTRON ACTIVATION ANALYSIS

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ABSTRACT

Different types of therapies have been introduced as an alternative treatment to various types of human disorders, among them, the use of herbal teas have been highlighted due to its low cost, easiness of acquisition and administration. The aim of this study was to evaluate the concentrations of the elements As, Ba, Br, Ca, Co, Cr, Cs, Fe, Hf, K, Na, Rb, Sb, Sc, Se, Ta, Th, U, Zn and Zr by neutron activation analysis in extracts of medicinal plants whose use is regulated by ANVISA. The relevance of this analysis is justified by the need of contributing to the recommendation of these plants as secure sources of mineral elements both for therapeutic and dietary purpose. The technique showed good sensitivity in determining the appropriate concentration of all the determined elements. Elements potentially toxic were found at concentration that do not present threats to the organism and the elements that present important roles in metabolism were determined at concentrations that can assist both therapeutic and nutritional purposes.

1. INTRODUCTION

In the present days the demand for alternative sources of unconventional treatment for individual health care and their families has been growing [1]. Among the treatment types available, therapy with medicinal plants has stood out, and among the main factors contributing to this are the capacity that some plants have in slowing down certain types of disorder [2], decreasing possible undesirable side effects, bioavailability of the Brazilian flora, facility of acquisition and low cost.

In general, medicinal plants are used preventively once they do not have immediately therapeutic action and, for the same reason, they are not recommended in medical emergencies [3, 4]. Concerning the therapies that make use of medicinal herbs with therapeutic purposes, drinking teas are the most often used, as they provide substances that moisturize, detoxify and aid in digestion [5], are ease to prepare and administrate. Worldwide, herbal products are classified as natural and, therefore, can be freely grown and marketed. This is a negative aspect due to the fact that it favors self-medication, especially in simple and ordinary diseases [6] cases. It is also important to emphasize that the user needs prior knowledge about therapeutic effects of the plants; properly use, benefits and its potential harms in case of excess of consume [7].

The plants have three main sources of elements supply. The CO₂ and water are the main source of carbon, hydrogen and oxygen, while, through the soil, they absorbed by the roots,

without much discrimination, essential (macronutrients and micronutrients), beneficial and toxic elements due their presence in soil, water or air. Another possible element source is the atmospheric dust deposited in the leaves. Knowing the concentration of these elements in the plant extracts commonly used in traditional medicine can help in verifying possible interferences in the therapeutic action of the plants or even possible toxic effects of these elements, as well as contribute to the recommendation of these plants as mineral sources in regular diet [8].

The purpose of this study was to determine the concentration of the elements As, Ba, Br, Ca, Cs, Co, Cr, Fe, Hf, Rb, Sb, Sc, Se, Ta, Th, U and Zn by neutron activation analysis in medicinal plant extracts whose use is regulated by ANVISA. The relevance of this analysis is justified by the need of contributing to the recommendation of medicinal plants as source of these minerals in the diet, as well as its compliance with ingestion limits where they exist.

2. METHODOLOGY

Samples of medicinal plants analyzed in this study were obtained from pharmacies, natural product homes and in the informal market. It was analyzed 59 species available in the market from the 66 plant species cataloged by ANVISA in the Resolution RDC No. 10 of March 9, in 2010. In Table 1 it is shown the plans selected for this study, their used part and application. After the acquisition, the samples underwent a cleaning process, with the help of white paper sheets and clamp to remove any foreign material.

The aqueous extracts were obtained as provided in ANVISA resolution for each type of plant, the processes involved were infusion, decoction and maceration. In the infusion process, approximately 150 ml of ultra pure water from a Milli Q[®] system was heated in heating plate until boiling and then the plant sample, precisely weighted according the ANVISA recommendation, was added and stood for a three minutes period. After that the sample was took out the plate, allowed to cool for a period of 30 minutes and then filtered. In the decoction processes the plant samples, precisely weighted, were added at the beginning of the procedure, the beaker containing the mixture water and sample was heated in a heating plate and after boiling the sample stood for a three minutes period. After that the sample was took out the plate, allowed to cool for a period of 30 minutes and then filtered. In the maceration processes the sample was precisely weighted and putted to soak in boiling water in a capped beaker, stood for a period of 15 minutes and after that, filtered.

Once the aqueous extract was obtained, it was taken to the heating plate and evaporated almost to dryness. The residue was treated with 5 mL of concentrated nitric acid and 2 mL of hydrogen peroxide for complete dissolution of organic matter. The final solution was pipetted onto a filter paper sheet. To ensure quantitative transfer of the solution, after the transference of the sample, the beaker was rinsed with ultra-pure water twice. The rinsing water was also evaporated and transferred to the paper sheet. The filter paper sheet, after drying, was packed in polyethylene bags to be analyzed.

The element concentrations in the extracts were obtained by instrumental neutron activation analysis (INAA), a multi-elemental technique. This technique involves the bombardment of a material followed by the induced radioactivity measurement. In general, the irradiation is

performed with thermal neutrons and the resulting radioactivity is measured using gamma ray spectrometry (9).

Table 1: Plants analyzed in this study: scientific name, used part of the plant to extract preparation and medicinal indication according to ANVISA.

