Quim. Nova, Vol. 30, No. 7, 1573-1577, 2007

Artigo

ANALYTICAL INVESTIGATION OF CHROMIUM AND ZINC IN SWEET, SOUR AND BITTER TASTING FRUITS, VEGETABLES AND MEDICINAL PLANTS

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Recebido em 14/7/06; aceito em 1/2/07; publicado na web em 24/7/07

Sweet, sour and bitter tasting fruits, vegetables and medicinal plants are an important component of human diet. The role of chromium and zinc in carbohydrate metabolism for control of diabetes is highlighted in selected commodities. Average levels of chromium and zinc in sweet taste were 0.69 ± 0.48 mg kg^{-1} and 4.81 ± 4.31 mg kg^{-1} respectively with correlation of 0.545, while in sour taste the values were 22.5 ± 22.0 mg kg^{-1} and 24.5 ± 11.8 mg kg^{-1} respectively with the correlation of 0.239 and in bitter taste, 0.61 ± 0.33 mg kg^{-1} and 4.70 ± 3.54 mg kg^{-1} respectively with correlation of 0.343. Overall, sour tasting commodities were found higher in levels of chromium and zinc and are recommended as food supplement for diabeties. None of these species contain metals above the toxic level.

Keywords: chromium, zinc, sweet.

INTRODUCTION

The trace elements chromium and zinc merit special attention when evaluating the nutritional adequacy of diabetes[1]. Phytic acid and possibly other constituents of some plant foods can reduce the bioavailability of dietary chromium and zinc considerably[1]. Metalloenzymes are important to the structural and functional integrity of the living cells[2]. Diabetes mellitus (DM) has been shown to be associated with abnormalities in the metabolism of zinc and chromium[3-7]. In Type 1 diabetes there is a lack of insulin production, in Type 2 diabetes resistance to the effects of insulin are predominant. Both Type 1 and Type 2 have the same long-term complications[8].

The biological activity of chromium depends on its oxidation number and the chemical form of the complex of which it is a part[9-11]. Glucose tolerance factor (GTF) is associated with a trivalent form of chromium and has high biological activity[10,12] (indeed Cr is one of the constituent ingredients of GTF). As age advances[13], glucose intolerance is seen, which has been attributed to chromium deficiency[13,14]. Nanogram quantities of chromium are required in every insulin dependent system[12]. It has been reported that by acting on ribosome, chromium facilitates the incorporation of insulin-stimulated amino acid into protein[11,12]. Insulin dependent diabetics excrete more chromium than the control subjects[15,16]. Chromium deficiency has also been held responsible for vascular complications associated with diabetes mellitus[17,18]. Other signs of chromium deficiency include hyperinsulinemia, hypecholestrolemia (common in type 2 DM), decreased insulin receptor number, decreased insulin binding and DM-associated neuropathies and vascular pathologies[19,20].

Only 2% of the chromium delivered by the diet is absorbed in the small intestine[16]. Chelation with dietary amino acids, especially L-histidine; or complexation with oxalate, abundant in many dietary vegetables and grains, or with nicotinic acid or ascorbic acid or picolinic acid, can significantly augment the intestinal absorption of this mineral[21,22]. Blood, urine and hair chromium values do not provide ample information on chromium storage in the human body. Despite this limitation, chromium rich herbs have been shown effective in many studies on patients with DM[23]. Medicinal plants used specifically for DM had a chromium content approximately 3X than found in other plants from the same area that were not used for DM[23].

Zinc plays a clear role in the synthesis, storage and secretion of insulin[24-31] as well as conformational integrity of insulin in the hexameric form. Normal human pancreas contains significant quantities of zinc, whereas the diabetic human pancreas contained very little quantities of zinc. Organic compounds, which are capable of reducing zinc content of pancreas, are found to be diabetogenic. The hypersecretion of insulin associated with obese type 2 DM patients has also been shown to cause hyperzincuria. The decreased zinc level affects the ability of the islet cell[32] to produce and secrete insulin. Many complications of diabetes may be related to increased intracellular oxidants and free radicals associated with decreases in intracellular zinc and in zinc dependent antioxidant enzymes[31,32].

