Rapid Detection of Nutrients in "Zea mays L" using Inductively Coupled Plasma Mass Spectrometry

SHAISTA QAMAR[1,*], MUHAMMAD ASLAM[2], FAHRUL ZAMAN HUYOP[1], MUHAMMAD ARSHAD JAVED[1,*]
[1]*Faculty of Biosciences and Medical Engineering (FBME), University Technology Malaysia (UTM), 81310 UTM Skudai, Johor Bahru, Malaysia.* [2]*Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan*
 *Address for correspondence to: Faculty of Biosciences and Medical Engineering (FBME), University Technology Malaysia (UTM), 81310 UTM Skudai, Johor Bahru, Malaysia.
Tel: +60 17 776n9049; Email: shaistapbg@gmail.com, majaved@fbb.utm.my

Abstract

Trace elements and minerals found in our food are essential for our health but at the same time may become hazardous if taken in high concentrations. Therefore, the analysis of trace elements in the food is very crucial and indispensable. The present work reports for the first time results of the analysis of mineral composition (macro-, micro- and trace-elements) in yellow maize flour using inductively coupled plasma-mass spectrometry (ICP-MS). Concentrations of 20 elements including toxic elements (Pb, Cd, As, Ni, Cr, Sr, U), essential elements (Ca, Co, Se, Zn, Cu, Fe, Mn, Mo) and probably essential elements (Mg, K, Na, Ba, Al) were detected in yellow maize flour. The results of the analysis of the inorganic elements such as Al, As, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, Pb, U, Zn yellow maize flour using ICP-MS, revealed contents of 200.05, 0.17, 2.53, 1290.27, 0.04, 0.145, 9.85, 11.46, 115.13, 914.98, 1594.14, 21.48, 1.71, 63.89, 6.08, 17.05, 5.23, 0.027, 33.89 µg/g in yellow maize flour, respectively. The detailed analysis of Cd or Pb in yellow maize flour revealed that none of them exceeded the maximum limit set by the European Legislation or values recommended by the codex alimentarius. The distinctive features of ICP-MS for rapid sample analysis demonstrated by this study suggests that this method offers promise for precision measurements of maize flour inorganic nutrients as compared to conventional methods.
Keywords: maize, flour, quantitative, inorganic nutrients, inductively coupled plasma mass spectrometry.

1. Introduction

Maize, or corn, (Zea mays L.) belongs to the grass family (Poaceae) and is the top cereal grain as measured by production. It ranks third, after wheat and rice, as a staple food for both humans and animals worldwide [1,2]; it also serves as a raw material for agriculture-based industries. It is cultivated on more than 142 million ha of land worldwide and its annual production exceeds 900 metric tons [3], with approximately 60 % produced in developed countries, particularly the United States of America. China produces 27 % of the world's maize, and the rest is grown throughout Africa, Latin America, and southern Asia, with a large proportion being produced in the tropics and subtropics [4]. The most important step in maize processing is milling, which produces different types of flour, by-products such as bran and germ, and impurities extracted during processing [5,6].

Numerous essential trace elements and minerals found in maize are important for humans to consume and absorb, as they are necessary for healthy development, but they can be hazardous to our health in high concentrations [7,8]. Trace elements are usually categorized into three groups from a dietary point of view: essential trace elements (iron, zinc, copper, cobalt, chromium, fluorine, iodine, manganese, molybdenum, and selenium); probably essential trace elements (nickel, tin, vanadium, silicon, and boron), and non-

essential trace elements (aluminium, arsenic, barium, bismuth, bromine, cadmium, germanium, gold, lead, lithium, mercury, rubidium, silver, strontium, titanium, and zirconium) [9-11]. Insufficient intake of essential trace minerals can cause symptoms of nutritional deficiency [12]. Therefore, detection and analysis of these trace mineral elements in foods, including maize, can provide useful assessment and control of safe and healthy food. Such detection requires sensitive and accurate methods of analysis; currently, most of the trace elements are estimated with colorimetric and spectroscopic methods. We investigated essential, probably essential, and non-essential trace elements by using inductively coupled plasma mass spectrometry (ICP-MS) in the grains of yellow maize grown at the University of Agriculture, Faisalabad, Pakistan. ICP-MS has some distinctive advantages over other detection techniques. For example, it can measure several elements simultaneously, has very low detection limits, and offers a wide linear dynamic range that allows the determination of major and trace elements in the same sample, which is not possible with other techniques.

