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Determination of elements in Salvia officinalis and Polygonum equisetiforme

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Abstract

The elemental content of two herbs, Salvia officinalis and Polygonum equisetiforme, grown in Libyan wilderness was analysed using four different digestion methods for sample preparation. the samples. The elements which determined in these plants were Ca, Mg, Na, K, Fe, P, Si, Mn, Zn, Cu, Co, Ni, Cr, Al, Hg, Pb and Cd, measured by ICP-AES technique. The macronutrients Ca, Mg, K, Na, P and Fe represent the highest concentrations in these samples ranging from Ca which owns the highest continent to Fe with the lowest concentration. The result also exhibits the highest micronutrient composition was detected for Si, Mn and Zn respectively. The toxic element, Al, was found at high level compared to other toxic elements which could be related to environmental content.
Statistical analysis (PCA, PLS-DA and HCA) data reveal that there are significant differences between water extraction method and other digestion methods. These differences could be explained by the ability of water to extract soluble salts only. Meanwhile there is a slight difference between ashing procedure and other digestion methods HNO3/H2O2 and HNO3/HClO4 methods). The results also show significant difference in terms of elemental content. For example, the samples have significant differences in the concentrations of Mn, Cu and Ni. While no significant differences were observed in the concentrations of Cr and Sn.
Key words: herbs, elements, digestion, water extract.

Introduction

From prehistoric time a many of herbal plants are being utilized as potential treatment for many diseases [1-5]. Lots patients have been turning to herbal treatment instant of chemical medical. The herbal plants are considered to be a good source for new medicine for fight many diseases like malaria, cancer, central nervous and disorder of cardiovascular [5, 6].

Herbal plants consist of essential and trace elements which could support the human body from any type of consumption of medical plants and their extracts. For most of the elements, there is a narrow range between the toxicity and deficiency for the body of human. Therefore, the health guidelines for herbal consumption are often difficult. The geochemical features of the soil

usually governed the content of major and trace elements in the medical plants. In addition, plants have the ability to accumulate elements from the environment, enabling many plants to be utilized as indicators for pollution of environment [7-10]

Due to the lake of data base of the content of elements in many Libyan herbal plants, this research was carried out to determine major, minor, trace elements and some toxic elements in Salvia officinalis and Polygonum Equisetiforme. In addition, to develop a reliable digestion method to extract the elements from the complex matrix of the selected medical plants.

Methodes

Chemicals

Deionized water produced through distillation and ion exchange resins treatment was supplied by West Tripoli Power Station. The chemicals which were used in this study were analytical grade and supplied by Farmitalia Cario Erba, Fluka chemika, Merck (Darmstadt, Germany) and Ventron Gmbh.

Sample preparation

Herbal plants were collected at the end of spring and washed with distilled and deionized water then air dried for approximately three weeks, after that, the plant species were finely chopped and dried in an oven at 90C° for about 24 hours. these plants were grounded in a ceramic mortar and stored in glass jar. In this work, three digestion procedures and water extraction were used to extract the elements from plant matrix.

Wet digestion Procedure 1

20 grams of individual plant samples were introduced into 80-mL beakers then 20 ml of con. HNO_3 to each beaker and covered with watch glasses after NO_2 bubble was removed. The next procedure, the mixtures of the samples were refluxed gently until the samples volume became less, then the contents were allowed to cool for around 5 minutes at room temperature, after that 5 ml of $HClO_4$ were added and boiled to almost dryness then the content were allowed to cool to room temperature. The content was dissolved in 0.1N HNO_3, boiled, then cold to room temperature then filtered through a glass filter into 50-ml volumetric flasks and diluted with 0.1N HNO_3 to the marks.

Wet digestion Procedure 2

The wet digestion procedure followed the same steps as the pervious method, except that of H_2O_2 was used instead of $HClO_4$ for the treatment of the sample mixture.

