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Atomic absorption spectrometry and flame photometry for determination of minerals elements in fresh pomegranate fruit juice

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ABSTRACT

Pomegranate is an excellent source of many nutrients able to contribute to a healthy diet. Taif is the first leading Pomegranate producers in Saudi Arabia. To our knowledge, there has been no research on the Taif Pomegranate fruit juices. this study, for determination of minerals elements (sodium , potassium, copper, lead and iron) was carried out for four different pomegranates origins commercially available in Taif province, KSA such as (Abha, Egyptian, Yamani 1, Yamani 2) using Atomic absorption spectrometry and flame photometry. The results showed that the concentrations of sodium and potassium were (0.3-0.36 mg/100 gm.) and (50-95 mg/100 gm.) respectively. Lead and iron were not detected whereas copper was 0.1ug/100 gm. In conclusion, the authors recommend these methods for analysis of minerals concerning the food and human health safety.

Key words: Taif pomegranate, Atomic absorption spectrometry and flame photometry, pomegranate fruit juices, minerals elements.

INTRODUCTION

Food is routinely analyzed for a variety of elements to assess possible nutritional or toxicological implications and to ensure compliance with government regulations or product quality. Fruits are generally acceptable as good source of nutrient and supplement for food in a world faced with problem of food scarcity. Fruit and fruits products are known to be an excellent source of nutrients such as minerals and vitamins [1]. Minerals are of prime importance in determining the fruit nutritional value. Potassium, calcium and magnesium are the major ones. In the tissue of many fruits, calcium is one of the mineral believed to be an important factor governing fruit storage quality [2]. It has been reported to delay ripening and senescence [3] and to reduce storage disorder [4]. The importance of minerals such as potassium, calcium, sodium etc. to human health is well known. Required amounts of these elements must be in human diet to pursue good healthy life [5]. Trace elements play an important role in human biology, because they are either inadequately synthesized or not synthesized in the body. Trace amounts of some metals manganese, copper, and zinc, for example, are essential micro nutrients and have a variety of biochemical functions in all living organisms. While these elements are essential they can be toxic when taken in excess. In addition, some metals like lead, do not occur naturally in the body, and their presence, usually a result of occupational or pollution-related exposure, is harmful to health and children are more sensitive to these metals than adults [6, 7]. The content of mineral elements in plants depends to a high degree on the soils abundance, including the intensity of fertilization [8].

Mineral components of fruit and vegetables contribute to their nutritive value. A number of mineral elements are required in varying amounts by humans for proper growth, function and overall well-being [9, 10]. In recent years, food analysts have been interested in the amounts of the various nutritive components and their changes in fruits, and the importance of dietary minerals in the prevention of several diseases such as bone demineralization, arterial hypertension, and overall cardiovascular risk [11- 13]. Mineral nutrients are divided into two broad groups; major and trace elements. Major minerals represent 1 per cent of bodyweight and are required in amounts greater than 100 mg per day, while trace minerals make up less than 0.01 per cent of bodyweight and are essential in much smaller amounts [12]. It is estimated that 70 biological elements are needed by all living things for the normal function of their metabolism, reproductive and immune system [9,12].

At present the determination of major, minor and trace elements has taken on considerable important in both pomegranate juice and pomegranate beverages not only from nutritional, but also from the technological point of view. These elements can have a considerable influence on the production process with , in some cases, negative effect such as oxide- reduction reaction, precipitation, colloidal alteration, formation of gel [14]. Analysis of thirteen minerals content in pomegranate juice by inductively couple plasma- optical emission spectrometry (ICP-OES) in commercial pomegranate juice consumed in Turkish market [15]. A comparative study on elemental composition of seven pomegranate cultivars was determined in the different parts of the pomegranate fruit include the arils. The concentrations of major elements (N, P, K, Ca, Mg, S, Cl and Na) and trace elements (Mn, Fe, Cu, Zn, B, Ni, Co, Cr, Pb, Cd, Se, Al, As, Li, Sr, Ti and V) were determined using (ICP-OES) calibrated with different concentrations of standard solutions of the minerals [16].