Scientific name	Used part	Indication
<i>Achillea millefolium</i>	Shoots	Lack of appetite, fever, inflammation and cramping
<i>Achyrocline satureioides</i>	Inflorescence	Poor digestion and intestinal cramps, mild sedative, and anti-inflammatory
<i>Aesculus hippocastanum</i>	Seeds in shell	Capillary fragility, venous insufficiency (varicose veins and hemorrhoids)
<i>Ageratum conyzoides</i>	Shoots without flowers	Joint pain (arthritis, arthrosis) and rheumatism
<i>Allium sativum</i>	Bulb	High cholesterol, as expectorant
<i>Anacardium occidentale</i>	Under bark	Noninfectious diarrhea
<i>Arctium lappa</i>	Roots	Dyspepsia, diuretic and anti-inflammatory such as the joint pain
<i>Arnica montana</i>	flowers	Trauma, bruises, sprains, swelling due to fractures and sprains
<i>Baccharis trimera</i>	Shoots	Dyspepsia
<i>Bidens pilosa</i>	Leaves	Jaundice
<i>Calendula officinalis</i>	Flowers	Inflammations and injuries, bruises and burns
<i>Caesalpinia ferrea</i>	Beans	Injuries as hemostatic astringent and antiseptic healing
<i>Casearia sylvestris</i>	Leaves	Pain and injuries, as an antiseptic and healing topic
<i>Cinnamomum verum</i>	Bark	Lack of appetite, mild cramping, flatulence and feeling of fullness
<i>Citrus aurantium</i>	Flowers	Mild cases of anxiety and insomnia, sedative
<i>Cordia verbenacea</i>	Leaves	Inflammation in bruises and pain
<i>Urcuma longa</i>	Rhizomes	Dyspepsia, Anti-inflammatory
<i>Cymbopogon citratus</i>	Leaves	Intestinal and uterine cramping, mild anxiety cases, insomnia, sedative
<i>Echinodorus macrophyllus</i>	Leaves	Edema by fluid retention and inflammation
<i>Equisetum arvense</i>	Shoots	Edema by fluid retention and inflammation
<i>Erythrina verna</i>	Bark	Mild cases of anxiety and insomnia, sedative
Table 1: continuation		
<i>Eucalyptus globulus</i>	Leaves	Colds and flus to clear airway as an adjunct in the treatment of bronchitis and asthma
<i>Eugenia uniflora</i>	Leaves	Noninfectious diarrhea
<i>Glycyrrhiza glabra</i>	Root	Coughs, colds and flus
<i>Hamamelis virginiana</i>	Bark	Skin inflammations and mucous membranes, hemorrhoids
<i>Harpagophytum procumbens</i>	Root	Joint pain (arthritis, arthrosis, arthralgia)
<i>Illicium verum</i>	Fruit	Bronchitis, expectorant
<i>Lippia sidoides</i>	Leaves	Gargles, mouthwashes and rinses
<i>Malva sylvestris</i>	Leaves and flowers	Respiratory expectorants
<i>Matricaria recutita</i>	Flowers	Intestinal cramps, mild anxiety cases, mild tranquilizer
<i>Maytenus ilicifolia</i>	Leaves	Dyspepsia, heartburn and gastritis, adjuvant ulcer prevention treatment
<i>Melissa officinalis</i>	Inflorescence	Abdominal cramps, mild anxiety and insomnia cases, mild tranquilizer
<i>Mentha x piperita</i>	Leaves and inflorescence	Colic, flatulence, liver problems
<i>Mentha pulegium</i>	Shoots	Respiratory expectorant, appetite stimulant, digestive disturbances, gastrointestinal spasms
<i>Mikania glomerata</i>	Leaves	Colds and flus, allergic and infectious bronchitis, expectorant
<i>Momordica charantia</i>	Fruit, and seeds	Dermatitis and scabies

<i>Passiflora alata</i>	Leaves	Mild anxiety and insomnia cases, mild tranquilizer
<i>Passiflora incarnata</i>	Shoots	Mild anxiety and insomnia cases, mild tranquilizer
<i>Paullinia cupana</i>	Seeds	Fatigue, stimulant
<i>Peumus boldus</i>	Leaves	Dyspepsia, choleric and cholagogue
<i>Phyllanthus niruri</i>	Shoots	Elimination of small kidney stones
<i>Pimpinella anisum</i>	Fruit	Dyspepsia, gastrointestinal cramps
<i>Plantago major</i>	Leaves	Inflammations of the mouth and pharynx
<i>Polygonum punctatum</i>	Shoots	Varicose veins and varicose ulcers
Table 1: continuation		
<i>Psidium guajava</i>	Young leaves	Noninfectious diarrhea
<i>Punica granatum</i>	Fruit peel	Inflammation and infection of the mouth and pharynx anti-inflammatory
<i>Rhamnus purshiana</i>	Bark	Eventual intestinal constipation
<i>Rosmarinus officinalis</i>	Leaves	Circulatory disorders, antiseptic and healing
<i>Salvia officinalis</i>	Leaves	Dyspepsia and excessive sweating
<i>Sambucus nigra</i>	Flowers	Colds and flus
<i>Schinus terebinthifolia</i>	Bark	Vaginal inflammation, leukorrhea, hemostatic, astringent and healing
<i>Senna alexandrina</i>	Fruit and folioles	Eventual intestinal constipation
<i>Solanum paniculatum</i>	Whole plant	Dyspepsia
<i>Stryphnodendron dromadstrigens</i>	Bark	Injuries, healing and topical antiseptic on the skin, oral mucosa and genital
<i>Taraxacum officinale</i>	Whole plant	Dyspepsia, appetite stimulant and as a diuretic
<i>Uncaria tomentosa</i>	Bark	Joint pain (arthritis and osteoarthritis) and acute muscle anti-inflammatory
<i>Vernonia condensata</i>	Leaves	Pain and dyspepsia
<i>Zingiber officinale</i>	Rhizome	Sickness, nausea and vomiting of pregnancy, postoperative motion, dyspepsia