To the best of our knowledge, this is the first study regarding the evaluation of trace elements using ICP-MS in maize. Detection and analysis of trace elements using ICP-MS may also provide assessment and monitoring of the pollution of the environments (soil, water, and air) with which maize, and all agricultural products, are in direct contact.

2. Materials and Methods

The investigation was carried out at the research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan. A randomized complete block design was used in growing experimental material, with three replications under normal conditions. Plant-to-plant and row-to-row distances were 20 cm and 40 cm, respectively. At maturity, plants were harvested manually and grains were used for the study of proximate chemical composition and detection of inorganic nutrients. The grains were grinded and passed through a 1-mm sieve of a milli micro mill (Model DFH-48, Culatti, Switzerland), then shaken for 8 h at 50 °C. Nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) were purchased from Sigma Aldrich (St. Louis, MO, USA). High-purity distilled water obtained from a Millipore Milli-Q water purification system (Merck Millipore, Billerica, MA, USA) was used throughout the work. Samples of white and yellow maize flour were pre-digested in nitric acid (HNO_3) as described in the literature [13]. In brief, 500 mg of the yellow maize flour samples was added to 7 mL of 70 % HNO3 and kept at room temperature for 6 h. After that, 1 mL of H_2O_2 was added, then digestion was carried out using a microwave oven. The digestion temperature was increased from 30 °C to 180 °C in 50 min, and the digestion was completed when the sample was colourless. Finally, the digested sample volume was adjusted to 50 mL with high-purity distilled water. A working standard solution for each tested element was prepared fresh daily by serial dilution from a stock solution containing 1000 mg L^{-1} of the element (Merck, Kenilworth, NJ, USA). Analytical determination of trace metals was carried out using a NexION 300 D ICP-MS (Perkin Elmer, Waltham, MA, USA). All analyses were performed in triplicate and checked by standard addition. The ICP-MS calibration was carried out by external calibration. The calibration curves of six elements (As, Cd, Cr, Ni, Pb, and Zn) were obtained using a blank and four working standards (0, 10, 20, 40, and 100 µg L^{-1}) and for Al obtained by the instrument using the blank and three working standards (0, 100, 200, and 300 µg L^{-1}) for all elements, starting from a 1000 mg L^{-1} single standard solutions for ICP-MS (Aristar grade, BDH Laboratory Supplies, England) for the trace elements. Correlation coefficients for all elements were 0.998–0.999.

3. Results and Conclusions

A sample of the yellow maize flour used in this experiment is shown in Fig. 1. Small-size debris aggregates with large-size grains and irregular residues, which could correspond to non-starch components, were observed in the ground maize.
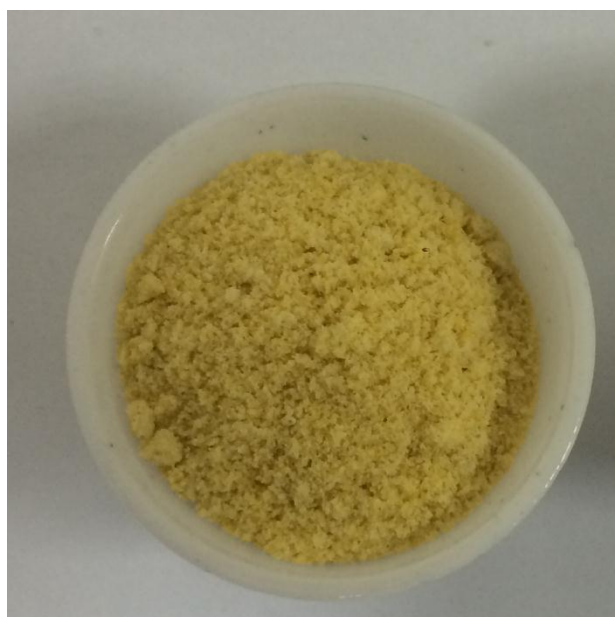

Figure 1: Photograph of the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan.

