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Determination of Mineral Elements in Soil, Citrus Limunum Juice and Citrus Limetta Juice Samples by Flame Photometry and Atomic Absorption Spectrometry Methods

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ABSTRACT

A number of researches have been performed in the field of elemental analysis of fruits from different areas of Iran and there is a vast unstudied area in this field. In this study, seven mineral elements such as Na, K, Ca, Mg, Fe, Zn and Cu in the tree soil, citrus limunum juice of Gorgan and Iranshahr gardens and citrus limetta juice of Gorgan and Bandarabas gardens were determined by flame photometry and atomic absorption spectrometry methods using standard addition technique. The results showed that the concentration of K and Cu elements in the samples of fruits was higher and lower than other elements, respectively. The concentration of Na and K elements in the citrus limetta sample of Bandarabas was more than that of the Gorgan sample. The concentration of Fe and Mg elements in the citrus limunum sample of Gorgan was more than that of the Iranshahr sample. The measurments showed that the concentration of Ca and Zn elements in the tree soil samples of citrus limunum, was higher and lower than the other elements, respectively. Moreover, although the concentration of the Ca element in the soil sample of Gorgan was more than that of Iranshahr, the concentration of this element in the citrus limunum of Gorgan was less than that of Iranshahr, this is probably due to problems in the Ca absorption of the citrus limunum plant of Gorgan. The obtained information in this study can be useful for some patients in the preferred consumption of these kinds of fruits. In addition, the used methods are reliable and simple analytical procedures for the mineral element characterisation of citrus limunum and citrus limetta fruits.

KEYWORDS: Citrus Limunum; Citrus Limetta; Soil; Element; Atomic Absorption Spectrometry; Flame Photometry.

1. INTRODUCTION

Fruit and fruit products are important source of minerals. In a human diet minerals are provided mainly by fruits, vegetables and juices [1]. Minerals are unique nutrients because of their important role in metabolism. They are an essential part of many important enzymes. They also play roles as catalyst and antioxidant, iron and copper for example are essential in blood formation, copper is also involved in carbohydrate and lipid metabolism [2].

Citrus limunum (Fam. Rutaceae) is a species of citrus. It is a small evergreen tree native to Asia which is typically round, green to yellow in colour, 3 to 6 cm in diameter and contains a sour and acidic pulp. It is often used to accent the flavour of foods and beverages. Citrus limunum is a medicinal plant which is used in the treatment of brain troubles [3], it has also anticancer and antibacterial activities [4].

Citrus limetta is also a species of citrus native to South and South East Asia which is cultivated in the Mediterranean region [5]. It is a small tree which may reach 8 m in height. Despite having little acidity, it has high levels of vitamin C and is used in the healing of common cold, chronic asthma, fever, inflammation, digestive disorders, coronary diseases, tumors and blood clotting [6-9].

Mineral elements in the soil, juice of citrus limunum and citrus limetta mainly affect the quality of fruit and plant growth. The quantitative estimation of various element concentrations is important for determining the effectiveness of the medicinal plants in treating various diseases and also in understanding their pharmacological action [10]. Imbalance in human health has been linked to an excess or deficiency of elements in water, soils, plants and animals [11]. The continuous intake of diets that are excessively high in a particular element can influence changes in the functioning, forms and activities of some organs or increase the concentrations of the element in body tissue and fluids above permissible limit [12].

For this reason, taking into account the importance of knowing the mineral composition of citrus limunum and citrus limetta fruits, the aim of the present study was to determine the Na, K, Ca, Mg, Fe, Cu and Zn content in the soil, juice of citrus limunum (Gorgan and Iranshahr) and juice of citrus limetta (Gorgan and Bandarabas) by flame photometry and atomic absorption spectrometry methods. These methods have a wide range of applications in analytical chemistry. The advantages of these methods are accuracy, speed, high sensitivity and precision.