Dry ashing Procedure

2.00 g of each plant sample were introduced into crucibles and then placed in a muffle furnace, where they were slowly heated to 500°C for 8 hrs. The crucibles were transferred from the furnace and allowed to cool. The ash was then dissolved in 10 ml of nitric acid (1:1) and filtered through a glass filter. The filtrate was transferred into 50-mL volumetric flasks and diluted to the marks with 0.1N HNO_3.

Water infusion procedure

2.000 grams of plant samples were introduced into 80-mL beakers and covered with watch glasses. A sufficient water was added to each beaker and refluxed for 1 hour, then filtered and transferred into 50-mL volumetric flasks and diluted with 0.1N HNO_3. This method is similar to traditional procedure to extract some of pharmacologically active compounds and minerals.

Instruments

Samples were analyzed for, Ca, Mg, K, Na, P, Fe, Si, Mn, Zn, Cu, Co, Cr, Ni, Sn, Al, Hg, Pb and Cd by Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-MS) technique, Model S (Industrial research center, Tripoli-Libya). Na and K were measured by flame photometer.

Result and discussion

Macro, micro and some of toxic element's concentrations were determined in two of Libyan wild medicinal plants. The studied medicinal plants were: Salvia officinalis and Polygonum Equisetiforme. Three digestion methods and water infusion method were used to extract elements from plant tissues. The screening was performed by Inductively Coupled Plasma Atomic Emission Spectroscopy technique (ICP-MS).

The first part of the discussion deals with the concentration of each studied element in selected Libyan medicinal plants. The concentration of these elements was presented in the corresponding labeled graphs and tables.

Macronutrients elements

Regardless of sample preparation method, the macronutrients, Calcium, Magnesium, Potassium, Sodium, phosphorus and iron were found to be dominate contents in both plants.

The mean concentration of macronutrients elements relay on digestion methods with a mixture of (HNO_3/$HClO_4$), a mixture of (HNO_3/H_2O_2) acids, dry ashing and water infusion procedures were shown in graph 1 and table 1:

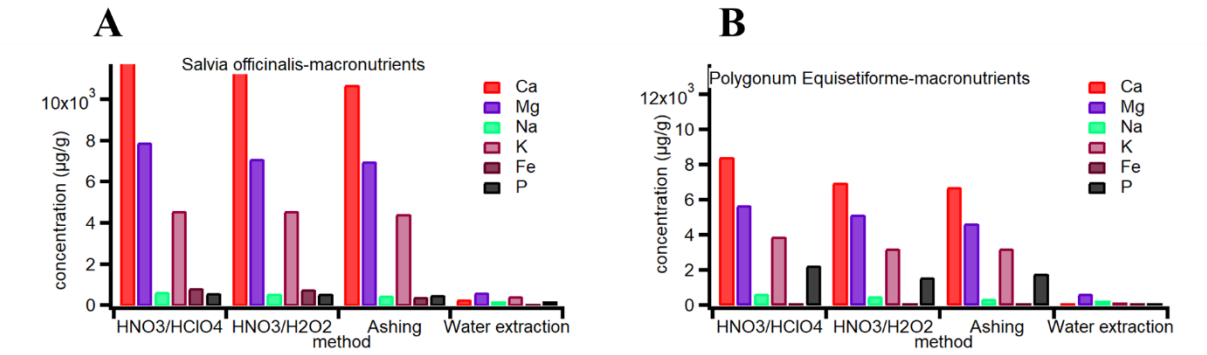

Figure 1: The concentrations of macronutrients elements in S. officinalis and P. equisetiforme using different digestion methods.