Zinc serves an essential role as a cofactor for more than 200 metalloenzymes[33-35], many of which regulate the metabolism of carbohydrates, lipids and protein. Zinc levels in plasma, leucocyte and erythrocyte are significantly lower in diabetics than in non-diabetics[34-36]. Insulin itself is stored in an inactive form in the presence of zinc. About one-third of the zinc consumed is absorbed in the duodenum[35,36]. Zinc competes for absorption with calcium, non-heme iron and copper because they all are divalent and are present in Irving-William series. Zinc's ability to decrease copper absorption makes it an effective part of treatment for hypercupremia associated with Wilson's disease[37,38]. Two proteins located within the intestinal lumen, metallothionein (MT) and cysteine-rich intestinal protein (CRIP) sequester zinc in the intestine and control the amount made bioavailable[36,38].

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Sweet, sour and bitter taste fruits, vegetables and medicinal plants are used in our daily life without knowing their nutrients. The present research is designed to discuss the availability of chromium and zinc in selected commodities. The documented data may be useful for health practitioners in connection to diabetes mellitus and for vegetarian diets.

EXPERIMENTAL

Reagents and materials

All reagents used were of AR grade. Graduated pipettes up to 0.005 ml accuracy were used. All other volumetric Pyrex glassware used were of A grade calibrated. Deionised water was used throughout the studies.

Collection of samples

Three samples of each sweet, sour and bitter taste fruits, vegetables and medicinal plants were purchased from the local markets and were claimed to be fresh. Botanists of Cholistan Institute of Desert Studies (CHIDS), Islamia University, Bahawalpur, identified all the collected samples.

Sweet and bitter taste samples

The collected samples were washed with deionised water to eliminate dust, possible parasite or their eggs and excess moisture was eliminated by placing the washed samples on filter paper. Washed samples were ground, if necessary, in pestle and mortar. Each sample was weighed (3 g) and digested with concentrated HNO_3 (20 mL) at 70 °C till near to dryness. The process was repeated until the entire sample was digested and organic matters were converted into CO, CO_2, NO etc. During digestion, when 1-2 mL of reacting HNO_3 was left, 10 mL of $HClO_4$ was added to boost up the oxidizing power of HNO_3. The solution was heated slowly near to dryness. The residue was dissolved in water and heated (if necessary) to dissolve any leftover residues. The solution was filtered through coarse type sintered funnel and made the total volume of 100 mL[2-4].

Sour taste samples

The samples were cleaned, washed with deionised water to remove mud or dust particles and dried at 150 °C to a constant weight. The dried samples were then ground to fine powder. Each ground sample was weighed in a precleaned porcelain crucible and heated in a muffle furnace at 400 °C till there was no evolution of smoke. The ash was then moistened with concentrated H_2SO_4 (0.5-0.8 mL) and then heated till the fumes of H_2SO_4 ceased. The crucible with the ash was heated at 600 °C till the weight of the contents was constant. This sulphated ash was dissolved in concentrated HCl and then diluted to 250 mL with deionised water and filtered through Whatman 42 filter paper. The prepared solutions were directly used for the determination of chromium and zinc[2-4].

Atomic absorption spectrophotometer measurements

The digested samples were analysed by using air-acetylene flame in combination with single element hollow cathode lamps into an atomic absorption spectrophotometer Hitachi model A-1800. Calibration of the instrument was repeated periodically during operation. Chromium and zinc contents were calculated by comparison of their standards solutions[2-4]. The experimental setting values for chromium and zinc are given in Table 1. The blanks were used for zeroing the instrument before each analysis to avoid matrix interference. All standard used were of ultrahigh purity (certified > 99.9%) procured from E-Merck, Germany, or British Drug House Chemicals Ltd., Poole, UK (BDH). Triplicate sub-samples of each sample were run separately in order to record average metal concentrations.

Table 1. Specific instrumental conditions for analysis of chromium and zinc by FAAS

Parameters	Chromium	Zinc
Wavelength (nm)	357.9	213.8
Band Pass (nm)	1.3	1.3
Lamp Current (mA)	7.5	10
Fuel Pressure (kg cm^{-2})	0.40	0.30
Burner height (mm)	7.5	7.5
Calibration Range (mg L^{-1})	0.2 – 5.0	0.05 – 0.9
Detection Limit (mg L^{-1})	0.01	0.01
Flame Composition	Air : C_2H_2	
Oxidant Pressure (kg cm^{-2})	1.60	
Atomizer	Standard Burner	
Measurement Mode	Absorbance	

Analytical precision

A parallel comparative check on the accuracy of quantified results was made through the use of standard reference material, SRM-1572, provided by National Bureau of Standards, Washington USA and the reproducibility of the results was checked by carrying out a triplicate analysis of each sample. The triplicate results did not differ by more than 5% of the mean. Citrus leaves, SRM-1572, were digested and then analysed following the same procedures. Certified values for chromium and zinc were 0.8 ± 0.2 and 29.0 ± 2.0 μg g^{-1} respectively. The analysed levels for chromium and zinc were 0.76 ± 0.23 and 30.4 ± 1.6 μg g^{-1} respectively. The percentage recoveries were 95 and 105 respectively.

