Research Article

Profiling of various elements in *Haloxylon griffithii* and *Convolvulus leiocalycinus* using atomic absorption spectroscopy and flame photometry

Warda Saifullah¹, Samiullah^{1*}, Naqeebullah Khan¹, Attiq-ur-Rehman¹, Pari Wash Mengal¹, Abdul Baqi^{1,2} and Abdul Manan³

1. Department of Chemistry, University of Balochistan, Quetta 87300-Pakistan

2. Colleges, Higher and Technical Education Department, Balochistan, Quetta 87300-Pakistan

3. Institute of Biochemistry, University of Balochistan, Quetta 87300-Pakistan

*Corresponding author's email: sami435889@gmail.com

Citation

Warda Saifullah, Samiullah, Naqeebullah Khan, Attiq-ur-Rehman, Pari Wash Mengal, Abdul Baqi and Abdul Manan. Profiling of various elements in *Haloxylon griffithii* and *Convolvulus leiocalycinus* using atomic absorption spectroscopy and flame photometry. Pure and Applied Biology. Vol. 8, Issue 2, pp1535-1542.

<http://dx.doi.org/10.19045/bspab.2019.80094>

Received: 05/02/2019

Revised: 11/05/2019

Accepted: 16/05/2019

Online First: 18/05/2019

Abstract

The concentration of nine different elements including heavy metals were analyzed in selected medicinal plants i.e. *Haloxylon griffithii* and *Convolvulus leiocalycinus*. The mineralization of plant samples was done using wet digestion method in which three strong acids were used i.e. nitric acid (HNO₃), sulphuric acid (H₂SO₄) and perchloric acid (HClO₄) followed by filtration and analyzed through flame atomic absorption. For sodium and potassium, flame photometer was used and samples were diluted to 100 folds for obtaining results within the range of the instrument. The decreasing order of concentration of elements in *H. griffithii* was K>Na>Fe>Ni>Mn>Cu>Co>Pb>Cd whereas, in *C. leiocalycinus* was K>Na>Fe>Cu>Mn>Ni>Co>Pb>Cd. The study revealed that heavy metals accumulate in the plants with different concentrations and their concentrations were within the permissible ranges established by international organizations. This will provide knowledge in the invention of many new drugs and the research helps the legislators to provide protection to the public regarding adverse effects of heavy metals.

Keywords: Flame atomic absorption; Flame photometer; Flame emission spectroscopy; Heavy metals; Wet digestion

Introduction

The climatic change, soil and ecological zones in Pakistan bring its flora rich in medicinal plants. Among six thousand species of flowering plants in Pakistan in which 400-600 are therapeutically active to cure various diseases [1, 2]. The climate of Balochistan makes rich in diverse species of medicinal plants [3]. According to World Health Organization (WHO), 80% of the

people use these medicinal plants as traditional drugs for curing ailments. The extract of plants primarily involved in the traditional therapies [4]. In the past few years, the consumption of herbal medicine proliferates because of its ease of use and less side effects among the third world countries. The use of herbal medicines give cure against various ailments and fulfil the need of essential and non-

essential minerals in the body [5]. Experimentally, it has been revealed that the metabolic maladies are cured by use of traditional medicinal plants. The constituents like essential oils, glycosides and vitamins are responsible for the activeness of medicinal plants but their excessive intake lead to many health problems owing to the existence of heavy metals [6]. The healing latent of curative plants relates to the trace elements existing in them. These trace elements provide cure against various ailments and disorders [7]. The presence of these trace elements in medicinal plants might be harmful in higher concentration. It is important to determine the concentration of these trace elements in medicinal plants while treating different human diseases in order to understand their different pharmacological properties [8, 9]. Metallic and non-metallic elements are needed for the human body in order to maintain good health. Subsequently, the elemental composition in edible plants and food is necessary for knowing their beneficial significance [6, 10-12]. For the endurance of human being, body systems and plants, more than 40 trace elements are vital below its certain limit [13]. In metabolism, there is a crucial role of trace elements present in the therapeutic plants [14]. These trace elements are heavy metals that are found in all kinds of biomes. Human is the main source of their existence mainly from industrial and agriculture sector [15]. The presence of heavy metal pollutes the environment of soil and water in higher concentration that affects both the flora and fauna of that environment [16]. The existence of heavy metals in higher concentration is considered as pollutants found in dietary supplements, cosmetics, herbal medicines and various saleable

products [17]. It has been reported that long term exposure towards heavy metals even in low concentration is noxious to human health [18]. Few heavy metals are considered important or essential to maintain the normal body functions of human in trace amounts and above their certain limit they might be lethal [19, 20]. Lead, mercury, cadmium and arsenic are the types of heavy metals that are fatal to human health and triggering bio accumulation in the body that leads to the disruption of main body functions [21]. The spectroscopic technique like atomic absorption is mainly used for the elemental analysis of various medicinal plants [8, 9]. In this study, different heavy metals including Fe, Mn, Cu, Co, Cd, Pb and Ni present in *H. griffithii* and *C. leiocalycinus* and their quantity in above mentioned plants were determined through Atomic Absorption Spectrophotometer. However, flame photometer was used for elements like Na and K.