Table 1: The mean concentrations (µg/g) of macronutrients in S. officinalis and P. equisetiforme using different digestion methods*.

plant	method	Ca	Mg	Na	K	Fe	P
S. Officinalis	$HNO_3/HClO_4$	12279	7942	673	4623	859	635
	HNO_3/H_2O_2	11578	7129	593	4622	795	590
	ashing	10712	7003	518	4458	431	536
	water infusion	313	663	234	464	4.8	239
P. equisetiforme	$HNO_3/HClO_4$	8462	5722	695	3923	139	2284
	HNO_3/H_2O_2	6987	5199	537	3250	125	1623
	ashing	6755	4698	426	3259	117	1847
	water infusion	12.9	684	299	195	8.4	25

*Average of triplicate determinations, %R.S.D.= 0.04-0.20

In both plant samples, the concentration of calcium is higher than magnesium and potassium over the sodium regardless any method used for sample preparation, which could be related to the selectivity of these plants towards the elements. The data also showed that the concentrations of phosphorus and iron were lower compared to calcium and magnesium; however, phosphorus and iron were still present in significant amounts in these plants, indicating that they could serve as a good source for human body. It is noticed that, the elemental contents which extracted by water are lower compared to other digestion methods. This could be related to the lower solubility of inorganic and organic compound of the elements in water. In spite of that, these macronutrients exist in aqueous extract in sufficient amounts which could be contribute to daily allowance recommended by WHO. It is clear from the data for most samples, that concentrations of the macronutrients are found to be in the given order:

Ca > Mg > K> Na > P > Fe

According to the natural abundance of micronutrients, the concentrations of most these elements were found to follow ~~with~~ the expected ~~given~~ order [5, 11-19]

The micronutrients elements

The concentrations of micronutrients in both herbal plants extracted using acid digesting, methods, dry ashing, and water infusion methods are shown in Figure 2 and Table 2.

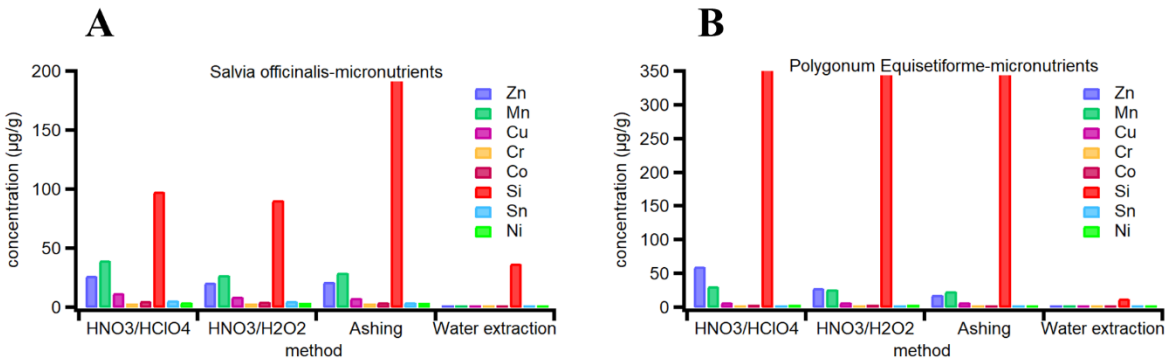

Figure 2: The concentrations of micronutrients elements in S. officinalis and P. equisetiforme using different digestion methods

Table 2: The mean concentrations (µg/g) of micronutrients in medical plants*

plant	method	Zn	Mn	Cu	Cr	Co	Si	Sn	Ni
S. Officinalis	$HNO_3/HClO_4$	26.97	40.34	12.69	3.66	5.86	98.67	6.31	4.74
	HNO_3/H_2O_2	21.53	27.71	9.54	3.67	5.00	91.07	5.74	4.37
	ashing	21.76	29.80	8.18	3.64	4.56	250	4.80	3.94
	water infusion	1.83	0.73	0.86	0.39	1.15	137.6	0.00	0.37
P. equisetiforme	$HNO_3/HClO_4$	61	32	8.1	4.1	4.8	498	2.07	4.82
	HNO_3/H_2O_2	29	27	8.0	3.3	4.2	414	2.01	4.14
	ashing	19	25	7.8	3.6	3.6	577	2.05	3.8
	water infusion	1.4	1.0	0.74	0.34	0.39	14.1	0.00	0.7