For irradiation, a set with samples, synthetic standards obtained by pipetting standard solutions (SPEX CertiPrep) in filter paper sheets, and certified reference material (Syenite, Table Mountain, STM) and (Rhyolite, Glass Mountain RGM, RGM) from United States Geological Survey were prepared. In IEA-R1 nuclear reactor at IPEN, this set was irradiated under a thermal neutron flux varying from 1 to $5 \times 10^{12} \text{ n cm}^{-2} \text{ s}^{-1}$, for a 8 h period. The samples were counted from 7 days to 15 days after cooling, depending on the half-life of the radionuclide produced in the irradiation. Gamma spectrometry was performed using an EG & G Ortec Ge-Hiperpure detector and associated electronic, with a resolution of 1.9 keV and 2.04 keV for ^{57}Co and ^{60}Co , respectively. The spectra analysis was done using the program VISPECT2 and the calculations using a spreadsheet. The concentrations were obtained by comparing the peak area of the element of interest in the sample spectrum with that in the reference materials according to the following expression:

$$C_{ai} = \frac{(A_{ai} w_p C_{pi}) e^{-\lambda(ta - tp)}}{A_{pi} w_a}$$

where:

C_{ai} = concentration of i component in the sample (mg g^{-1} or %)

C_{pi} = concentration of component i in the reference material (mg g^{-1} or %)

A_{ai} = activity of the element i in the sample (cps)

A_{pi} = Activity of the element i in the reference material (cps)

W_p and W_a = sample and reference material mass, respectively (g)

λ = decay constant of the radioisotope (t^{-1})

t_a , t_p = cooling time of the sample and reference material, respectively (min)

3. RESULTS AND DISCUSSION

According to Vaz (1995) many ions can be present initially in the soil, adsorbed in solid or in liquid phase, in conditions immediately available to plants and, therefore, can be moved through the central cylinder and cortex plants to shoots [10]. Desorption depends on the equilibrium constants that vary with factors such as type of nutrient, compound chemical form, temperature and pH. Despite the aerial parts of the plants are adapted to photosynthesis, they also have the ability to absorb water and nutrients from the external environment. Therefore, medicinal herbs contain chemical elements that can be made available to the human body on any type of herbs and their extracts consumption. Not all elements have biological functions fully described, but most of them is responsible for major activities in human metabolism and, therefore, are directly related to health maintenance and proper functioning of the body. Table 2 presents the results of the element concentrations determined by INAA.

Only eight elements were measured in all the samples (Ba, Co, Cr, Cs, Fe, Na, Rb and Zn). For the others many samples were below de detection limit or the element was not determined. The element Ta was determined only in the *Stryphnodendron adstringens* and *Solanum paniculatum* samples. Elements such as Hf, Th, and Zr were measured only in few samples such as *Erythrina verna*, *Polygonum punctatum*, *Calendula officinalis*, *Passiflora incarnata* and *Aesculus hippocastanum*.

Elements potentially toxic, depending on their concentration, were found in extract samples ranging from 0.006 to 0.45 mg g⁻¹ for As in *Achyrocline satureioides* and *Momordica charantia* respectively; Ba was found in the range of 0.5 to 24.7 mg g⁻¹ in *Sambucus nigra* and *Passiflora alata*, respectively; Rb ranged from 0.86 to 170 mg g⁻¹ in samples of *Lippia sidoides* and *Equisetum arvense*, respectively; Sb ranged from 0.0023 to 0.28 mg g⁻¹ in *Cymbopogon citratus* and *Psidium guajava*, respectively, and U was found in concentrations varying from 0.0057 to 0.05 mg g⁻¹ in the extract of the plants *Stryphnodendrom adstringens* and *Echinodorus macrophyllus*, respectively. It was also noted that the same plant extract can presente two or more of these potentially toxic elements as *Psidium guajava*, *Stryphnodendrom adstringens* and *Echinodorus macrophyllus*.

Table 2: Results obtained in INAA for the extracts of medicinal plants analyzed. The values showed correspond to the concentration in mg g⁻¹ ± and the uncertainty of the measurement obtained by error propagation.