Table 1 summarizes the operating conditions of the ICP-MS. Quantitation of the elements was obtained using the external calibration curve method.

Table 1: *ICP-MS (NexION 300 D) operating conditions and data acquisition parameters.*

RF power	1600 W
Nebulizer gas flow	0.65 L/min
Lens Voltage	9.55 V
Analog Stage Voltage	-1745 V
Pulse Stage Voltage	950 V
Number of Replicates	3
Reading / Replicates	20
Scan Mode	Peak Hopping
Dwell Time	40 ms
Integration	1200ms

Three- to five-point calibration curves, with appropriate dilutions, were used for quantitation, and the concentration ranges for the twenty elemental STDs are shown in Table 2. The calibration curves had good linearity ($R^2 > 0.994$) within the investigated range. The limit of detection (LOD) and limit of quantitation (LOQ) for all twenty elements were determined for each calibration curve using the equations $LOD = 3 \times SD/S$ and $LOQ = 10 \times SD/S$, where SD is the standard deviation of a response, and S is the slope of the calibration

curve (Shrivastava and Gupta, 2011). In the current study, the LOD ranged from 0.0017 to 0.280 µg mL-1, and the LOQ from 0.0057 to 0.934 µg mL-1, as shown in Table 2.

Table 2: *Calibration curves, limit of detection (LOD), and limit of quantitation (LOQ) of 20 elements examined in the present study.*

Element	Concentration µg/mL	Linearity (r^2)	Slope (S)	SD of Response	LOD µg/mL	LOQ µg/mL
Al	0.01-0.1	0.9984	15273.4	63.0	0.012	0.041
As	0.1-1	0.9995	529.5	49.5	0.280	0.934
Ba	0.01-0.1	0.9997	6614.1	12.0	0.0054	0.0181
Ca	0.01-0.1	0.9980	1312.7	32.0	0.0731	0.243
Cd	0.01-0.1	0.9998	1819.4	13	0.0214	0.0714
Co	0.01-0.1	0.9991	3249.4	13.5	0.0124	0.0415
Cr	0.01-0.1	0.9990	2929.2	27	0.0276	0.0921
Cu	0.01-0.1	0.9992	14530.8	42	0.0086	0.0288
Fe	0.1-1	0.9989	1120.4	30	0.080	0.2677
K	0.01-0.1	0.9971	11946.9	32.96	0.0082	0.0275
Mg	0.01-0.1	0.9991	13907.9	58	0.0125	0.041
Mn	0.01-0.1	0.9991	4358.4	24	0.165	0.055
Mo	0.01-0.1	0.9999	5001.5	11	0.0065	0.0219
Na	0.01-0.1	0.9970	54780.8	50	0.0027	0.0091
Ni	0.01-0.1	0.9981	6790.1	35	0.015	0.0515
Pb	0.01-0.1	0.9995	9601.0	49	0.0153	0.0510
Se	0.1-1	0.9998	770.7	4.7	0.0183	0.0610
Sr	0.01-0.1	0.9997	17985.1	18	0.0030	0.010
U	0.01-0.1	0.9998	17267.9	10	0.0017	0.0057
Zn	0.1-1	0.9991	378.4	1.7	0.0134	0.044

In general, the results for most of the elements indicate the efficiency of ICP-MS and its ability to provide suitable detection limits and LOQ for maize flour analysis. Concentrations of the twenty essential, probably essential, and toxic trace elements in the maize flour, as detected by ICP-MS, are presented in Table 3.

Table 3: *Concentration of 20 elements determined by ICP-MS in the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan.*

Elements	Maize Flour (µg/g)
Al	200.052 (1.3)
As	0.173 (4.1)
Ba	2.53 (1.2)
Ca	1290.272 (1.0)
Cd	0.04 (7.2)
Co	0.148 (2.7)
Cr	9.854 (1.0)
Cu	11.465 (0.7)
Fe	115.13 (1.2)
K	914.984 (0.9)
Mg	1594.137 (0.7)
Mn	21.481 (1.9)
Mo	1.171 (1.4)