*Average of triplicate determinations, %R.S.D.= 0.03-0.2

The data indicate that the micronutrient silicon has the highest concentration followed by manganese while, the tin has the lowest concentration. For most samples, the order of the concentrations of micronutrients was found to follow this order:

Si >> Mn > Zn > Cu > Co > Ni > Cr > Sn

The concentrations of these micronutrients were found to be in a good agreement with many other working on herbal plants in different places around the word [5, 14, 16, 17, 19, 20].

The toxic elements

Aluminum concentration is found relatively high compared to other toxic elements which could related to soil and environmental content of aluminum or due to the selectivity of these herbals towards this element, as illustrated in Figure 3 and Table 3.

The relative concentrations of toxic elements were found to be in agreement with the finding of many others studies on herbal plants from different region of the world [14, 18].

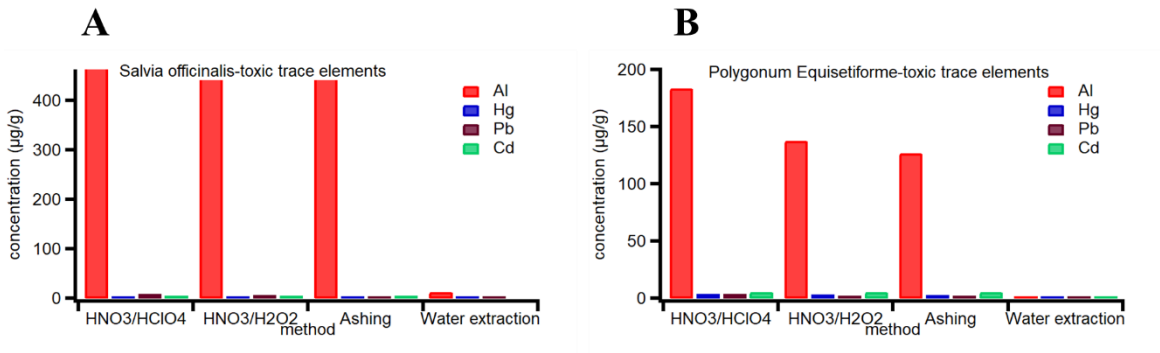

Figure 3: The concentration of toxic elements in S. officinalis and P. equisetiforme with different digestion methods

Table 3: The mean concentration of toxic elements in medical plants*

plant	method	Al	Hg	Pb	Cd
S. Officinalis	$HNO_3/HClO_4$	1093	4.58	9.34	5.65
	HNO_3/H_2O_2	984	3.84	7.73	5.63
	ashing	919	2.39	4.43	5.67
	water infusion	13.12	0.00	0.40	0.00
P. equisetiforme	$HNO_3/HClO_4$	184	4.29	4.31	5.63
	HNO_3/H_2O_2	138	3.52	2.75	5.60
	ashing	127	2.88	2.74	5.61
	water infusion	0.79	0.00	0.36	0.51

*Average of triplicate determinations, %R.S.D.= 0.04-0.26.

Statistical analysis

To examine the significant differences between selected medical plants in terms of the concentration of elemental composition and also to discover the significant differences between the methods which used to prepare the samples, statistical analyses including Principal Component Analysis (PCA), Partial Least Squares Discriminant Analysis (PLS-DA), and Hierarchical Cluster Analysis (HCA) were employed. The PCA results demonstrated a strong separation between each plant from the others. For example, the PCA result showed that there is a significant difference between S. officinalis and P. equisetiforme in terms of elemental composition, with the first two principal components explaining 94% of the total variance, as depicted in Figure 4**.** Also, the PCA result exhibit that there is significant difference between water extracting method and other methods (wet and dry methods). The difference could be explained by the elemental composition that extracted by water only represent the soluble salts only. There is a slight difference between ashing method and other two methods (HNO_3/H_2O_2 and $HNO_3/HClO_4$ methods), as seen in Figure 4.