H. griffithii, as shown in figure 1, belongs to the family of Chenopodiaceae, familiar with name of Cat tail Family. It covers 100 genera and 1200 species [22]. The class of Haloxylon covers 13 species growing in dry places of the North-African and Arabian deserts and South-West Asia. The species of the genus Haloxylon are shrubbery [23]. Five species of this class are found in Pakistan [24].

C. leiocalycinus, as shown in figure 2, belongs to the family of Convolvulaceae. However, Convolvulus, a genus, comprising of 200-250 species of flowering plants. *C. leiocalycinus* is found in Balochistan on open stony ground and stony slopes at the height of 1500m, also found in the hilly areas of Hanna Lake, Ziarat, Brewery close to Quetta city.

Figure 1. Shows the image of *H. griffithii*

Figure 2. Shows the image of *C. leiocalycinus*

Materials and methods

Reagents and solutions

Different standards (1000ppm) of analytical grade were purchased from Merck, Marker for the analysis of heavy metals. Concentrated acids were used like perchloric acid (HClO_4), nitric acid (HNO_3) and sulphuric acid (H_2SO_4) which are the strong oxidizing agents used in the digestion of plants and were of analytical grade.

Instrument and glasswares

Atomic absorption spectrophotometer of Thermoelectron S4 AA system was used for the determination of heavy metals with hollow cathode lamps of different metals and air acetylene gas as a flame was used in the analysis of heavy metals. Whereas, Na and K was detected through flame emission spectroscopy of Jenway PFP7 for the above mentioned plants.

Various glass wares including round bottom flask, conical flask and beakers of pyrex glass were used and rinsed with deionized water before analysis.

Sample collection

The entire plant of *H. griffithii* was collected from valley of Hanna and Spinni road of Quetta, Balochistan on May 17, 2018. The whole plant of *C. leiocalycinus* was collected from hilly regions of Hanna Lake on May 17, 2018 and recognition of both above mentioned plants was done by taxonomist Dr. Rasool Bakhsh Tareen.

Sample preparation

Crushing and grinding

The collected plants were first dried in shady area then dried plants were crushed into powder with the help of electrical blender. These powdered forms of plants were further used for the analysis through atomic absorption spectrophotometer.

Digestion method

As various methods are used for the digestion of metals from plants but here wet digestion method was used followed the protocol of Zafar *et al.* [25]. Each plant sample in powdered form 0.25g were taken in a 50ml round bottom flask then the mixture of acids with a total volume of 6.5ml i.e., 1ml conc. sulphuric acid, 5ml conc. nitric acid and 0.5ml conc. perchloric acid was added in a 50ml round bottom flask containing plant materials. Individually, each plant sample containing mixture of acids was heated on a hot plate (JENWAY 1000) at 80-85 degree Celsius until white fumes come out from the flask. These white fumes indicate that the digestion was complete. Subsequently, added few drops of distilled water in it, removed it from the hot plate and allowed it to cool then transferred the solution to 50ml volumetric flask and made the volume up to the mark i.e. 50ml by pouring distilled water in it. Filtered the extract using filter paper Whatmann No.1 in labelled plastic bottles. These prepared solutions after the

wet digestion were analyzed for the detection of elements using atomic absorption spectrophotometer of Thermoelectron S4 AA system with selective hollow cathode lamps. Dilutions of different concentrations were prepared from analytical grade stock standards of 1000ppm for the purpose of calibration. These dilutions were prepared immediately before running the samples. Deionized water was used throughout the investigation. FES of Jenway PFP7, the technique was applied for the evaluation of sodium and potassium. For obtaining results within the range of flame photometer, the samples were diluted 100 folds with deionized water.

Results and discussion

The results are given below in table 1 and their comparison is given in figure 3.

