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ELEMENTAL INVESTIGATION OF MOMORDICA CHARANTIA LINN. AND SYZIGIUM JAMBOLANA LINN. USING ATOMIC ABSORPTION SPECTROPHOTOMETER

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Elemental investigation of very important medicinal plant i.e. m*omordica charantia* linn and s*yzigium jambolana* linn, and its decoction has been carried out using flame atomic absorption spectrophotometer. In present study fifteen essential, trace and toxic elements such as Zn, Cr, K, Mg, Ca, Na, Cu, Fe, Pb, Al, Ba, Mn, Co, Ni and Cd were determined in different parts of both plants and in its decoction. The level of essential elements was found high as compared to the level of toxic elements. Both plants are useful in the treatment of diabetes. The validation of the method was checked by employing NBS- 1570 (Spanich) as a standard reference material . The measured values of elements are in close agreement with certified values.

Keywords : Momordica charantia linn, Syzigium jambolana linn, Medicinal plants, Atomic absorption spectrometry, Elemental analysis.

1. Introduction

Medicinal plants have long been the subject of human curiosity and need. Plants have been used as medicine since man's existence on the earth [1-5]. Herbal medicines have usually been in the form of fruit and vegetable; drugs are their extracts for the treatment of the disease and maintaining of improved health.

Momordica charantia linn belongs to family Cucurbitaceae, commonly called bitter gourd or karela. It has many pharmacological actions such as stomachic, carminative, antithelmintic, hypoglycemic, hypocholestermic activity. The fruit and seed of *M. charantia* yields a polypeptide viz. p-insulin, which is considered to be similar to bovine insulin, it rapidly decreases and normalizes the blood sugar level [6-8]

The hypoglycemic property of bitter gourd fruit juice is superior to synthetic drugs, as it appears to regulate the blood sugar toward the normal limit.

Syzigium jambolana belongs to family Myrtaceae commonly called Jaman. The fruit and bark is astringent, carminative and useful in dysentery. The dry powdered seeds are considered to be useful in the treatment of diabetes.

1.1. Plant samples

Five to ten samples of momordica charantia linn. and syzigium jambolana linn. were collected from different areas of Hyderabad city and Sindh University Jamshoro campus. Reference vouchers specimens were grown in the plot of centre of Excellence in Analytical Chemistry, University of Sindh, Jamshoro and Botany Department of Sindh University. The reference samples were identified by Botany Department, Sindh University Jamshoro, Pakistan. These reference samples were also analysed separately are and reported in Tables 1 and 2.

1.2. Reagents and apparatus

All the reagents and chemicals used were of AR grade. All the solutions of standards and samples were prepared in deionized water. Mineral elemental analysis of all parts of samples and reference materials was carried out by Atomic

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Table 1. Determination of metals in Momordica Charantia Linn family Cucurbitaceae by atomic absorption spectrophotometer (mg/100 g driy weight basis).