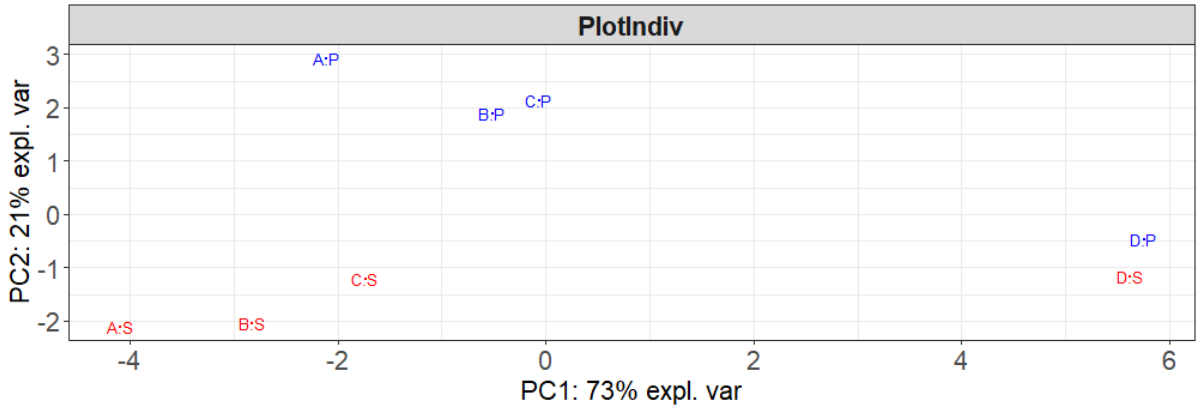

Figure 4: PCA analysis of elemental composition of S. officinalis and P. equisetiforme; AS ($HNO_3/HClO_4$ for S. officinalis), BS (HNO_3/H_2O_2 for S. officinalis), CS (dry ashing for S. Officinalis), DS (water extraction for S. officinalis, AP ($HNO_3/HClO_4$ for P. equisetiforme), BP (HNO_3/H_2O_2 for P. equisetiforme), CP (dry ashing for P. equisetiforme), DP (water extraction for P. equisetiforme).

The results of the PLS-DA analysis demonstrate a clear distinction between S. officinalis and P. equisetiforme in terms of elemental concentrations. The analysis also reveals distinct clustering for certain groups, water extraction method and other methods., as seen in Figure 5.