FAAS was used for the evaluation of different heavy metals because of its accuracy and FES is a simple method for the evaluation of sodium and potassium. For sodium and potassium, the dilution of samples was done using deionized water considering the range of calibration curves of different elements. As flame atomic absorption was attached with the computer device and it gives calculated concentration of various elements upon detection whereas flame photometer was not able to give the calculated concentrations so, the concentration of sodium and potassium were obtained through the regression equation.

Satisfactory results were obtained through the wet digestion method for the mineralization of plant materials. This method eradicates the surrounding of organic compounds around the minerals, only the minerals are left behind in an aqueous solution by using the strong oxidizing agents. This method is simple and faster than other methods of digestion like dry ashing and microwave digestion.

Table 1. Concentration of various elements in selected medicinal plants (µg/ml)

No.	Species Name	Cu	Co	Ni	Fe	Mn	Cd	Pb	Na	K
1	<i>H. griffithii</i>	0.2137	0.0657	0.7544	4.8342	0.5174	0.0126	0.0568	819.83	836.67
2	<i>C. leiocalycinus</i>	0.8492	0.0838	0.1871	5.6601	0.2294	0.0139	0.0309	423.83	504.67

Figure 3. Comparison of elements among two species of medicinal plants

Copper helps in the development and growth of plants [26]. Chest, wounds inflammation and arthritis diseases are mainly cured through the presence of copper in plants [27, 28]. In addition, 10mg/kg is the permissible limit of copper in plants that are used therapeutically and 2-3mg/day for human consumption [29]. The mean concentration of Cu in *H. griffithii* and *C. leiocalycinus* was 0.2137µg/ml and 0.8492µg/ml respectively.

Cobalt in a very minute amount is required for all mammals. Anemic patients are treated with cobalt and various cancers are cured with the small quantity of cobalt. The excessive intake of cobalt leads to many cardiovascular problems [27, 28]. Cobalt contains an important component i.e. vitamin B-12 [30]. The acceptable limit of cobalt for human consumption is 1.6-8µg/kg body weight per day [31]. The mean concentration of Cobalt in *H. griffithii* and *C. leiocalycinus* were 0.0657µg/ml and 0.0838µg/ml respectively.

Nickel is essential for human body as well as for plants in a very less amount as it

takes part in the production of insulin and its deficiency leads to liver malfunction. In high concentration, its toxicity leads to many diseases including loss of body weight, cardiac and liver problems. According to WHO, the permissible limit of Ni is 1.5mg/kg in the plants and for dietary intake it is 1mg/day [29]. The mean concentration of Cobalt in *H. griffithii* and *C. leiocalycinus* were 0.7544µg/ml and 0.1871µg/ml respectively.

Iron is a vital element for the production of red blood cells in the body. Low concentration of iron in the body leads to anemia [32]. The high concentration of iron cause damage to the tissues in humans [33]. In plants, the acceptable limit of iron according to WHO is 20mg/kg while for human consumption it is 10 to 28mg/day [29]. The mean concentration of iron in *H. griffithii* and *C. leiocalycinus* were 4.8342µg/ml and 5.6601µg/ml respectively.

Manganese is a very vital element for the growth of both plants and animals. In mammals the reproductive and skeletal problems are due to the deficiency of this trace metal in the body. Excessive intake

of manganese leads to many lung problems and affects the human brain [34]. The permissible limit of manganese according to WHO in plants is 200mg/kg and for human intake it is 11mg [29]. The mean concentration of iron in *H. griffithii* and *C. leiocalycinus* were 0.5174µg/ml and 0.2294µg/ml respectively.

Cadmium is noxious for human health even in the low concentration, as it is a non-essential heavy metal. Learning debilities and hyperactivity in progenies all due to cadmium [35]. According to WHO, the permissible limit of cadmium in therapeutic plants is 0.3mg/kg [29]. The average concentration of cadmium in *H. griffithii* and *C. leiocalycinus* were 0.0126µg/ml and 0.0139µg/ml respectively.

Lead is a non-essential heavy metal. It is accumulated in the bones and teeth which causes the bones to stiff and weakens the wrists and limbs. It affects the reproductive, renal, immune and nervous system of the body through the deposition of lead in the soft tissues. The safety limit of lead for human consumption is 1.5 ppm [32, 36] and 10mg/kg in medicinal plants [29]. The average concentration of lead in *H. griffithii* and *C. leiocalycinus* were 0.0568µg/ml and 0.0309µg/ml respectively.