Atomic absorption and flame photometer are a popular technique for the determination of metals in many types of samples. It's commonly used for the analysis of food [14,17-20]. The different of Omani fruit including the Omani pomegranate fruit were assessment in terms of moisture o f the arils and minerals elements in the juice using atomic absorption spectrometry monitoring potassium sodium and iron [21]. The physical and physicochemical studied the changes during different stages of maturation including total seed juice extracted from unripe, half-ripe and full-ripe stage are reported. The researcher concluded knowledge of changes in mineral contents would be very useful for determination of fruit quality. The measurement of minerals elements observed using Atomic absorption spectrometry [22]. Twenty different varieties of pomegranate from Turkey were analyzed for vitamin C, lipid content and elemental analysis such as (Ca, Mg, Fe, Na, and K) using Atomic absorption spectrometry (AAS) technique [23]. Physico-chemical properties and mineral contents using Atomic absorption spectrometry (AAS) of two commercial pomegranate fruits from Tunisia were established [24]. Seven different brands of pomegranate molasses from Turkish market were analyzed for minerals elements some which are important for nutrition and health using (AAS) instrument [25]. The pomegranate fruit has become an international high-value crop for the production of commercial pomegranate juice. The comprehensive chemical characterization of 45 commercial juice sample from 23 different manufactures in the United State in addition to several country producers such as Iran and turkey considering the pomegranate juices adulteration, the concentration of the minerals elements in juices take apart from these analysis using the atomic absorption and emissions techniques [26].

According to our knowledge a part of this research , no scientific study has been reported in KSA so far particularly assessment of nutrition characterize to evaluated the minerals contents and compered with the region pomegranate products such as (Abha, Egypt, Yaman1, Yaman 2). In this work describes a detailed procedure for the analysis of minerals (Copper, Iron, Lead, Potassium and Sodium) using flame atomic absorption spectrometry and flame photometer.

MATERIALS AND METHODS

Atomic absorption spectrophotometer (AAS) Buck scientific model 210 VGP equipped with a deuterium background corrector and automatic hollow cathode lamp switch was used for absorbance measurement and digital concentration readout device to analyze Pomegranate fresh fruit samples for copper, iron and lead by direct aspiration into the burner thought the sample introduction system . The method was interference free and Flame photometer Jenway clinical PFP7. The PEP7 flame photometer is a low-temperature (air/ natural gas) flame atomic emission photometer designed for the routine determination of sodium and potassium in aqueous solutions, to very important foods industry and physiological felids. The low-temperature (about 1700 ^{0}C) generates strong emission only from the most easily excited elements.

Wavelength isolation is by use of a simple narrow-band pass interference filter that is designed to transmit only the interest line. The PFP7 and PFP7/C flame photometer are low temperature single channel emission flame photometers designed for the routine determination of mineral salts [27]. The hot flame evaporates the solvent, atomizes the metal, and excites a valence electron to an upper state. Light is emitted at characteristic wavelengths for each metal as the electron returns to the ground state. Optical filters are used to select the emission wavelength monitored for the analyte species. Comparison of emission intensities of unknowns to either that of standard solutions, or to those of an internal standard, allows quantitative analysis of the analyte metal in the sample solution [28, 29].

2.1 Reagents and chemical materials :
Copper , Iron and lead stock standard solutions obtained from Buck Scientific. The stock solutions of these elements at a contraction of 1000 mg/L were already prepared in water and 5% HCL. Working solution prepared in suitable concentration used for direct determination were made in water.

-Sodium chloride Extra Pure assay: 99.45% Merkland (Germany)

-Potassium chloride Extra Pure assay: 99.56% Merkland (Germany) Preparation of 1000 mg/L Sodium stock standard solution**:**

Accurately weigh 1.2705gm of dry quality NaCl dissolve in pure deionised water and transfer into a 500ml volumetric flask . fill to the mark with pure acidified deionised water to prepare the standard solution for use with the flame photometer.

Preparation of 1000 mg/L Potassium stock standard solution**:**

Accurately weigh 0.9534 gm of dry quality KCl dissolve in pure deionised water and transfer into a 500ml volumetric flask, fill to the mark with pure acidified deionised water to prepare the standard solution for use with the flame photometer.

2.2 Sample Preparation:
Pomegranate fresh fruit from Taif product, Abha product and imported pomegranate Yamani and Egyptian) were purchased from local market. The seeds of pomegranate get from the fruit by make a shallow slit at the top the pomegranate around the crown cut all the way around the top of the rind, creating a shallow circle. Don‟t push the knife too deep, or you may burst some seeds. Pull the crown of the pomegranate off to reveal the inner seeds. Turn the pomegranate over. Repeat the process of creating a narrow slit in the rind, cutting a circle around the base of the fruit. Pull the bottom off the fruit. Turn the pomegranate back over. Cut slits in the sides of the rind, following the lines of the pith, from the top of the fruit to the bottom. Pull the fruit apart, you will now have sections of the fruit with the seeds fully exposed.