2. EXPERIMENTAL

2.1 Materials and Methods

Citrus limunum fruit, tree soil samples of Gorgan and Iranshahr and Citrus limetta fruit samples of Gorgan and Bandarabas were collected and analyzed in December 2013. In the first step, all fruits were cleaned and washed with double distilled water. The obtained juices were filtered through whatman No.2 filter paper. Soil samples also were collected from a depth of approximately 0-15 cm and stored in plastic containers for analysis. Soils were dried in an oven at 40 ^{0}C for 24 h before any further treatments and were ground to a fine powder using a mortar and pestle. To achieve a standard size of soil particles, the ground materials were sieved through a 2mm metal sieve. The process was repeated until all the material passed through the sieve. The soil powder (0.5 g) was placed in a 100 mL volumetric flask that contained 100 mL double distilled water. The contents were stirred for 48 h and allowed to stand for 5-8 min at room temperature. After that, the suspensions were filtered through whatman No.42 filter paper.

2.2. Reagents and Apparatus

All used chemical reagents were of analytical or chromatographic grade and were purchased from Merck (Darmstadt, Germany). Freshly prepared double-distilled water was used in all experiments. The pure salt of each element was used to prepare the standard solutions.

Analysis of metals such as Ca, Mg, Fe, Zn and Cu was performed by atomic absorption spectrometry (Philips-Pu 9100x). This system was equipped with a hollow monoelement cathode lamp (Hollow Cathode Lamp, England) for the analysis of each element. To achieve maximum sensitivity and precision, the equipment was equilibrated by aligning the lamp and lighter and adjusting the selected wave-length prior to analysis. The hollow cathode lamps were operated at 4.8 mA for Ca, 13.5 mA for Mg, 11 mA for Fe, 7.5 mA for Zn and 10 mA for Cu. The wavelengths for Ca, Mg, Fe, Zn and Cu were set to 422.7, 285.2, 284.3, 213.9 and 324.8 nm, respectively. Analysis of Na and K elements was performed by flame photometer (Corning-410). The blanks were used for zeroing the instrument before each analysis. The mineral element contents of the samples were determined by standard addition technique

3. RESULTS AND DISCUSSION

Standard addition curves for absorbance versus mineral element concentration (mg L^{-1}) were plotted for the elements. Some of the curves are shown in Fig. 1. The limits of detection (DL), the limits of quantification (LOQ) and the limits of linearity (LOL) for Na, K, Ca, Mg, Fe, Zn and Cu were determined and results were summarized in Table 1. The limits of detection were 0.47, 0.38, 0.09, 0.04, 0.3, 0.02 and 0.09 mg L^{-1}, the limits of quantification were 7.72, 59, 0.29, 0.15, 1.30, 0.26 and 0.31 mg L^{-1} and the limits of linearity were 100, 200, 3.5, 0.75, 13, 3 and 6 mg L^{-1} for Na, K, Ca, Mg, Fe, Zn and Cu, respectively.

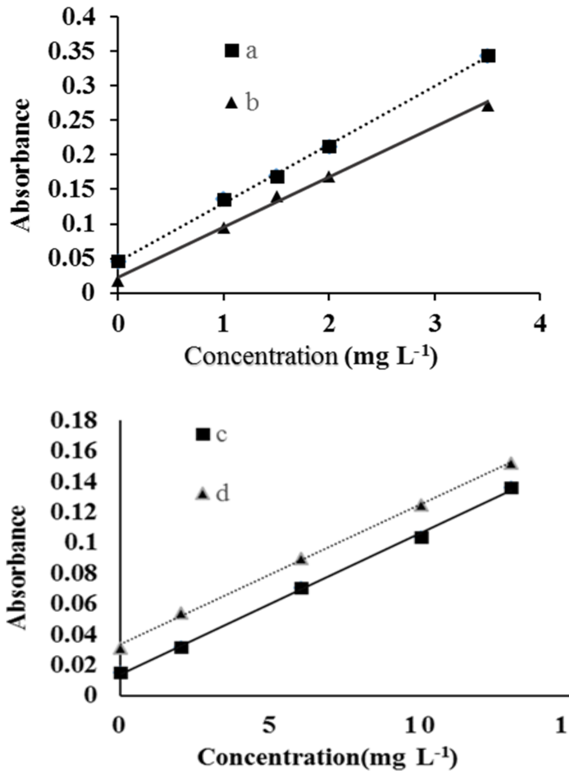

Fig. 1. Standard addition curves for absorbance versus mineral element concentration: (a) Ca in Citrus limetta of Bandarabas, (b) Ca in Citrus limetta of Gorgan, (c) Fe in Citrus limunum of Iranshahr, (d) Fe in Citrus limunum of Gorgan