Plantas	As	±1σ	Ba	±1σ	Br	±1σ	Ca	±1σ	Co	±1σ	Cr	±1σ	Cs	±1σ	Fe	±1σ	Hf	±1σ	K	±1σ	
1- <i>Achillea millefolium</i>	0.048	0.007	1.5	0.1	0.86	0.01			0.041	0.003	0.33	0.02	0.011	0.002	13.7	0.2	0.0031	0.0003	34342	14174	
2- <i>Achyrocline satureioides</i>	0.006	0.002	6.5	0.7	0.798	0.009			0.026	0.002	0.68	0.08			15.7	0.7					
3- <i>Aesculus hippocastanum</i>							220	36	0.023	0.002	0.59	0.05	0.003	0.001	38.6	0.7	0.0013	0.0004			
4- <i>Ageratum conyzoides</i>			10.6	0.6	3.79	0.02	245	24	0.01	0.288	0.017	0.26	0.02	0.012	0.001	31.1	0.3	0.0102	0.0004		
5- <i>Allium sativum</i>	0.05	0.005			0.117	0.005	467	175		0.031	0.002	1.61	0.09	0.026	0.002	25.2	0.5			8090	1635
6- <i>Anacardium occidentale</i>			11.6	0.7	0.680	0.006	370	57	0.007	0.12	0.01	0.32	0.02	0.0020	0.0006	4.8	0.2	0.010	0.001		
7- <i>Arctium lappa</i>	0.031	0.008	2.7	0.5	1.90	0.02				0.044	0.003	0.57	0.06	0.022	0.003	17.5	0.6	0.007	0.001		
8- <i>Arnica montana</i>			0.9	0.1	0.259	0.005			0.003	0.036	0.002	1.2	0.1	0.044	0.005	17.2	0.3	0.0021	0.0003		
9- <i>Baccharis trimera</i>			2.4	0.3	0.354	0.008			0.01	0.066	0.004	0.23	0.02	0.19	0.02	7.6	0.2			20271	8367
10- <i>Bidens pilosa</i>			8.1	0.5	2.33	0.01	757	182		0.099	0.006	0.28	0.02	0.025	0.001	14.8	0.2			17206	2030
11- <i>Calendula officinalis</i>			2.179	0.222	0	0.0	720.5	79.116		0.078	0.004	1	0.1	0.1	0.005	17.1	0.3			45018	21863.2
12- <i>Caesalpinia ferrea</i>			2.6	0.2	0.228	0.001	368	129	0.003	0.017	0.001	0.18	0.01	0.0029	0.0005	25.0	0.3			2433	491
13- <i>Casearia sylvestris</i>	0.026	0.004	3.1	0.3	2.05	0.01	222	20	0.01	0.045	0.003	0.23	0.02	1.6	0.1	8.8	0.2				
14- <i>Cinnamomum verum</i>					0.186	0.004	402	153	0.003	0.074	0.006	0.53	0.03	0.069	0.004	19.6	0.3			2525	518
15- <i>Citrus aurantium</i>	0.02	0.003	2.6	0.2			455	158		0.011	0.001	0.83	0.05	0.023	0.002	12.6	0.3			4076	829
16- <i>Cordia verbenacea</i>					0.13	0.01			0.01	0.081	0.005	2.6	0.2	0.11	0.01	18.0	0.4	0.0011	0.0004	9097	3773
17- <i>Curcuma longa</i>	0.015	0.004	1.4	0.3	1.59	0.012	370	34		0.037	0.002	0.30	0.02	0.039	0.003	10.3	0.2				
18- <i>Cymbopogon citratus</i>			6.9	0.5	0.337	0.006	1689	126		0.034	0.002	1.8	0.1	0.118	0.007	11.9	0.3				
19- <i>Cynara scolymus</i>			10.3	0.7	0.13	0.01			0.01	0.068	0.004	2.7	0.2	0.024	0.004	24.3	0.5	0.0015	0.0005	84502	34872
20- <i>Echinodorus macrophyllus</i>			16	1	17.14	0.08	521	66	0.02	0.63	0.04	1.5	0.1	0.064	0.005	69.0	1.1	0.0143	0.0016		
21- <i>Equisetum arvense</i>					0.96	0.01				0.093	0.006	0.56	0.04	3.0	0.4	17.8	0.4			30759	12701
22- <i>Erythrina verna</i>			17	1	0.197	0.003	492	72	0.003	0.45	0.04	0.24	0.02	0.115	0.007	6.3	0.2	0.006	0.001		
23- <i>Eucalyptus globulus</i>			8.2	0.6	0.47	0.01	134	31	0.009	0.062	0.005	0.48	0.05	0.011	0.002	8.5	0.4			5044	1470
24- <i>Eugenia uniflora</i>			26	2	0.259	0.008	605	55	0.008	0.57	0.05	0.58	0.06	0.037	0.004	9.0	0.4			9395	2719
25- <i>Glycyrrhiza glabra</i>			0.56	0.05	0.198	0.005	51	12	0.001	0.010	0.001	0.078	0.008	0.009	0.001	3.00	0.07			2044	597
26- <i>Hamamelis virginiana</i>	0.034	0.003	2.2	0.1	0.001	0.001	1240	187	0.002	0.0076	0.0007	0.30	0.02	0.0074	0.0004	4.54	0.05				
27- <i>Harpagophytum procumbens</i>			8.7	0.7	2.91	0.02	819	152	0.01	0.13	0.01	1.4	0.1	0.013	0.002	21.9	0.8			7145	2062
28- <i>Illicium verum</i>	0.044	0.018	2.1	0.4	0.45	0.01	315	141	0.009	0.033	0.002	0.19	0.04	0.24	0.02	20.5	0.6			12346	5638
29- <i>Lippia sidoides</i>			0.9	0.2	3.60	0.03	280	134	0.004	0.022	0.002	0.26	0.04	0.004	0.001	8.6	0.4			2285	1060
30- <i>Malva sylvestris</i>	0.044	0.015	6.0	0.5	4.88	0.04	634	297	0.02	0.16	0.01	1.2	0.2	0.048	0.005	23.2	0.6			23009	10461