Statistical analysis

The data was statistically analysed by using SPSS 12 and STATISTICA (StatSoft 1999) softwares on P-IV system. The levels of chromium and zinc in collected samples were correlated by linear correlation coefficient matrix (pearson).

RESULTS AND DISCUSSION

Sweet, sour and bitter taste fruits, vegetables and medicinal plants are an important component of human diet. The World Health Organization estimates that 4 billion people, or 80% of the world's population, use herbal medicine for some aspects of primary health care. Herbal medicines are economical and well effective. Considering the significance of metals and consumption of these species, this investigation shows the levels of chromium and zinc in selected commodities.

The levels of chromium and zinc in sweet taste fruits, vegetables and medicinal plants are summarized in Table 2 and shows that maximum level of chromium is found in apple, banana and carrot, which are 1.71, 2.20 and 1.47 mg kg^{-1} respectively. Similarly maximum level of zinc is observed in apple and banana, which is 15.4 and 12.4 mg kg^{-1} respectively. This shows that nature always

Table 2. Levels of chromium and zinc in sweet taste fruits, vegetables and medicinal plants

Sample code	Common name	Botanical name	Family name	Chromium mg kg^{-1}	Zinc mg kg^{-1}
St – 1	Almond	*Prunus amygadalus*	Rosaceae	0.34	5.89
St – 2	Apple	*Pyrus malus*	Rosaceae	1.71	15.4
St – 3	Apricot	*Prunus amenica*	Rosaceae	0.73	8.74
St – 4	Banana	*Musa paradisica*	Musaceae	2.20	12.4
St – 5	Carrot	*Dacus corota*	Umberlifera	1.47	6.67
St – 6	Date palm	*Phoenix dactylifera*	Palmae	0.50	3.46
St – 7	Glycine	*Glycyrrhiza glabra*	Leguminoseae	0.60	4.06
St – 8	Guava	*Psidium guayava*	Myrtaceae	0.54	2.93
St – 9	Jambolan (pulp)	*Eugenia jambolana*	Myrtaceae	0.82	0.75
St – 10	Persimon	*Diospyros kaki*	Ebenaceae	0.71	0.06
St – 11	Lassori	*Cordia myxa*	Boraginaceae	0.17	7.90
St – 12	Raisin (Partially dried grapes)	*Vitis vinefera*	Vitaceae	0.70	7.49
St – 13	Mulbery	*Morus lavigata*	Moraceae	0.69	2.19
St – 14	Musk melon	*Cucumis melo*	Cucurbitaceae	0.79	5.57
St – 15	Potato	*Solanum tuberosum*	Solamacceae	0.74	5.93
St – 16	Pumpkin	*Bannicasa cerifera*	Cucurbitaceae	0.43	0.70
St – 17	Sew ber	*Zizyphus hysteria*	Rahamnaceae	0.55	4.08
St – 18	Sugar cane (juice)	*Saccharum officinarum*	Garminae	0.25	0.20
St – 19	Sweet lime (juice)	*Citrus limeta*	Rutaceae	0.23	0.01
St – 20	Sweet orange (juice)	*Citrus sinensis*	Rutaceae	0.26	0.94
St – 21	Water cress	*Trape bispinosa*	Nymphacaciae	0.61	10.3
St – 22	Water melon	*Citrus vulgaris*	cucurbitaceae	0.21	9.14
St – 23	Brown sugar (Partially refined)	———	———	0.77	0.34
St – 24	Honey	———	———	0.74	0.29
St – 25	White sugar (refined)	———	———	0.02	0.05
	Average levels	———	———	0.69	4.81
	±SD	———	———	±0.48	±4.31
	Correlation between chromium and zinc	———	———	0.545	

pertains toward perfection. The refined white sugar, which has negligible amount of chromium and zinc as compared to brown sugar, is dangerous for human health especially for diabetic patients and is sometime called as white poison. In modern society, the use of white sugar is considered as a matter of prestige, but really we are going away from natural ways. For such type of deviation we are not forgiven, so several complicated diseases are seen in people using white sugar and diabetes is one of them. The regular use of fruits and vegetables, as a part of diet, maintains health but can cover the deficiencies of metallic elements in the body[12,15].