Gently loosen the seeds from the pith and place them into the clean bow. The pomegranate seeds were washed under distilled water then dried.

The triplicate fresh pomegranate from each kinds were exactly weight (100 gm). The total amounts of the seeds hand squeezed to get pure fresh pomegranate juice. The suspension solution were passed through a stainless steel mesh sieves before filtering by Whatman filter paper using Buchner funnel.

The pomegranate juice sample final volume of solution sample 60 ml. Centrifuge and stored in refrigerator. An aliquot of the samples was diluted with purified water to bring the concentration of the samples within the range of the calibration curves. All standards and solutions ready to analysis were passed through the 0.45 um desk filter.

2.3 Analysis of Mineral Elements:
Mineral compositions were carried out for fresh pomegranate juice samples for direct measurements of (Fe, Cu, Pb,

Na and K) using air- acetylene flame in combination with single element hollow cathode lamps into Buck Scientific Atomic Absorption spectrophotometer was used to evaluate the Iron, Copper and Lead . The samples solution diluted within range of the calibration curves of the elements by deionized water.

Jenway Flame Emission Spectrophotometer for evaluation of sodium and potassium. Calibration of the instrument was repeated periodically during operation. Mineral contents were calculated by comparison of their standards solutions. The blanks were used for zeroing the instrument before each analysis to avoid matrix interferences.

2.4 Moisture content and pH measurement:
The pH for the fresh pomegranates juices were measured by pH meter (Jenway pH 3305) after standardized the pH electrode .The percent moisture content is determined by measuring the mass of the pomegranates arils before and after the water is removed by evaporation in memmert oven (Germany) at 120 ^{O}C ±5 for 2 hours [30].

RESULTS AND DISCUSSION

3.1 Atomic absorption:
Table (1) list the optimum parameter for the AAS used for determination of iron, copper and lead. These conditions used for the aspiration of the standards solutions to the external calibration curve for each elements under study. Equation y= 0.069x for iron and y=0.104x+0.024 for copper. Each curve done by aspirations of a series of standards solutions diluted from the stocked standards 1000 ug/ml and 100 µg/ml. A series of dilution were made, The correlation coefficient was higher than 0.99 for all the elements evaluated. The linearity for copper was (0.1-8µg/ml) and (0-10 µg/ml) for Iron. The fresh pomegranates fruit juices samples previously prepared and measured in the same
time of analysis.

Table (2) shows the result of copper, Iron and lead in the Taif pomegranate juice, Abha and the imported pomegranate (Egypt,Yaman1 and Yaman 2). The results was that the copper concentration in Taif pomegranate is in similar concentration with Abha, Egypt and Yaman 1 and Yaman 2 pomegranates. The results are the mean of the triplicate samples for each kind. It shows negligible standards deviation for all kinds ,this indicated that the method used it is highly precise. Iron and lead is not detected in all kinds of the pomegranates for the operation parameter in this AAS techniques [15] reported that the copper, lead and iron were found at trace levels which agree which was in agreement with what found in the pomegranates under study.

3.2 Flame photometer:
After setting the flame photometer for the best emission signal of the elements under study (sodium and potassium). y=1.301and y= 0.96x+0.036 are representative the equations for calibration curves for sodium and potassium respectively. The correlation coefficient was higher than 0.99 for all the elements evaluated. The linearity for sodium was (0.1-4 µg/ml) and (0.2-1.0 µg/ml) for potassium. The fresh pomegranates fruit juices samples previously prepared and measured in the same time of analysis. The calibration curves were used to find and calculated the concentration of pomegranates juice samples. Table (3) shows the result of sodium and potassium in the Taif pomegranate, Abha and the regions pomegranate (Egypt ,Yamani1 and Yamani 2). It is the mean of the triplicate samples for each kinds.

It shows negligible standards deviation for sodium in all kinds of pomegranate and less than 2% for potassium. The sodium element concentration in Taif pomegranate are similar with that measured for Egypt and Yamini 1, Yaman 2 pomegranates. On the other hand, the result was obtained for determination the potassium was present in all sample in different concentration. the highest potassium content, (95±1.48mg/100g) was found in Abha and the lowest (50±1.59mg/100g) in Yaman 2. While the concentration of potassium in Taif pomegranate juice was 78±00 mg/ 100 gm. It is clear that potassium is the most abundant element in fruit followed by Na then Cu, while Iron and lead is not detected in all kinds of the pomegranates . These results show the significant different in the concentration of sodium and potassium in Turkish, Tunisun, Omans and the pomegranates (Taif, Abha, Egypt, Yaman 1, Yaman 2) under study. But the results agree what they reported that the potassium and sodium content the highest among the minerals elements in pomegranates juice and similar order of mineral concentration. Most of the findings in the present study with regard to the mineral components of the pomegranate Juice are consistent with the result of the other studies. Undoubtedly duo to the strong influence of the climate of the place, different environmental place, types of the soil and agricultural procedures on the contents of these elements in which is study [21- 24].