Table 2: Continuation

Plantas	As	Na	$\pm 1\sigma$	Rb	$\pm 1\sigma$	Sb	$\pm 1\sigma$	Sc	$\pm 1\sigma$	Se	$\pm 1\sigma$	Ta	$\pm 1\sigma$	Th	$\pm 1\sigma$	U	$\pm 1\sigma$	Zn	$\pm 1\sigma$	Zr	$\pm 1\sigma$
1- <i>Achillea millefolium</i>	243	6	6.3	0.2	0.025	0.002	0.0037	0.0001						0.0032	0.0004			8.5	0.4	0.49	0.09
2- <i>Achyrocline satureioides</i>	273	7	16.3	0.5	0.0080	0.0009	0.0036	0.0002										8.4	0.6		
3- <i>Aesculus hippocastanum</i>	14.9	0.4	1.99	0.08	0.0117	0.0009	0.00063	0.00005						0.003	0.001			5.5	0.2	19	1
4- <i>Ageratum conyzoides</i>	624	14	13.2	0.5			0.0117	0.0004	0.04	0.01				0.016	0.001	0.021	0.003	5.7	0.2	0.45	0.07
5- <i>Allium sativum</i>	843	20	5.6	0.2	0.0134	0.0008	0.00132	0.00008	0.11	0.03								16.0	0.7		
6- <i>Anacardium occidentale</i>	388	9	4.9	0.2	0.0032	0.0003	0.00118	0.00005	0.016	0.006				0.0116	0.0006			1.77	0.07		
7- <i>Arctium lappa</i>	107	3	29.4	0.8	0.019	0.002	0.0009	0.0001						0.20	0.01			26	2		
8- <i>Arnica montana</i>	281	7	16.7	0.5	0.0072	0.0008	0.0062	0.0002										16	1		
9- <i>Baccharis trimera</i>	554	13	82	2			0.0034	0.0002						0.0034	0.0007			6.3	0.3		
10- <i>Bidens pilosa</i>	129	3	44	2	0.011	0.001	0.0027	0.0001						0.0041	0.0005						
11- <i>Calendula officinalis</i>	864.3	19.5	50	2.0	0.02	0.001	0.004	0.0002	0.046	0.01				0.009	0.001						
12- <i>Caesalpinia ferrea</i>	74	2	3.0	0.1	0.0025	0.0001	0.00060	0.00003	0.011	0.005				0.0031	0.0003			1.72	0.07		
13- <i>Casearia sylvestris</i>	166	4	27	1			0.0049	0.0002						0.005	0.001			7.2	0.3		
14- <i>Cinnamomum verum</i>	181	4	6.6	0.3	0.0047	0.0004	0.00139	0.00005						0.0024	0.0003			3.6	0.1		
15- <i>Citrus aurantium</i>	466	11	7.8	0.3	0.0069	0.0006	0.00057	0.00005										2.02	0.09	0.5	0.2
16- <i>Cordia verbenacea</i>	3124	71	16.2	0.4			0.0107	0.0003						0.009	0.001			2.5	0.1		
17- <i>Curcuma longa</i>	192	4	50	2			0.0068	0.0002						0.010	0.001			9.1	0.4		
18- <i>Cymbopogon citratus</i>	119	3	41	2	0.28	0.04	0.0025	0.0001										5.2	0.2		
19- <i>Cynara scolymus</i>	1952	44	22.56	0.52	0.024	0.003	0.0059	0.0002						0.0091	0.0012			9.1	0.4		
20- <i>Echinodorus macrophyllus</i>	387	9	75	3			0.0072	0.0003						0.018	0.002	0.05	0.01	2.6	0.1		
21- <i>Equisetum arvense</i>	415	9	170	7	0.07	0.01	0.0005	0.0001										8.0	0.3		
22- <i>Erythrina verna</i>	39	1	12.2	0.5	0.0027	0.0002	0.00056	0.00003						0.0018	0.0004			2.4	0.1		
23- <i>Eucalyptus globulus</i>	286	7	14.6	0.6	0.008	0.001	0.0080	0.0003						0.008	0.001			4.0	0.2	1.0	0.3
24- <i>Eugenia uniflora</i>	142	4	48	2	0.006	0.001	0.0034	0.0002						0.0037	0.0007			7.3	0.3	1.8	0.4
25- <i>Glycyrrhiza glabra</i>	114	3	0.94	0.04	0.031	0.006	0.0024	0.0001						0.0027	0.0002			1.09	0.05	0.56	0.07
26- <i>Hamamelis virginiana</i>	190	4	2.56	0.09	0.0030	0.0002	0.00034	0.00001										2.6	0.1		
27- <i>Harpagophytum procumbens</i>	0.20	0.05	4.6	0.2	0.025	0.002	0.0153	0.0006	0.036	0.020				0.017	0.002			8.4	0.4	2.6	0.6
28- <i>Illicium verum</i>	71	2	26	1	0.018	0.002	0.0019	0.0001						0.0023	0.0007			3.7	0.2		
29- <i>Lippia sidoides</i>	27.9	0.8	0.86	0.04	0.008	0.001	0.0031	0.0001						0.0027	0.0005			5.5	0.2	0.4	0.2
30- <i>Malva sylvestris</i>	169	4	20.7	0.9	0.050	0.005	0.0118	0.0004						0.034	0.002	0.016	0.002	18.2	0.8	3.2	0.7