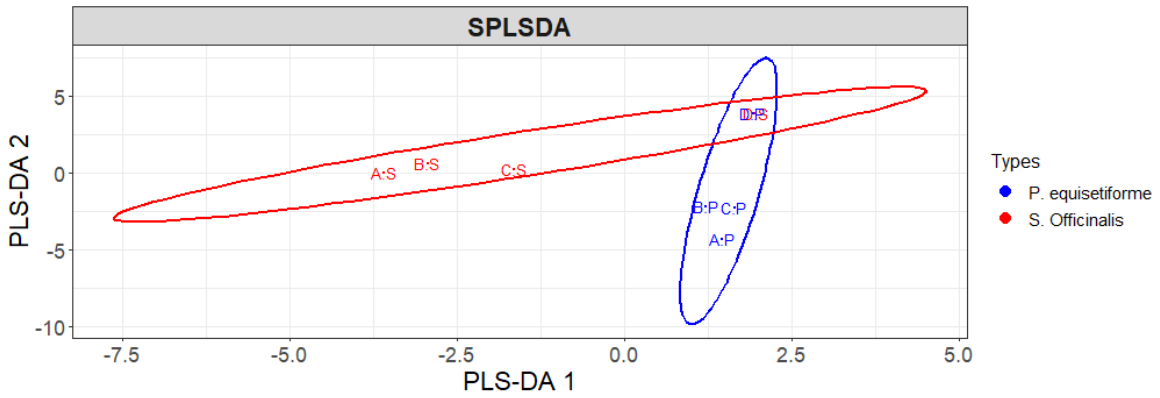

Figure 5: PLS-DA analysis of elemental composition of S. officinalis and P. equisetiforme; AS ($HNO_3/HClO_4$ for S. officinalis), BS (HNO_3/H_2O_2 for S. officinalis), CS (dry ashing for S. Officinalis), DS (water extraction for S. Officinalis, AP ($HNO_3/HClO_4$ for P. equisetiforme), BP (HNO_3/H_2O_2 for P. equisetiforme), CP (dry ashing for P. equisetiforme), DP (water extraction for P. equisetiforme).

The cluster dendrogram plots, as an example shown in Figure 6, reveal clear separation between water extraction method and other digestion methods. The analysis groups similar methods

together, indicating that these methods have a similar ability to extract the elements from the plants.

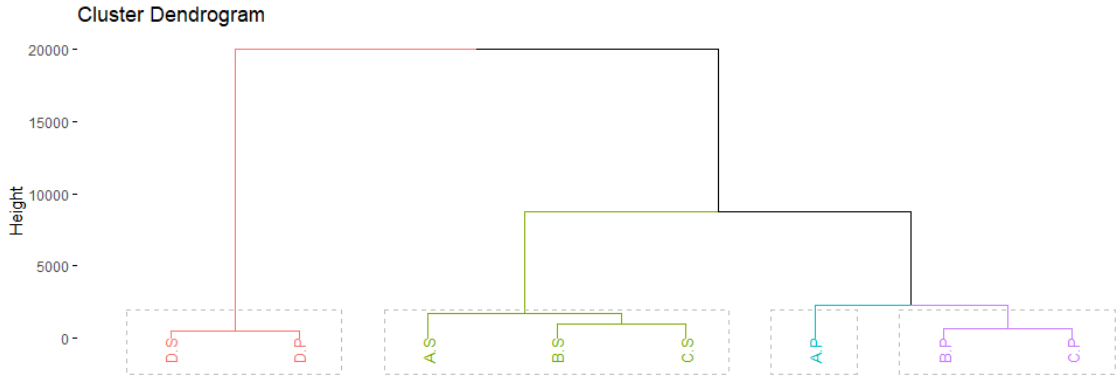

Figure 6: Dendrogram plots showing separation of digestion methods that used to extract elements from S. officinalis and P. equisetiforme; AS ($HNO_3/HClO_4$ for S. officinalis), BS (HNO_3/H_2O_2 for S. officinalis), CS (dry ashing for S. officinalis), DS (water extraction for S. officinalis, AP ($HNO_3/HClO_4$ for P. equisetiforme), BP (HNO_3/H_2O_2 for P. equisetiforme), CP (dry ashing for P. equisetiforme), DP (water extraction for P. equisetiforme).

Statistical analysis, Boxplot graph, also show there is significant differences in the selected medical plants in term of elemental concentration. For instance, the plants have a significant difference in concentration of manganese, coper and nickel. This may relate to the different ability of the plants to absorb these metals. While there are no significant differences between these plants according to the concentration of Cr and Sn, as seen this example, Figure 7.