3.3 Percentage of Moisture and pH:

Fruits c o n t a i n large quantities of water in proportion to their weight. Table (3) provides the water content of several pomegranates under study. Moisture content varied within among pomegranate fruit cultivars, the lowest was in Yamani 2 while Taif pomegranate contained the highest in most of kinds. On average for triplicate samples, among all the pomegranate kinds , the moisture contents in pomegranate 81.75±0.73 per cent (Taif) , 75.77±1.35 per cent (Abha), 79.79±1.27 per cent (Egypt),78.45±0.97 per cent (Yamani1) and 65.63±2.01 per cent (Yaman 2). The maximum moisture vales are obtained for Taif pomegranate closer to what the AL- Aiman reported.

Variations in moisture contents have been attributed to the influence of climatic, genetic and geographical factors as well as differences in harvest year and growth conditions of plants.

pH was measured with a pH meter the highest pH value was obtained for Yaman1 pH (4.18) and the lowest for Egypt pH (3.79) Table (3). That main the Egyptian pomegranate fruit juice is more sour fruit juices than the other under study are common sources of weak acid.

Table (1): Recommended analysis conditions for Cu, Fe and Pb in (AAS)

Element	Cu	Fe	Pb
Wave length	327.4 nm	372.0 nm	217.0 nm
Current	1.5mA	3mA	3mA
Slit	0.7 nm	0.2 nm	0.2
Air flow meter reading	5	5	5
Acetylene flow meter reading	4	4	4
Background Correction deuterium	ON	ON	ON

Table (2): Composition of minerals elements in fresh pomegranate fruit juice in 100 grams weight seeds samples using atomic absorption spectrometry (AAS)

Original / Elements	Taif	Abha	Egypt	Yamani 1	Yamani 2	linearity	R2
Cu ug/100 grams (AAS)	0.1±NG	0.1±NG	0.1±NG	0.1±NG	0.1±NG	0.1-8	0.995
Fe ug/100 grams (AAS)	ND	ND	ND	ND	ND	0-10	0.997
Pb ug/100 grams (AAS)	ND	ND	ND	ND	ND	5-50	0.995
Water %	81.75±0.73	75.77±1.35	79.79±1.27	78.45±0.97	65.63±2.01		
PH	3.99	4.02	3.79	4.18	4.15		

Table (3): Composition of minerals elements in fresh pomegranate fruit juice in 100 grams weight seeds samples using flame emission spectrometry (FES)

Original Elements	Taif	Abha	Egypt	Yamani 1	Yamani 2	linearity	R2
Na mg/100 grams(FES)	0.300±NG	0.360±NG	0.300±NG	0.300±NG	0.300±NG	0.1-4	0.991
K mg/100 grams(FES)	78±1.33	95±1.48	84±1.65	60±1.45	50±1.59	0.2- 1.0	0.994

1 triplicate samples were analysis.; R^2: square of correlation coefficient; ND: Not detected with the operation condition for AAS. NG: Neglected

CONCLUSION

The composition of pomegranate Juice depends on the cultivar type, soil type, environmental conditions and bioavailability of trace elements, have been implicated in the variability of trace elements and heavy metals in pomegranate fruit cultivars [31]. In this study five minerals elements were determined in five kinds of pomegranates. The results presented in this work show that consuming pomegranate arils is good source of mineral element in human diet which are essential for human health, consuming a fruit per day may cover the needs amounts of the daily requirement of many minerals elements for an average . The accurate determination of trace metals in products food is important. Here in the method was used in this project for determination of iron, cupper, sodium ,potassium and lead in pomegranate was simple and does not required the sample to be subjected to any harsh and further treatment or time consuming treatments environmental friend " Green chemistry". In addition the procedure is advantageous because it has shown good perspective of being applied without further chemical treatments. The method may be applied by the industry's quality control laboratory.

Atomic emission and atomic absorption are fast, simple, and sensitive method for the determination of trace concentration for sodium, potassium, iron and copper in solution. The method is free from interferences from other elements.