Table 2: Continuation

Plantas	As	±1σ	Ba	±1σ	Br	±1σ	Ca	±1σ	Co	±1σ	Cr	±1σ	Cs	±1σ	Fe	±1σ	Hf	±1σ	K	±1σ	
31- <i>Matricaria recutita</i>			1.7	0.4	1.28	0.02	1653	813	0.14	0.01	1.1	0.2	0.074	0.007	12.5	0.4			43632	19838	
32- <i>Maytenus ilicifolia</i>			4.2	0.4	1.18	0.01	472	240	0.007	0.035	0.002	0.43	0.07	0.020	0.003	5.3	0.3		20541	9336	
33- <i>Melissa officinalis</i>			8.7	0.6	0.337	0.003	2505	436	0.081	0.007	0.69	0.07	0.067	0.006	7.6	0.3	0.0011	0.0004	11993	3459	
34- <i>Mentha x piperita</i>			0.9	0.1	0.068	0.002	287	32	0.031	0.002	0.40	0.02	0.02	0.002	7	0.1			17939	8712	
35- <i>Mentha pulegium</i>			3.4	0.7			457	118	0.026	0.827	0.094	1.4	0.3	0.019	0.005	75.9	1.8				
36- <i>Mikania glomerata</i>			1.8	0.2			1680	146	0.004	0.036	0.004	0.8	0.2	0.169	0.020	11.4	0.2				
37- <i>Momordica charantia</i>	0.45	0.03	18	1	0.44	0.01			0.05	0.48	0.03	0.79	0.08	0.10	0.01	13.5	0.4	0.011	0.001		
38- <i>Passiflora alata</i>			24.7	1.5			1603	139	0.010	0.031	0.004	1.8	0.4	0.021	0.003	19.4	0.5	0.0018	0.0007		
39- <i>Passiflora incarnata</i>	0.011	0.003	1.4	0.2			1655	454	0.010	0.038	0.004	0.48	0.07	0.552	0.061	31.8	0.6	0.0053	0.0009		
41- <i>Peumus boldus</i>	0.02	0.01	1.2	0.2			249	28	0.008	0.034	0.004	0.8	0.2	0.017	0.003	13.4	0.5				
42- <i>Phyllanthus niruri</i>	0.02	0.004	6.38	0.50	0.60	0.01	753.35	175.01	0.01	0.12	0.01	0.29	0.02	0.01	0.001	18.00	0.42		10320.32	1218.01	
43- <i>Pimpinella anisum</i>			4.8	0.3	65	1	3310	2381	0.045	0.003	0.39	0.03	0.013	0.001	14.5	0.3			8035	2941	
44- <i>Plantago major</i>			10.9	0.7	0.200	0.003	1994	511	0.01	0.584	0.033	0.34	0.02	0.038	0.002	15.1	0.3	0.0122	0.0010	8273	977
45- <i>Polygonum punctatum</i>			6.4	0.5	0.582	0.009	139	23	0.01	0.30	0.02	0.46	0.03	0.048	0.003	17.9	0.4	0.0012	0.0005	25734	3044
46- <i>Psidium guajava</i>	0.015	0.006	1.1	0.2			73	23	0.031	0.004	0.55	0.08	0.008	0.002	8.3	0.3					
47- <i>Punica granatum</i>	0.014	0.006	3.0	0.3	21.9	0.4	255	185	0.006	0.043	0.003	0.37	0.03	0.025	0.002	10.4	0.2		7010	2566	
48- <i>Rhamnus purshiana</i>			13	1	94	3	1319	899	0.072	0.006	2.3	0.2	0.016	0.006	29	1					
49- <i>Rosmarinus officinalis</i>	0.063	0.013	3.8	0.3			2640	253	0.008	0.091	0.010	1.07	0.15		26.4	0.7	0.0030	0.0007			
50- <i>Salvia officinalis</i>	0.026	0.003	2.6	0.2	2.190	0.009	1037	261	0.01	0.066	0.004	0.53	0.03	0.044	0.002	21.3	0.3	0.0090	0.0006	21480	2534
51- <i>Sambucus nigra</i>			0.5	0.2			333	99	0.028	0.005	0.3	0.1	0.006	0.002	9.3	0.4					
52- <i>Schinus terebinthifolia</i>	0.033	0.005	25	1	0.338	0.007	813	29	0.009	0.071	0.006	0.89	0.05	0.019	0.002	15.6	0.3	0.031	0.001		
53- <i>Senna alexandrina</i>	0.014	0.004	2.8	0.3	0.18	0.01	2481	905	0.038	0.002	1.71	0.10	0.016	0.002	13.8	0.3			9693	1965	
54- <i>Solanum paniculatum</i>			3.6	0.3			275	36	0.016	0.096	0.011	2.65	0.36	0.113	0.013	28.5	0.5				
55- <i>Stryphnodendrom adstrigens</i>	0.011	0.002	3.0	0.2	0.289	0.004	302	47	0.007	0.0108	0.0009	0.45	0.02	0.016	0.001	39.4	0.4	0.0028	0.0002		
56- <i>Taraxacum officinale</i>			7.3	0.4	0.717	0.006	2263	337	0.006	0.19	0.02	0.89	0.07	0.111	0.006	41.2	0.6	0.013	0.001		
57- <i>Uncaria tomentosa</i>	0.050	0.008	2.1	0.3	0.180	0.009			0.012	0.032	0.003	1.15	0.06	0.021	0.002	34.9	0.7				
58- <i>Vernonia polyanthes</i>			12.1	0.7			842	82	0.007	0.057	0.006	0.44	0.06	0.034	0.004	11.3	0.2				
59- <i>Zingiber officinale</i>	0.20	0.01	2.3	0.3	0.536	0.004			0.102	0.009	0.52	0.03			34.3	0.5					