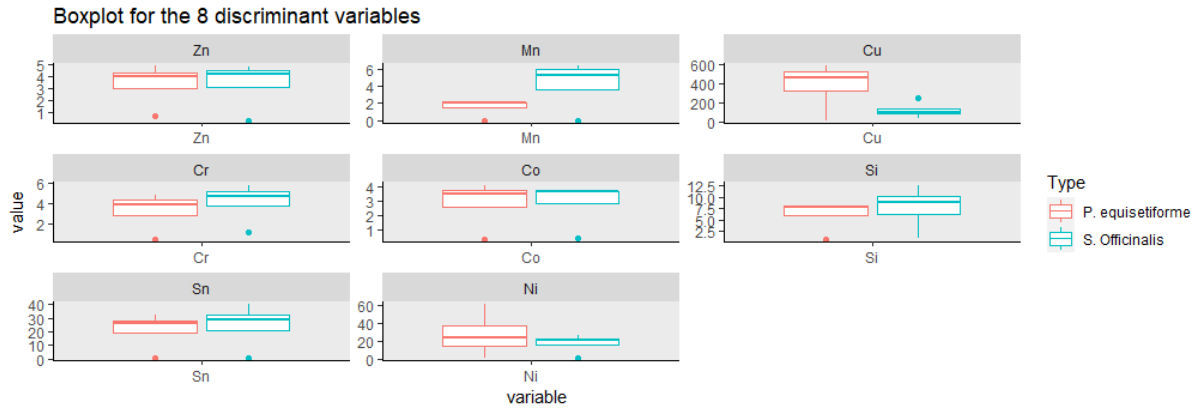

Figure 7: Grouped boxplots showing the possible elements that considered as marker to distinguish between S. officinalis and P. equisetiforme.

Conclusion

Macro, micro and some of toxic element's concentrations were estimated in S. officinalis and P. equisetiforme. Based on the above data, it can be concluded that the concentration levels for the most measured elements in these plants sticked to the following trend: Ca >> Mg > K > Si > P > Al > Na > Fe > Mn > Zn > Cu > Co > Cd > Pb > Ni > Cr > Sn > Hg.

Based on our arbitrary division, the macronutrients calcium was found at the major level, whereas the macronutrients magnesium, potassium, sodium, phosphorus, iron and micronutrient silicon, along to the toxic trace element aluminum, were present at a `minor level. The micronutrients zinc, manganese, copper, chromium, tin, nickel and cobalt, as well as the toxic element lead, were found at trace levels.

The data also indicate that, there is significant amounts of micronutrients exist in the water extract of these plants. Therefore, this water extract could contribute to the daily allowance recommended by WHO.

In general, the statistical analysis indicated the clear superiority of ($HNO_3/HClO_4$) as wet digestion mixture over the other discussed procedures to destroy the complex organic matrix of the studied plant tissues to extract most of the elements. However, the results also indicated that the extraction efficiency of the (HNO_3/H_2O_2) mixture is comparable to that of dry ashing for most elements. Statistical analysis also demonstrated that there are significant differences in the herbal plants in terms of elemental concentrations. For instance, the plants have a significant difference in concentration of manganese, coper and nickel. While there are no significant differences between these herbals according to the concentrations of chrome and tin.

References:

1 Silagy, C.A. and Neil, H.A.W.J.J.o.h., *A meta-analysis of the effect of garlic on blood pressure.* 1994. **12**(4): p. 463-468.

2 Mukhtar, H. and Ahmad, N., *Green tea in chemoprevention of cancer.* Toxicological sciences : an official journal of the Society of Toxicology, 2000. **52**: p. 111-7.

3 Auer, W., Eiber, A., Hertkorn, E., Hoehfeld, E., Koehrle, U., Lorenz, A., Mader, F., Merx, W., Otto, G., and Schmid-Otto, B.J.B.j.o.c.p.S., *Hypertension and hyperlipidaemia: garlic helps in mild cases.* 1990. **69**: p. 3.6-

4 Belman, S., *Onion and garlic oils inhibit tumor promotion.* Carcinogenesis, 1983. **4**(8): p. 1063-1065.

5 Sahito, S., MEMON, A., KAZI, T., KAZI, G., PIRZADO, Z., and SHAR, G.J.J.o.T.C.S.o.P., *Comparison of Wet Ashing Methods for Medicinal Plants, Celosia argentea and Cubeba officinalis by Atomic Absorption Spectrophotometer.* 2011. **26**(4): p. 135.