The results of Table 3 reveal that most of the sour taste plants contain reasonable amounts of chromium and zinc. Maximum level of chromium is found in sour grapes and sour mango, which is 74.0 and 62.3 mg kg^{-1} respectively. Similarly, the maximum level of zinc is found in sour mango powder and dried pomegranate, which is 45.0 and 40.0 mg kg^{-1} respectively. According to these

Table 3. Levels of chromium and zinc in sour taste fruits, vegetables and medicinal plants

Sample code	Common name	Botanical name	Family name	Chromium mg kg^{-1}	Zinc mg kg^{-1}
Sr – 1	Oxalis (Khatti booti)	*Oxalis corymbosa*	Oxalidaceae	18.6	29.0
Sr – 2	Tamarind (Imli)	*Tamarindus indica*	Caesalpiniodceae	1.50	11.0
Sr – 3	Plum (Aloo bukhara)	*Prunus bokherensis*	Rosaceae	7.60	33.0
Sr – 4	Olive (Amla)	*Phylanthus emblica*	Euphorikceae	10.0	15.0
Sr – 5	Tomato	*Lycopersicum*	Solanaceae	3.30	20.0
Sr – 6	Sour apple	*Pyrus malus*	Rosaceae	6.90	5.90
Sr – 7	Sour grapes	*Vitis vinifera*	Vitaceae	74.0	9.00
Sr – 8	Sour mango powder (Aamchoor)	*Mangifera indica*	Anacardiaceae	62.3	45.0
Sr – 9	Pomegranate	*Punica granatum*	Puniaceae	30.7	40.0
Sr – 10	Orange (Malta)	*Citrus sinensis*	Rutaceae	21.5	30.0
Sr – 11	Lime (Khatti)	*Citrus acida*	Rutaceae	17.7	28.0
Sr – 12	Citron (Lemon)	*Citrus medica*	Rutaceae	20.0	25.0
Sr – 13	Spinach	*Spinacia oleracea*	Chenopodiaceae	18.1	27.0
	Average levels	———	———	22.5	24.5
	±SD	———	———	±22.0	±11.8
	Correlation between chromium and zinc	———	———	0.239	

results the amount of chromium and zinc present in sour taste plant samples is much larger than that present in sweet tastes (Table 2) and bitter taste plant samples (Table 4). Although the bitter taste edible species are being recommended by traditional herbal practioners for the treatment of diabetes and hypercholesterolemia but the practioners are not aware of the actual chemical nature of the edible things. An interesting thing is found that those fruits which are sweet in taste after ripening, e.g. grapes, apples etc, contain less amount of chromium and zinc as compared to those fruits which become sour in taste after ripening e.g. oxalis, rumax, etc. Within a single species like mango and oranges, sweet ones contain less chromium and zinc than that of sour ones. The reason to explain this phenomenon remains to be elucidated.

The results of chromium and zinc determinations in different bitter taste fruits, vegetables and medicinal plants are shown in Table 4. It is found that chromium contents in *Gymnema sylvester* (1.56 mg kg^{-1}), *Melia azadriachta* (0.93 mg kg^{-1}) and *Terminalia chebula* (0.91 mg kg^{-1}) are found higher. Higher levels of zinc are found in *Cesalpini bandocella* (11.6 mg kg^{-1}), *Momordica dioica* (11.0 mg kg^{-1}) and *Gymnema sylvester* (1.56 mg kg^{-1}). These plants are widely used for preparation of antidiabetic herbal medicines. Bitter tasted commodities levels closely resemble as those for sweet taste fruits, vegetables and medicinal plants. Rich quantities of these trace elements are also found in the medicinal plants like *Gymnema sylvester*. This plant is known as 'sugar killer herb' because on chewing, followed by eating of sugar, it gives no sense of sweetness to tongue. Indigenous herbalists (Hakeems) used this herb for the cure of diabetes. This is probably due to the presence of chromium and zinc, which instigate pancreas to produce insulin that metabolize the sugar. They also advise the patients to use hairy mordica, seeds of jambolan, neem tree, fruit of bitter gourd, *vincea rosa* (evergreen), flowers and seeds of fenugreek and horse radish moringa (suhanjna) legumes etc. as a remedy for the cure of water retention and diabetes.