Table 1. The values (mg L^{-1}) of detection limit (DL), limit of quantification (LOQ) and limit of linearity for the elements

Element	Na	K	Ca	Mg	Fe	Zn	Cu
DL	0.47	0.38	0.09	0.04	0.3	0.02	0.09
LOQ	7.72	59	0.29	0.15	1.30	0.26	0.31
LOL	100	200	3.5	0.75	13	3	6

Soil minerals play a vital role in soil fertility since mineral surfaces serve as potential sites for nutrient storage. Different types of soil minerals hold different amounts of nutrients. Soil factors such as organic matter, type and amount of clay or soil pH influence the quantity of elements available for mobilization or sorption in a soil. Sometimes competition between different metals may prevent optimal uptake of a particular nutrient, such as decreased Zn availability induced by the presence of relatively high concentrations of metals such as Fe and Ca. Under equal conditions, different plant species may adsorb the same metal from the soil at different rates [13]. The mineral element contents in the samples of tree soils of citrus limunum (Gorgan and Iranshahr gardens), citrus limetta juice (Gorgan and Bandarabas gardens) and citrus limunum juice (Gorgan and Iranshahr gardens) were measured by atomic absorption spectrometry and flame photometry methods. Because there is no certified reference material for metals in the samples of soils and fruits, these studies were performed by the standard addition technique to ensure accuracy. The results are given in Table 2. As evident from Table 2, the highest element concentration in the soil samples belongs to Ca and the lowest element concentration belongs to Zn.

There was a significant difference between the concentration of Ca and Mg elements in the soil samples of Iranshahr and Gorgan. Although the concentration of the Ca element in the soil sample of Gorgan was more than that of Iranshahr, the concentration of this element in the citrus limunum of Gorgan was less than that of Iranshahr. Moreover, the concentration of the Mg element in the soil sample of Iranshahr was more than that of Gorgan, but the concentration of this element in the citrus limunum of Iranshahr was less than that of Gorgan which is probably due to problems in the Ca and Mg absorption of the citrus limunum plant in these two areas. There were no significant differences in the values of K, Na, Fe and Zn elements and also the concentration of Cu was found to be same in both soil samples.

The results showed the highest element concentration in the fruit samples belonged to K and the lowest concentration belonged to Cu element. The amount of the Cu element in the fruit samples was below the detection limit and has not been inserted in Table 2. Significantly, the concentration of Na, K and Ca elements in the citrus limetta sample of Bandarabas was more than that of the Gorgan sample and also the concentration of the Fe and Mg elements in the citrus limunum juice of Gorgan was more than that of Iranshahr.

Although the scientific literature data concerning mineral composition of citrus limunum and citrus limetta fruits are poor, these results can also be compared with other fruits such as olive, banana and apple.

Fernandez-Hernandez et al. in 2010 reported the mineral composition in fresh olive fruits of cultivars such as Arbequina, Picual, Hojiblanca, Frantoio and Bella de España. The K levels were found to be highest, followed by Ca, Na, and Mg. Of the minority elements, Fe had the highest level, followed by Zn and Cu [14]. Anhwange et al. in 2009 reported the mineral elements in banana fruits. The K levels were found to be highest, followed by Ca, Na, Fe and Mg [15]. Juranovic´ Cindric´ and colleagues in 2012 reported the major part of elements in an apple consists of K, followed by Na, Ca, Mg and other minor elements, such as Zn and Cu [16]. The comparison of results showed that in the citrus limunum juice, citrus limetta juice, olive, banana and apple fruits the K element was more than other elements.

The elements play both a curative and preventive role in combating diseases. The preventive medicinal aspects of various elements remain vastly unexplored [17]. Potassium (K) is helpful in reducing hypertension and maintaining cardiac rhythm. In the human body, potassium plays a vital role in many physiological reactions and its deficiency or excess can affect the human health [18]. Potassium is the principal intracellular caution and with sodium helps regulate osmotic pressure and pH equilibrium [19]. The highest amount of K was detected in the citrus limetta of Bandarabas (3075 mg L^{-1}) and the lowest amount of K was in the citrus limetta of Gorgan (1274 mg L^{-1}).