Table 2: Continuation

Plantas	Na	$\pm 1\sigma$	Rb	$\pm 1\sigma$	Sb	$\pm 1\sigma$	Sc	$\pm 1\sigma$	Se	$\pm 1\sigma$	Ta	$\pm 1\sigma$	Th	$\pm 1\sigma$	U	$\pm 1\sigma$	Zn	$\pm 1\sigma$	Zr	$\pm 1\sigma$
1- <i>Achillea millefolium</i>	243	6	6.3	0.2	0.025	0.002	0.0037	0.0001					0.0032	0.0004			8.5	0.4	0.49	0.09
31- <i>Matricaria recutita</i>	275	7	60	3	0.009	0.002	0.0019	0.0001									13.4	0.6		
32- <i>Maytenus ilicifolia</i>	41	1	33	1	0.013	0.001	0.0043	0.0002					0.0033	0.0007			6.7	0.3		
33- <i>Melissa officinalis</i>	112	3	15.4	0.6	0.026	0.002	0.0046	0.0002					0.0058	0.0008			8.9	0.4	1.2	0.3
34- <i>Mentha x piperita</i>	344.4	7.8	20	1			0.0017	0.0001	0.018	0.006			0.004	0.000						
35- <i>Mentha pulegium</i>	655	21	19.6	1.2	0.0277	0.0023	0.0339	0.0012					0.0259	0.0026			27.7	1.8	4.8	1.0
36- <i>Mikania glomerata</i>	933	30	135.1	7.9	0.0153	0.0013	0.0032	0.0001					0.0027	0.0004			6.8	0.4	1.1	0.2
37- <i>Momordica charantia</i>	581	15	8.7	0.2	0.034	0.003	0.0034	0.0001									8.6	0.6		
38- <i>Passiflora alata</i>	146	5	54.3	3.2	0.0148	0.0006	0.0049	0.0002					0.0058	0.0008			13.8	0.9		
39- <i>Passiflora incarnata</i>	161	5	40.2	2.4	0.0320	0.0028	0.0277	0.0010					0.1093	0.0071	0.019	0.002	23.2	1.5		
41- <i>Peumus boldus</i>	264	9	9.0	0.5	0.0086	0.0010	0.0073	0.0003					0.0058	0.0009			5.9	0.4		
42- <i>Phyllanthus niruri</i>	100.01	2.23	5.83	0.22	0.001	0.0002	0.02	0.001					0.01	0.001					0.52	0.24
43- <i>Pimpinella anisum</i>	150	5	4.8	0.2	0.0043	0.0006	0.00136	0.00006									4.8	0.3		
44- <i>Plantago major</i>	85	2	14.7	0.5	0.006	0.001	0.033	0.001					0.0075	0.0006					1.4	0.3
45- <i>Polygonum punctatum</i>	165	4	22.2	0.8	0.009	0.002	0.0040	0.0002					0.005	0.001					2.0	0.4
46- <i>Psidium guajava</i>			6.3	0.4	0.0023	0.0007	0.0021	0.0001					0.0038	0.0007	0.023	0.007	8.7	0.6		
47- <i>Punica granatum</i>	97	3	14.1	0.5	0.0057	0.0005	0.00223	0.00009					0.0070	0.0007			2.5	0.2		
48- <i>Rhamnus purshiana</i>	838	29	12.8	0.5													128	9		
49- <i>Rosmarinus officinalis</i>	260	9	3.2	0.2	0.0042	0.0010	0.0115	0.0004					0.0091	0.0010			18.3	1.2		
50- <i>Salvia officinalis</i>	132	3	9.4	0.3	0.017	0.001	0.0232	0.0008					0.039	0.002	0.015	0.002			1.1	0.2
51- <i>Sambucus nigra</i>	109	4	5.6	0.3	0.0056	0.0007	0.0015	0.0001									13.7	0.9	0.7	0.3
52- <i>Schinus terebinthifolia</i>	668	16	5.3	0.2	0.026	0.001	0.0031	0.0001					0.011	0.001			14.6	0.6		
53- <i>Senna alexandrina</i>	749	18	4.6	0.2	0.0059	0.0008	0.00113	0.00007	0.15	0.02							5.3	0.2		
54- <i>Solanum paniculatum</i>	233	8	21.5	1.3	0.0359	0.0030	0.0067	0.0002			0.0014	0.0004	0.0224	0.0016			11.0	0.7	3.5	0.4
55- <i>Stryphnodendrom adstrigens</i>	455	11	9.3	0.3	0.114	0.004	0.0040	0.0001	0.018	0.003	0.0007	0.0001	0.017	0.001	0.0057	0.0006	6.3	0.2	4.7	0.4
56- <i>Taraxacum officinale</i>	333	8	64	2	0.0067	0.0005	0.0151	0.0005	0.023	0.004			0.0329	0.0004	0.023	0.002	7.6	0.3	0.31	0.06
57- <i>Uncaria tomentosa</i>	775	18	8.0	0.3	0.020	0.001	0.0204	0.0007					0.009	0.001			7.9	0.3	1.2	0.3
58- <i>Vernonia polyanthes</i>	175	6	36.1	2.1	0.0052	0.0007	0.0039	0.0001					0.0049	0.0005	0.017	0.005	14.4	0.9		
59- <i>Zingiber officinale</i>			8.4	0.3	0.0108	0.0004	0.0042	0.0002	0.090	0.024			0.0054	0.0009			16.6	0.6		

Among the elements that play important role in the body some features can be drawn, for example, for calcium that plays a key role in strengthening bones and teeth and also participating in blood coagulation which the dose reference for the population is 700 mg per day of intake and adequate intake for people over 51 years is 1200 mg [11] was found in highest concentration in *Melissa officinalis* species, with 2,505 mg g⁻¹. Plants *Agerantum conyzoides*, *Harpagophytum procumbens* and *Uncaria tomentosa* are known for their ability of improving function in joint pain (arthritis, arthrosis, arthralgia and reumastimos) also found with relatively high concentration of this element.

Most of the iron in the human body is present in erythrocytes as hemoglobin. Its main function is oxygen transport from lungs to tissues. Also, Fe is an important component of many enzyme systems, such as cytochromes involved in oxidative metabolism. Iron is an essential element for human nutrition and estimates of minimum daily requirement depends on the age, gender, physiological status varies from 10 to 50 mg per day (12). This element was measured in all samples ranging from 3 to 75 mg g⁻¹. The species *Schinus terebinthifolia*, having hemostatic action, has Fe concentration of 39.4 mg g⁻¹.