Average levels of chromium and zinc in sweet taste are 0.69 ± 0.48 and 4.81 ± 4.31 mg kg^{-1} respectively with correlation of 0.545, in sour taste 22.5 ± 22.0 and 24.5 ± 11.8 mg kg^{-1} respectively with correlation of 0.239 and in bitter taste 0.61 ± 0.33 and 4.70 ± 3.54 mg kg^{-1} respectively with correlation of 0.343. Strong correlation *i.e.* 0.545 in sweet taste samples suggests that chromium and zinc have also their role in taste development of natural products. Overall sour taste commodities are found higher in levels of chromium and zinc and are recommended for diabetes food supplement. None of these species contain metals above the toxic level.

CONCLUSION

In conclusion, zinc and chromium have potential beneficial antioxidant effects in people with type 2 DM and also help to prevent free radical damages in them. The increase in glomerular filtration rate associated with DM leads to an increased filtered load of various minerals in the urine. When the status of one of these discussed minerals is poor in a patient with DM, diet rich in that mineral will probably be beneficial. Risk-to-benefit ratio should be assessed and communication between the patient and healthcare providers should be open and frank. We suggest the use of unripe mango (sour), pomegranate, *Citrus sinensis, Citrus medica, Oxalis corymbosa, Spinacia oleracea, Phylanthus emblica, Gymnema sylvestre, Terminalia chebula, Melia azadriachta* (seeds) and *Euegnia jambolana* (seeds) in the diet of patient with DM and for preparation of antidiabetic herbal medicines.

ACKNOWLEDGEMENT

The authors highly acknowledge Dr. M. Arshad, Deputy Director, Cholistan Institute of Desert Studies (CHIDS), Islamia University, Bahawalpur, for his help and cooperation in sample

Table 4. Levels of chromium and zinc in bitter taste fruits, vegetables and medicinal plants

Sample code	Common name	Botanical name	Family name	Chromium mg kg^{-1}	Zinc mg kg^{-1}
Bt – 1	Bitter almond	*Prunus amygdalus*	Rosaceae	0.57	3.71
Bt – 2	Bitter gourd	*Citrulus colocynthius*	Cucurbitaceae	0.50	2.98
Bt – 3	Cheese maker	*Within coagulans*	Solanaceae	0.57	7.17
Bt – 4	Gillo (Dried shoots)	*Tinospora cordifolia*	Menispermaceae	0.34	0.57
Bt – 5	Gurmar buti	*Gymnema sylvestre*	Asclepiadaceae	1.56	9.91
Bt – 6	Hareer myrobalan	*Terminalia chebula*	Combretaceae	0.91	7.56
Bt – 7	Jambolan (Seed)	*Euegnia jambolana*	Myrtaceae	0.71	0.13
Bt – 8	Karir fruit pulp (Dela)	*Capparis aphylla*	Capparidaceae	0.67	1.16
Bt – 9	Hairy mordica (Unpeeled)	*Momordica dioica*	Cucurbitaceae	0.26	11.0
Bt – 10	Hairy mordica (Peeled)	*Momordica dioica*	Cucurbitaceae	0.27	4.91
Bt – 11	Fenugreek (Methi)	*Trigonella foenum graecum*	Fabacaea	0.18	2.85
Bt – 12	Neem leaves	*Melia azadriachta*	Meliaceae	0.69	1.14
Bt – 13	Neem fruits	*Melia azadriachta*	Meliaceae	0.93	5.41
Bt – 14	Neem seeds	*Melia azadriachta*	Meliaceae	0.21	5.27
Bt – 15	Purging cassia	*Cassia fistula*	Caesalpiniacae	0.89	6.18
Bt – 16	Salt blush root (Tooth brush tree)	*Salvadora persica*	Salvadoraceae	0.17	0.10
Bt – 17	Bonduc nut	*Cesalpini bandocella*	Sapinodaceae	0.56	11.6
Bt – 18	Soap nut	*Spindudus trifoliatus*	Sapinodaceae	0.88	6.47
Bt – 19	Evergreen (Pink)	*Vincea rosa*	Aponcynaceae	0.47	2.43
Bt – 20	Evergreen (White)	*Vincea rosa*	Aponcynaceae	0.59	1.06
Bt – 21	Snake gourd (Charungli)	*Caralluma edulis*	Asclepiadaceae	0.79	7.19
	Average levels	———	———	0.61	4.70
	±SD	———	———	±0.33	±3.54
	Correlation between chromium and zinc	———	———	0.343	

collection, identification and preservation. Dr. T. M. Ansari, Department of Chemistry, Bahauddin Zakariya University, Multan, is also acknowledged for providing AAS facilities.

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