Sodium is a transporter of amino acids, glucose and other metabolites in the cells of the body which in results produces energy. The deficiency of sodium in the body leads to mood swings, causing cramps in the muscles, dehydration in the body and hair loss. The recommended value of sodium for human intake is 13.8mg/day [37]. The average concentration of lead in *H. griffithii* and *C. leiocalycinus* was 819.83µg/ml and 423.83µg/ml respectively.

Potassium maintains the balance of hormones, secretion of insulin and response of immune system. Potassium works to reduce the blood pressure, maintains the cell volume and also the cell signaling of the body. It helps in the treatment of liver and kidney problems.

For adult women, the permissible limit of potassium is 2300mg/day and for adult men it is 3100mg/day [38]. The average concentration of lead in *H. griffithii* and *C. leiocalycinus* were 836.67µg/ml and 504.67µg/ml respectively.

Conclusion

This study revealed that the concentration of heavy metals in plants i.e. *H. griffithii* and *C. leiocalycinus* were below their safety limits given by different international organizations like WHO and FAO. Nine elements including essential and non-essential elements were evaluated in these plants among which the concentration of sodium and potassium were found to be higher in both plants and concentration of heavy metals were within the permissible ranges. These plants are safe to use as traditional medicines and therefore are not at risk. The accurate and precise evaluation of metals are very necessary. The results obtained in this investigation will be supportive for the manufacturers in the synthesis of new drugs with different combination of medicinal plants for curing various ailments.

Authors' contributions

Conceived and designed the experiments: W Saifullah, Samiullah & AU Rehman, Performed the experiments: W Saifullah, Samiullah & A Baqi, Analyzed the data: Samiullah, N Khan, AU Rehman, Contributed materials/ analysis/ tools: A Manan, N Khan, Wrote the paper: W Saifullah, A Baqi & P Mengal.

References

1. Nasir E& Ali SI (1971). Flora of West Pakistan. Islamabad, Pakistan. *Agric Research Council*, pp 1-190.
2. Hamayun M (2005). Ethnobotanical profile of Utror and Gabral valleys, district Swat, Pakistan. *Ethnobotanical Leaflets* (1): 9.
3. Hussain SZ& Sheikh SA (2008). Ethno-medicinal survey of plants from salt range of Pakistan. *Pak J Bot* 40(3): 1005-1011.

4. Farnsworth NR, Akerele O, Bingel AS, Soejarto DD & Guo Z (1985). Medicinal plants in therapy. *Bull World Health Organ* 63(6): 965.
5. Bello MO, Ibrahim AO, Ogunwande IA & Olawore NO (2004). Heavy trace metals and macronutrients status in herbal plants of Nigeria. *Food Chem* 85(1): 67-71.
6. Jabeen S, Shah MT, Khan S & Hayat MQ (2010). Determination of major and trace elements in ten important folk therapeutic plants of Haripur basin, Pakistan. *J of Medi Plants Resm* 4(7): 559-566.
7. Shirin K, Imad S, Shafiq S & Fatima K (2010). Determination of major and trace elements in the indigenous medicinal plant *Withaniasomnifera* and their possible correlation with therapeutic activity. *J of Saudi Chem Soc* 14(1): 97-100.
8. Lozak A, Soltyk K, Ostapczuk P & Fijalek Z (2002). Determination of selected trace elements in herbs and their infusions. *Sci of the Total Environment* 289(1-3): 33-40.
9. Ning P, Gong C, Zhang Y, Guo K & Bai J (2011). Lead, cadmium, arsenic, mercury and copper levels in Chinese Yunnan Pu'er tea. *Food Addit and Contaminants* 4(1): 28-33.
10. Khan SA, Khan L, Hussain I, Marwat KB & Akhtar N (2008). Profile of heavy metals in selected medicinal plants. *PakJ of Weed Sci Res* 14(1-2): 101-110.
11. Sharma RK, Agrawal M & Marshall FM (2009). Heavy metals in vegetables collected from production and market sites of a tropical urban area of India. *Food and Chem Toxicol* 47(3): 583-591.
12. World Health Organization. (1998). Quality control methods for medicinal plant materials.
13. Osae E (2001). Activation analysis of some essential elements in five medicinal plants used in Ghana. *J of Radioanalytical and Nuclear Chem* 250(1): 173-176.
14. Rajurkar NS & Damame MM (1997). Elemental analysis of some herbal plants used in the treatment of cardiovascular diseases by NAA and AAS. *J of Radioanalytical and Nuclear Chem* 219(1): 77-80.
15. Lokhande RS, Singare PU & Pimple DS (2011). Toxicity study of heavy metals pollutants in waste water effluent samples collected from Taloja industrial estate of Mumbai, India. *Resources and Environ* 1(1): 13-19.
16. Leung A, Cai ZW & Wong MH (2006). Environmental contamination from electronic waste recycling at Guiyu, southeast China. *J of Material Cycles and Waste Management* 8(1): 21-33.
17. Neustadt J & Pieczenik S (2007). Research Review: Heavy-Metal Toxicity-With Emphasis on Mercury. *IntegMedInno Comm* 6(2): 26.
18. Ernst E (2002). Toxic heavy metals and undeclared drugs in Asian herbal medicines. *J Pharma Sci* 2 3(3): 136-139.
19. Florea AM & Busselberg D (2006). Occurrence, use and potential toxic effects of metals and metal compounds. *Biometals* 19(4): 419-427.
20. Plum LM, Rink L & Haase H (2010). The essential toxin: impact of zinc on human health. *Int J Enviro Res Pub Hea* 7(4): 1342-1365.
21. Ray SA & Ray MK (2009). Bioremediation of heavy metal toxicity-with special reference to chromium. *Al Ameen J Med Sci* 2(2): 57-63.
22. Ali SI & Qaiser M (2001). Flora of Pakistan, Chenopodiaceae Jointly published by Department of Botany, University of Karachi, Karachi, Pakistan & Missouri Botanical Press: Missouri Botanical Garden, St. Louis, Missouri, USA 204: 185.
23. Baqi A, Samiullah, Tareen RB, Mengal A, Khan N, Behlil F, Achakzai AKK & Faheem M (2018). Determination of