Elements	Seed	Reference Seed	Fruit	Ref. Fruit
Sodium	1151.8 – 1227.58 (127.55 – 178.07)*	1123.56 (165.8)	444.56 – 659.26 (102.29 – 127.55)	580.85# (120.65)
Calcium	818.97 – 1108.32 (337.64 – 409.48)	987.6 (406.8)	962.64 – 1250.00 (563.94 – 599.86)	1120.5 (432.65)
Potassium	1448.8– 1842.51 (1059.0 – 1344.5)	1658.7 (1123.9)	1547.24 – 2570.8 (1073.8 – 1816.9)	2187.5 (1476.8)
Magnesium	996.75-1010.03 (228.72 – 249.97)	988.7 (248.85)	797.56 – 882.55 (255.28 – 282.90)	668.45 (216.68)
Manganese	1.90 – 2.69 (0.951 – 1.33)	2.34 (1.16)	1.58 – 3.32 (0.443 – 0.570)	2.89 (0.498)
Zinc	1.65– 4.14 (2.98 – 3.75)	3.18 (2.98)	4.14 – 5.71 (1.90 – 2.32)	5.87 (2.14)
Iron	18.72 – 24.57 (4.36 – 6.08)	20.56 (5.45)	57.11– 70.20 (5.46 – 7.64)	56.86 (6.58)
Nickel	1.65– 0.630 (0.096 – 0.118)	0.562 (0.12)	0.463 – 0.519 (0.118 – 0.129)	0.468 (0.186)
Cobalt	0.159 – 0.263 (0.105 – 0.161)	0.287 (0.153)	0.194 – 0.264 (0.063 – 0.119)	0.312 (0.108)
Chromium	0.165– 0.235 (0.077 – 0.110)	0.245 (0.12)	0.187– 0.319 (0.094 – 0.127)	0.285 (0.154)
Lead	0.127 – 0.188 (0.063 – 0.075)	0.187 (0.085)	0.127 – 0.188 (0.063 – 0.087)	0.214 (0.094)
Copper	1.03 – 1.47 (0.7556 – 0.9109)	1.54 (0.895)	0.802 – 1.061 (0.248 – 0.383	1.24 (0.412)
Cadmium	0.0756 – 0.0897 (0.0302 – 0.0472)	0.106 (0.0386)	0.0756 – 0.104 (0.0302 – 0.0415)	0.142) (0.0654)
Aluminum	5.54 – 7.34 (1.76 – 2.58)	6.85 (1.98)	2.93 – 3.81 (1.41 – 1.88)	3.12 (1.65)
Barium	2.45 – 3.12 (0.95 –1.25)	3.28 (1.325)	2.3 – 2.95 (1.15 – 1.94)	2.76 (1.27)

Values of reference samples, *The values of metals in Decoction (water extractable elements)

Table 2a. Determination of metals in syzigium jambolana, linn (cuminii) family myrtaceae by atomic absorption spectrophotometer *(mg/100g dried basis).*

Elements	Leaves	Reference# Leaves	Stem	Reference stem
Sodium	368.8 – 482.45 (77.03 – 95.98)*	512.78 (96.5)	803.5 – 935.8 (58.2 – 89.66)	948.87# (68.56)
Calcium	1074.71 – 1649.4 (481.3 – 564.5)	1187.8 (498.5)	2568 – 3545.9 (621.8 – 779.4)	2486.7 (756.47)
Magnesium	756.46 – 943.6 (425 – 495.8)	865.65 (380.48)	685.6 – 726.8 (425 – 451.6)	658.85 (456.78)
Potassium	605.42 – 881.9) (444.88 – 641.73)	7658.5 (750.65)	411.4 – 569.9 (104.3 – 124.0)	485.68 (134.65)
Manganese	3.96 – 5.86 (1.07 – 1.52)	5.98 (2.15)	2.85 – 3.48 (1.078 – 1.21)	2.86 (1.45)
Zinc	1.95 – 2.65 (1.30 – 1.69)	1.87 (1.56)	0.996 – 1.520 (0.782 – 0.957)	1.65 (0.873)
Iron	26.5 – 30.81 (3.90 – 6.08)	32.75 (5.85)	5.85 – 13.65 (3.74 – 4.21)	14.25 (4.87)
Nickel	0.379 – 0.463 (0.118 – 0.18)	0.547 (0.211)	0.35 – 0.435 (0.16 – 0.196)	0.532 (0.216)
Cobalt	0.194 – 0.403 (0.1333 – 0.161)	0.476 (0.218)	0.298 – 0.507 (0.105 – 0.091)	(0.613 (0.105)
Chromium	0.1521 – 0.2354 (0.077 – 0.1108)	0.286 (0.165)	0.152 – 0.2354 (0.062 – 0.127)	0.267 (0.173)
Lead	0.127 – 0.249 (0.051 – 0.075)	0.217 (0.081)	0.157 – 0.249 (0.051 – 0.075)	0.265 (0.082)
Copper	0.155 – 0.311 (0.518 – 0.611)	0.325 (0.487)	0.259 – 0.336 (0.465 – 0.611)	0.412 (0.0587)
Cadmium	0.3018 – 0.188 (0.030 – 0.047)	0.214 (0.065)	0.104 – 0.118 (0.024 – 0.035)	0.214 (0.041)
Aluminum	2.93 – 7.93 (1.4 – 2.7)	5.45 (1.65)	6.17 – 9.73 (1.6 – 1.95)	7.35 (2.25)
Barium	1.85 – 2.75 (0.93 – 1.65)	2.94 (1.18)	2.85 – 3.45 (1.00 – 1.85)	2.89 (1.25)