6 Elless, M.P., Blaylock, M.J., Huang, J.W., and Gussman, C.D.J.F.c., *Plants as a natural source of concentrated mineral nutritional supplements.* 2000. **71**(2 :(p. 181-188.

7 Calliari, I., Caniglia, G., Nardi, S., Tollardo, A., and Callegaro, R.J.X.R.S., *EDXRS study of lichens as biomonitors and effects of washing procedure on element concentrations.* 1995. **24**(3): p. 143-146.

8 Carvalho, M.L., Ferreira, J., G ،Amorim, P., Marques, M.I., Ramos, M.T.J.E.T., and Chemistry, *Study of heavy metals and other elements in macrophyte algae using energy-dispersive x-ray fluorescence.* 1997. **16**(4): p. 807-812.

9 Racz, L., Bumbalova, A., Harangozo, M., Tölgyessy, J., Tomeček, O.J.J.o.R., and Chemistry, N., *Determination of cesium and selenium in cultivated mushrooms using radionuclide X-ray fluorescence technique.* 2000. **245**: p. 611-614.

10 Richardson, D., Shore, M., Hartree, R., and Richardson, R.J.S.o.t.t.e., *The use of X-ray fluorescence spectrometry for the analysis of plants, especially lichens, employed in biological monitoring.* 1995. **176**(1-3): p. 97-105.

11 Kabata-Pendias, A. and Pendias, H.J.B.R., Florida, *Trace elements in plants and soils.* 1984: p. 233-237.

12 Mengel, K. and Kirkby, E.A., *Principles of plant nutrition*. 2012: Springer Science & Business Media.

13 Ekinci, N., Ekinci, R., Polat, R., Budak, G.J.J.o.R., and Chemistry, N., *Analysis of trace elements in medicinal plants with energy dispersive X-ray fluorescence.* 2004. **260**: p. 127-131.

14 Serfor-Armah, Y., Nyarko, B., Akaho, E., Kyere, A., Osae, S., Oppong-Boachie, K., and Osae, E.J.J.R.N.C., *Activation analysis of some essential elements in five medicinal plants used in Ghana.* 2001. **250**(1): p. 173.176-

15 Ražić, S., Onjia, A., Đogo, S., Slavković, L., and Popović, A.J.T., *Determination of metal content in some herbal drugs—Empirical and chemometric approach.* 2005. **67**(1): p. 233-239.

16 Gomez, M.a.R., Cerutti, S., Olsina, R.A., Silva, M.a.F., Martínez, L.D.J.J.o.P., and Analysis, B., *Metal content monitoring in Hypericum perforatum pharmaceutical derivatives by atomic absorption and emission spectrometry.* 2004. **34**(3): p. 569-576.

17 Ražić, S., Onjia, A., Potkonjak, B.J.J.o.P., and Analysis, B ،.*Trace elements analysis of Echinacea purpurea—herbal medicinal.* 2003. **33**(4): p. 845-850.

18 Olabanji, S., Omobuwajo, O., Ceccato, D., Buoso, M., De Poli, M., Moschini, G.J.J.o.r., and chemistry, n., *Analysis of some medicinal plants in South-western Nigeria using PIXE.* 2006. **270**(3): p. 515-521.

19 Bello, M.O., Ibrahim, A.O., Ogunwande, I.A., and Olawore, N.O.J.F.C., *Heavy trace metals and macronutrients status in herbal plants of Nigeria.* 2004. **85**(1): p. 67-71.

20 Ferreira, L.D.S., Lopes, R.P., Ulbrich ،M.N.C., Guaratini, T., Colepicolo, P., Lopes, N.P., Garla, R.C., Oliveira Filho, E.C., Pohlit, A.M., and Zucchi, O.L.A.D., *Research Article Concentration of Inorganic Elements Content in Benthic Seaweeds of Fernando de Noronha Archipelago by Synchrotron Radiation Total Reflection X-Ray Fluorescence Analysis (SRTXRF).* 2012.