Calcium (Ca) helps in overcoming the problems of high blood pressure, heart attack, premenstrual syndrome, colon cancer and retaining bone strength [11]. It also participates in muscle contractions, the conductions of nerve impulses, cell membrane permeability and blood coagulation. Calcium bioavailability within food is 25-50% and is depressed by anti-nutrient substances (oxalic acid) but raised by some amino acids, lactose and vitamin D [20-21]. The highest amount of Ca was detected in the citrus limunum of Iranshahr (208.3 mg L^{-1}) and the lowest amount of Ca was in the citrus limetta of Gorgan (39.4 mg L^{-1}).

Sodium (Na) is involved in the production of energy and the transport of amino acids and glucose into the body cells. The highest level of sodium was determined in the citrus limunum of Iranshahr (167.1 mg L^{-1}) and the lowest amount of Na was in the citrus limetta of Gorgan (8.5 mg L^{-1}).

Table 2. Mineral element content (ppm) in the samples of soils and fresh fruits of different cultivars (mg L^{-1}). Mean values ±SD[a] (n=4)

Sample						Element
Citrus limunum of Gorgan	Citrus limunum of Iranshahr	Citrus limetta of Gorgan	Citrus limetta of Bandarabas	Sample[c]	Sample[b]	
27.9±2.8	167.1±6.1	8.5±0.6	16.6±2.5	2546±116	1576±12	Na
2038±54	1702±53	1274±1	3075±18	100±6	134±28	K
149.9±0.5	208.3±0.3	39.4±0.4	68.3±0.1	11796±682	9690±54	Ca
401.6±0.1	55.2±0.1	77.3±0.1	121.4±0.1	2138±14	9376±24	Mg
3.7±0.3	1.6±0.1	5.1±0.7	5±0.2	346±4	272±14	Fe
1.5±0.2	0.9±0.2	0.7 ±0.7	0.8±0.1	BDL[d]	56±2	Zn
BDL	BDL	BDL	BDL	4±64	64±4	Cu

[a] Standard deviation, [b] The sample of tree soil of citrus limunum of Iranshar

[c] The sample of tree soil of citrus limunum of Gorgan, [d] Below of det1ection limit

Magnesium (Mg) improves insulin sensitivity, protect against diabetes and reduce blood pressure [19]. The highest content of Mg was in the citrus limunum of Gorgan (401.6 mg L^{-1}) and the lowest amount of Mg was in the citrus limunum of Iranshahr (55.2 mg L^{-1}).
Iron is a trace mineral that is essential for our health. Forming a part of the red pigment called hemoglobin in the blood; it gives blood its dark red color and helps transport oxygen to our cells. Apart from that, iron is also important for muscle protein and traces of it can be found in the liver, spleen, bone marrow and in our muscles [22]. The highest content of iron was in the citrus limetta of Gorgan (5.1 mg L^{-1}). The lowest amount of Fe was found in the citrus limunum of Iranshahr (1.6 mg L^{-1}).
Zinc (Zn) deficiency may contribute to arrested growth retardation and hair loss, delayed wound healing and emotional disturbance [11]. Its deficiency leads to impaired growth and malnutrition [23]. Zinc insufficiency may also lead to growth inhibition in children and changes in their appetite, taste, smell and body weight loss [1, 24]. The highest level of Zn was determined in the citrus limunum of Gorgan (1.5 mg L^{-1}). The lowest amount of Zn was found in the citrus limetta of Gorgan (0.7 mg L^{-1}).
Environmental factors including atmosphere and pollution, season of sample collection, age of plant and soil conditions in which a plant grows effect the concentration of elements as it varies from plant to plant and region to region [17].

4. CONCLUSION

In summary, it can be concluded that the determination of mineral elements by flame photometry and atomic absorption spectrometry methods is a reliable and simple analytical procedure for fresh citrus limetta and citrus limunum fruits mineral characterization.
The experimental results in this study showed that these fruit samples have mineral elements which are responsible for curing various diseases and are helpful to human health such that the preferred consumption of these kind of fruits can be useful for some patients. However, more detailed analysis of chemical composition of these fruit samples are required.

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