- antioxidants in two medicinally important plants, *Haloxylon griffithii* and *Convolvulus leiocalycinus*, of Balochistan. *P & Appl Bio* 7(1): 296-308.
24. Mhaskar KS, Blatter E & Caius JF (2000). Indian Medicinal plants published by Indian Books centre, Delhi, India I V: 1212-1214.
 25. Zafar M, Khan MA, Ahmad M, Jan G, Sultana S, Ullah K & Abbasi AM (2010). Elemental analysis of some medicinal plants used in traditional medicine by atomic absorption spectrophotometer (AAS). *J Med Plants Res* 4(19): 1987-1990.
 26. Diaconu D, Diaconu R & Navrotescu T (2012). Estimation of heavy metals in medicinal plants and their infusions. *Analele Universitatii " Ovidius" Constanta-Seria Chimie* 23(1): 115-120.
 27. Khan KY, Khan MA, Niamat R, Munir M, Mazari HFP, Seema N & Ahmed SN (2011). Element content analysis of plants of genus *Ficus* using atomic absorption spectrometer. *AfriJPhar Pharma* 5(3): 317-321.
 28. Obiajunwa EI, Adebajo AC & Omobuwajo OR (2002). Essential and trace element contents of some Nigerian medicinal plants. *J RadioanaNuc Chem* 252(3): 473-476.
 29. Shah A, Niaz A, Ullah N, Rehman A, Akhlaq M, Zakir M & Suleman Khan M (2013). Comparative study of heavy metals in soil and selected medicinal plants. *J Chem* 2013: 1-5.
 30. Imray P & Langley A (2001). Health-based soil investigation levels. National Environmental Health Forum.
 31. Arnich N, Sirot V, Riviere G, Jean J, Noel L, Guerin T & Leblanc JC (2012). Dietary exposure to trace elements and health risk assessment in the 2nd French Total Diet Study. *Food & Chem Toxicol* 50(7): 2432-2449.
 32. Agrawal J (2011). Determination of heavy metal contents in samples of different medicinal plants: 1126-1132.
 33. Fuortes L & Schenck D (2000). Marked elevation of urinary zinc levels and pleural-friction rub in metal fume fever. *Veter & Hum Toxicol* 42(3): 164-165.
 34. Jarup L (2003). Hazards of heavy metal contamination. *Brit Med Bull* 68(1): 167-182.
 35. Hunt JR (2003). Bioavailability of iron, zinc, and other trace minerals from vegetarian diets. *The Am J Clin Nutr* 78(3): 6335-6395.
 36. Todd GC (1996). Vegetables grown in mine wastes. *Environ Toxicol & Chem* 19(3): 600-607.
 37. Harper ME, Willis JS & Patrick J (1997). Sodium and chloride in nutrition. Handbook of nutritionally essential mineral elements. (Eds BL O'Dell, RA Sunde), pp 93-116.
 38. Fernandes JC & Henriques FS (1991). Biochemical, physiological, and structural effects of excess copper in plants. *The Botan Rev* 57(3): 246-2.