Selenium, which has a high antioxidant capacity, i.e., neutralizes the action of free radicals (formed by the sunlight action, pollution, cigarette smoke, etc.) in the body, slowing the aging process and preventing the onset of some cancer forms, aids in maintaining the body in defense against infection and modulates growth and development. Recommended daily doses were fixed at 0.9 mg per kg of the body weight for adults [13]. In the samples, Se is present in concentrations ranging from 0.011 to 0.15 mg g⁻¹ in samples of *Casealpinia ferrea* and *Senna Alexandria*, respectively. *Allium sativum*, a well-known anti-hypercholesterolemic plant contains this element in the amount of 0.11 mg g⁻¹.

Zinc is an essential component of a large number of enzymes and plays a central role in cell growth and tissue differentiation. This element acts in tissue repair and wound healing. Deficiency results in several autoimmune diseases and causes severe lymphopenia (large decrease in the number of lymphocytes). The recommended daily intake for adult men is 15 mg [14]. Zinc was measured in almost all the analyzed samples. In the extract of the plants with healing and antiseptic action this element was found in concentrations ranging from 1.72 to 8.6 mg g⁻¹ in the species *Casealpinia ferrea* and *Momordica charantia*, respectively.

Sodium is the major electrolyte found in extracellular fluid and potassium is the predominantly intracellular electrolyte. They have mainly the function in regulating the sodium and potassium pump, the maintenance of cellular physiology and the maintenance of the homeostatic pressure [15]. The extract samples of *Echinodorus macrophyllus* and *Equisetum arvense*, which are indicated to assist the edema treatment caused by fluid retention and in inflammatory process showed concentrations of 387 and 415 mg g⁻¹ of sodium, respectively.

3. CONCLUSIONS

The technique used in this study showed adequate sensitivity for the determination of the elements present in the plant extracts analyzed, the concentrations of those elements potentially toxic As, Ba, Rb, Sb, Th and U were not found in concentrations that may present

some threat to human body. Furthermore, elements that play important role in the metabolism such as Ca, Fe, Se, Zn, Na and K species were found in concentrations that indicate possible influence in the pharmacological action of the plants considering their therapeutic application. Besides that, it can be also concluded that the introduction of these extract in the regular diet can assist in deficiency cases.

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REFERENCES

- 1- Oliveira, E. R.; Menini Neto, L. **Levantamento etnobotânico de plantas medicinais utilizadas pelos moradores do povoado de Manejo, Lima Duarte – MG.** Revista Brasileira de Plantas Medicinais, vol. 14, nº. 2, p. 311-320, 2012.
- 2- Gonçalves, R. D. M. R; Francisconi, L. S; Silva, P. S. C. **Inorganic constituents in herbal medicine by neutron activation analysis.** In: International Nuclear Atlantic Conference - INAC 2011, 2011, Belo Horizonte. Anais da International Nuclear Atlantic Conference - INAC 2011, 2011. Rates, S. M. K. **Plants as source of drougs.** Toxicon, n. 39, p. 603-613, 2001.
- 3- Yunes, R. A.; Pedrosa, R. C.; Cechinel, F. V. **Fármacos e fitoterápicos: a necessidade do desenvolvimento da indústria de fitoterápicos e fitofármacos no Brasil.** Quimica Nova, vol. 24, nº. 1, p. 147-152, 2001.
- 4- Calixto, J.B. Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines. *Brazilian Journal of Medical and Biological Research*, v. 33, p. 179-189, 2000.
- 5- Kobers, Vunibaldo Cirilo; SANTOS, Crescêncio Roque Ribeiro dos. **Plantas medicinais.** Francisco Beltrão: Grafti, 2007. 13p. 4Chen, C. Y.; Pan, L. K. **Trace elements of Taiwanese dioscorea spp. using instrumental neutron activation analysis.** Food Chemistry, vol.72, p. 255-260, 2001.
- 6- Amorozo, Maria Christina de Mello. **Agricultura tradicional, espaços de resistência e o prazer de plantar.** In: Simpósio brasileiro de etnobiologia e etnoecologia, 2002, Recife.
- 7- Machado, T.B. *In vitro* activity of Brazilian medicinal plants, naturally occurring naphthoquinones and their analogues, against methicillin-resistant *Staphylococcus aureus*. **International Journal of Antimicrobial Agents**, v.21, n.3, p.279-284, 2003.
- 8- Chen, C.Y., Pan, L.K.: **Food Chem.** 72, 255 (2001)
- 9- IAEA, **Practical aspects of operating a neutron analysis laboratory**, TECDOC – 564 (1990) International Atomic Energy Agency, Vienna.
- 10- Vaz, S. M. **Análise de extratos de plantas medicinais pelo método de ativação com nêutrons.** 1995. Dissertação (Mestrado) - Instituto de Pesquisas Energéticas e Nucleares, SP.
- 11- FAO/WHO. **Human vitamin and mineral requirements.** Report of a Joint FAO/WHO Expert Consultation, Bangkok, Thailand. Rome: World Health Organization and Food and Nutrition Organization of the United Nations, 2002

- 12- FAO/WHO. **Requirements of vitamin A, iron, folate and vitamin B12.** Report of a Joint FAO/WHO Expert Consultation. Rome, Food and Agriculture Organization of the United Nations, 1988 (FAO Food and Nutrition Series, No. 23).
- 13- Golden M. H. **Proposed recommended nutrient densities for moderately malnourished children.** 2009 *Food and Nutrition Bulletin*, vol. 30, no. 3 (supplement)
- 14- Hambidge H. **Human zinc deficiency.** *J Nutr* 2000;130:1344S-1349S.
- 15- Sheng, H-P. **Sodium, chloride and potassium.** In: Stipanuck MH, ed. *Biochemical and physiological aspects of human nutrition.* Philadelphia: WB Saunders Company, 2000:686-710.