Key: # *Values of reference samples, *The values in Decoction (water extractable elements)*

Table 2b. Determination of metals in syzigium jambolana, linn (cuminii family Myrtaceae byatomic absorption spectrophotometer (mg/100g dried basis)

Elements	Fruit	Reference # Fruit	Seed	Reference seed
Sodium	848.7-1050.7 (588.5 - 707.7)*	987.65 (678.78)	550.64-607.5 (519.0-588.5)	625.58# (548.85)
Calcium	500- 1132.2 (523.6-696.8)	876.65 (543.65)	1132.1-1477.5 (553.1-984.2)	1387.45 (674.65)
Magnesium	909.9- 978.9 (329.5 – 398.7)	895.67 (345.56)	373.4 – 628.3 (221 – 333.9)	467.68 (245.76)
Potassium	1442 -1787.8 (811.5-854.3)	1643.76 (823.56)	1635.-1832.7 (719.5-753.9)	1587.65 (763.86)
Manganese	1.42 –2.53 (0.697- 1.01)	2.65 (1.28)	0.951 – 1.58 (0.507-1.204)	2.15 (1.32)
Zinc	1.12 - 3.355 (0.608-0.713)	3.45 (0.945)	4.753-5.54 (0.433-0.503)	5.76 (0.621)
Iron	17.55-41.34 (6.55-7.33)	34.56 (5.68)	8.19-9.36 (6.08-6.86)	10.26 (5.89)
Nickel	0.295 – 0.35 (0.118-0.151)	0.246 (0.132)	0.293 – 0.352 (0.140-0.151)	0.416 (0.138)
Cobalt	0.298 -0.6111 (0.077-0.119)	0.712 (0.211)	0.298-0.4027 (0.1055-0.091)	0.389 (0.116)
Chromium	0.1521-0.277 (0.060-0.094)	0.310 (0.107)	0.1521-0.193 (0.060-0.094)	0.208 (0.098)
Lead	0.249-0.3712 (0.075-0.099)	0.286 (0.126)	0.157-0.4017 (0.075-0.111)	0.517 (0.172)
Copper	0.440-0.621 (0.113-0.134)	0.713 (0.142)	0.362-0.491 (0.165-0.196)	0.532 (0.237)
Cadmium	0.0897-0.118 (0.035-0.047)	0.104 (0.0514)	0.075-0.1039 (0.047-0.069)	0.126 (0.085)
Aluminum	4.40 – 7.1 (1.17 – 2.23)	6.12 (2.46)	2.93 – 3.81 (1.64 – 2.23)	3.87 (2.06)
Barium	1.80-2.45 (0.85-1.12)	1.89 (0.945)	2.48 – 3.75 (1.05-1.85)	2.98 (1.98)

Key: # *Values of reference samples, *The values in Decoction (water extractable elements)*

Absorption Spectrophotometer, Hitachi model 180-50 equipped with Zeeman background correction using flame absorption mode.

2. Experimental

The different parts of plants were washed with distilled water and dried at 120 °C in an electric oven to a constant weight. The dried plant material was then ground to powder.

Later, each part of sample plants, reference samples and certified sample was weighed into separate flasks and treated with 5 ml nitric acid, side by side 5 ml nitric acid was also added in an empty flask which served as a blank [9-12]. The flasks were covered with watch glasses and heated to reflux on an electric hot plate at 80^{o} to 100 °C. After heating for an hour, the contents of flasks were treated with additional 5 ml of nitric acid, followed by 2 ml of 30% hydrogen peroxide, and the heating at gentle reflux was continued for another hour. The watch glasses were removed from the top of flasks, and heating was continued until the volume of contents was reduced to semidried mass, the contents of the flasks were cooled, diluted appropriately with 2N HNO_3 and filtered through Whatman # 42 paper into volumetric flasks marked as stock sample solutions.