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REFERENCES

[1] Nahar N, Rahaman,S., Mosiihuzzaman, M. *Journal of the Science of Food and Agriculture,* **1990.;**5:185-192.
[2] Lechaudel, M. Joas J., Caro Y.,Genard M. And Jannoyer M. *Journal of the Science of Food and Agriculture* , **2005**;85:251– 260.
[3] Ferguson, I.B *Plant Cell Environ*. **1984**; 7: 477-489.
[4] Bangeruh F, *A review phytopathol*. **1979;**17: 97-122.
[5] San,B., Yildirim,A.N.,Pola, T, M. and Yildirim, F. Asian, *Journal of Chemical* **2009**;21(4):2898-2902.
[6] Peris, M, Nollet, L.M. L. Marcel Dekker, New York, **1996**; 2nd Ed.
[7] Manuel, O., Javier, F., Carmen, C., Miquel, N., Rafael, G., Lopez, M., *Journal of Agricultural and Food Chemistry* **2004**;52:2715-2720.
[8] Kruczek, A; *Acta.Sci. Pol. Agric***. 2005**.4(2):37-46.
[9] Obiajunwa, E.I., Adebiyi, F.M. and Omoda, P.E. *Pakistan Journal of Nutrition.* **2005**;4:393-395.
[10] Hussain, A., Larsson, H., Kuktaite, R. and Johansson, E. *International Journal of Environmental Research Public Health*, 2010; 7: 3442- 56.
[11] Glew, R.H., Ayaz, F.A., Sanz, C., VanderJagt, D.J., Huang, H.S., Chuang, L.T. and Strnad, M. *Food Chemistry*, 2003; 83:363-369.
[12] Ozcan, M. FoodChemistry. **2004**;84: 437-40.
[13] Segura, R., Javierre, C., Lizarraga, M.A. and Ros, E, *British Journal of Nutrition,* **2006**;96:36-44.
[14] Mahmood S, Mahmmed TA, Salih AB,, El-Sayed SAH. *Archives of Applied Science Research*. **2011**; 3 (6):488-496**.**
[15] Arzu Akpinar- Baxizil. *Asian Journal of Chemistry*, **2010**; 22 (8):6542-6546.
[16] Olaniyi Amos Fawole and Umezuruike Linus Opara. *British Food Journal*,**2012**; 1518 - 1532
[17] Fateh, A., lnes, T., and Ali, F *African Journal, of Biotechnology*, **2007**;6:599-602.
[18] Venkatesh K. Mutalik., Jagadish G. Baragi, Somaraddi B. Mekali, C. Veeresh Gouda , N.B. Vardhaman, *Journal of Chemical and Pharmaceutical Research* , **2011**;3(6): 1097-1102.
[19] Faizul Haq Shams ur Re hman, Habib Ahmad, Zafar Iqbal, Rahat Ullah. *American Journal of Biochemistry*, **2012**;2(4): 47-50.
[20] Valvi,S.R., Rathod, V. S. *International Journal of applied biology and pharmaceutical technology,* **2011**;2: 392-297.
[21] Salma MZA, Abdulrahman AO, Maryam MA. *Science and Technology*. **2001**;6:39-44.
[22] AL- Maiman SA, Ahmad D. *Journal of Food Chemistry*. **2002**;76: 437-441.
[23] Dumlu, MU,Gurkan E. *Journal of Medicinal Food*. **2007**;10:392-395.
[24] Elfalleh W., Nizar Tlili, Ma Ying, He sheng-Hua, Ali Ferchichi, Nizar Nasri, *International Journal of food engineering*, **2011**; 7(4): 1-13**.**
[25] Bige Incedayi, Canan Ece Tamer, Omer Utku Copur, *Journal of Agricultural Faculty of Uludage University*, **2010**; 2: 37-47.
[26] Yanjun Zhang, Dana Krueger, Robert Dupo, Rupo Lee, David Wang, Navindre Seeram, David Heber, *Journal of Agricultural and Food Chemistry*, **2009**;57:2550-2557.
[27] Mary, C. H., Gregory, A. T., and Jerald, R. S., **1995**.Wiley &Sons. Inc. New York, 4th Ed,
[28] Sawyer,H. B, Wiley&Sons. **1984 ;**Inc. New York, 1st Ed.
[29] Harris, C. D., W. H. Freeman and company, **1995** New York,4th Ed.
[30] Nirmal Sinha, Jiwan Sidhu, James Wu and M. Pilar Cano , wiley- Blackwell, Chichester, UK**, 2012.**

[31] Fadavi, A., Barzegar, M., Azizi, M.H. and Bayat, M. *Journal of Food Science and Technology*, **2005**;11:113-119.