2.1. Preparation of decoction

Each dried part of both plants and reference samples were boiled with deionized water for an hour on an electric hot plate. After cooling, filter through Whatman # 42 and keep it as a stock sample solution.

The aqueous extract gave +ve test for the presence of glycosides, saponin and sugar only which are water soluble, other water insoluble organic compounds were absent.

2.2. Determination of mineral elements

Working standard solutions of aluminum, calcium, cadmium, cobalt, chromium, copper, silver, iron, lead manganese, magnesium, nickel, potassium, sodium, and zinc were prepared from stock standard solution (1000 ppm), in 2N nitric acid and calibration curves were drawn for each element using atomic absorption spectrophotometer. The calibration curves obtained for concentration vs. absorbance data were statistically analyzed using least square method. These fifteen elements were determined in both medicinal plants. The results of these measurements are presented in Tables 1 ,2a and 2b.

A blank reading was also taken and necessary correction was made during the calculation of percentage concentration of various elements.

Employing the above mentioned experimental condition the NBS- 1570 Spanich was analyzed to check the accuracy of the procedure. The result are given in Table 3 which are fairly in good agreement with the reference values.

Table 3. Analysis of metals in standard reference materials NBS- 1570A (Spinach) mg/100 g.

Elements	Reference value	Our value
Na	----	608 ±2.8
K	3560 ± 5	3568 ± 12
Ca	-----	678± 5.6
Mg	------	145.0 ± 1.8
Fe	55 ± 2	53 ± 3.5
Zn	5.0 ± 0.2	4.8 ± 0.28
Mn	16.5 ± 0.6	16.6 ± 0.7
Co	0.15	0.47 ± 0.05
Cr	0.46	0.44 ± 0.05
Cu	1.2 ± 0.2	1.22 ± 0.3
Ni	0.6	0.62 ± 0.04
Pb	0.12 ± 0.02	0.12 ± 0.025
Al	87.0±2	86.5±3.5
Ba	----	2.56 ±0.56
Cd	0.15	0.148 ± 0.004

2.3. Percentage recovery test

The efficiency of extraction method was checked by standard addition method. The duplicate sample of each part of both plants spiked with known amount of metal standard (Fluka Kamica) prior to digestion. The matrix of standards and sample solutions was same by using 2N HNO_3. The percentage recovery test for different elements was 99 % in range of 98.5-99%.

3. Results and Discussion

The role of inorganic elements in human health and diseases is now an established fact [13-17]. Our goal is to confirm whether the elements occurring in a plant are responsible for the therapeutic action or it is due to some organic

matter. If it is due to certain elements, it can prevent the disease by giving elements in other form also.

Boiling in water, which is the aqueous, extract containing some water-soluble organic and mostly inorganic compounds. Mostly Unani crude drugs are used by boiling in water and used water extract.

Analysis of total elements and in decoction of different parts of both plants has shown that many essential and important elements are present in considerable amount e.g. calcium, iron, potassium and zinc etc.

It was observed that the level of toxic metals is very low in both plants. Essential elements Mg, Ca, K, Zn, Fe, Cr and Co are present in both plants and in their aqueous extracts, which may be directly or indirectly helpful in the management of many diseases.

Both plants are used as a remedy in diabetic. The considerable amount of chromium and zinc was present in total as well as in aqueous extract of both plants. Chromium is essential for normal glucose tolerance and carbohydrate metabolism. The daily recommendation of Cr for normal person is 50 to 80 μg/day. In both plants a good amount of Cr is present in total as well as in available form i.e. in Decoction. So the Cr present in biological form is more potent than simple Cr salts. Insulin is known to contain Zn and it is, therefore, of interest that the pancreas of diabetic contain only about one half the normal amount of zinc [18]. The zinc in the fruit juice of bitter gourd was found to be a potent scavenger of super oxide and hydroxyl radicals. These oxygen radicals are implicated in diabetes; the reported antidiabetic activity of the fruit juice may be mediated through these mechanisms.

There are numerous Zn containing enzymes, which help in synthesis and degradation of proteins, carbohydrates and lipids.

Self medication with herbal decoctions like joshanda formulation should be considered of safer as compared to attempt to self medication with modern remedies e.g. antiallergies, analgesic, sedative and antibiotic.

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