Na	63.887 (0.2)
Ni	6.083 (0.8)
Pb	17.047 (0.3)
Se	0.062 (6.9)
Sr	5.231 (0.9)
U	0.027 (1.3)
Zn	33.898 (0.6)

The results are the mean values for three replicates (n = 3) with RSDs. The precision was good, varying from 0.2 % (for Na) to 7.2 % (for Cd) in our samples. The concentrations of all twenty studied elements, grouped as essential, probably essential, or toxic, are shown in Figs. 2 and 3. As can be seen from Fig. 2(b), the essential elements are higher in concentration in yellow maize flour and can be arranged as follows: Se < Co < Mo < Cu < Mn < Zn < Fe < Ca. The concentrations of the elements detected using ICP-MS were in good agreement with those obtained using other techniques, although we found that Ca and Fe were higher than those obtained by [14-16]. The Zn and Cu content was significantly lower than the Ca and Fe content, but are comparable to those determined by Ullah et al. (2010), who determined that the Zn content of maize grains was 37.05–52.4 ppm, and Cu concentration was higher than those obtained by [16].
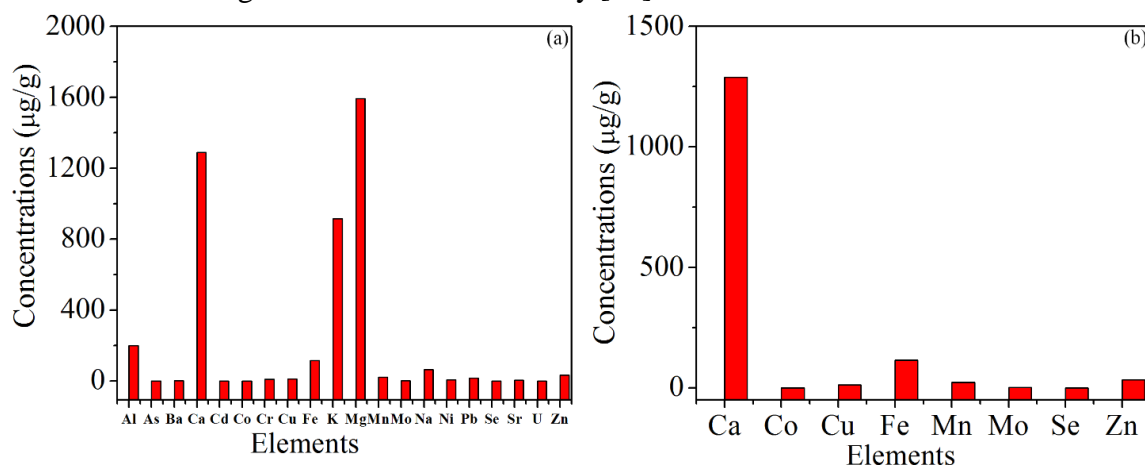

Figure 2: (a) Overall concentration of 20 elements determined by ICP-MS in the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan (b) Concentration of essential elements determined by ICP-MS in the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan.

Fig. 3(a) shows the concentrations of the probably essential elements (Mg, K, Na, Ba, Al) in the yellow maize flour. As seen in the Fig. 3(a), the concentrations of these elements can be arranged as Ba < Na < Al < K < Mg. The concentration of Mg found here is higher than those found by [14-16], whereas concentrations of Na and K are significantly lower than those obtained by [17]. The concentration of Al is slightly higher than those obtained by [17] in maize and those obtained by [13,18]. The concentration of Ba is relatively low (2.13–2.53 lg g-1), which is comparable to that obtained by [19] in rye crispbread and higher than those obtained by [13] in rice. Fig. 3(b) shows the concentration of toxic elements (Pb, Cd, As, Ni, Cr, Sr, and U) in the yellow maize flour used in the current study. Concentrations of these elements can be arranged as follows: U < Cd < As < Sr < Ni < Cr < Pb. The concentration of Pb was 9.49 µg g-1, which is comparable to that obtained by [18] but significantly higher

than that reported in other relevant studies [19-23]. The concentrations of Ni, Cr, and U were very low compared to those obtained by [18]. The Cd concentration found here was quite low compared to those reported by [19,21] and [20]. We believe that these differences could be due to the source of the cereals, including geographic area, nature of the soil, or different production processing.
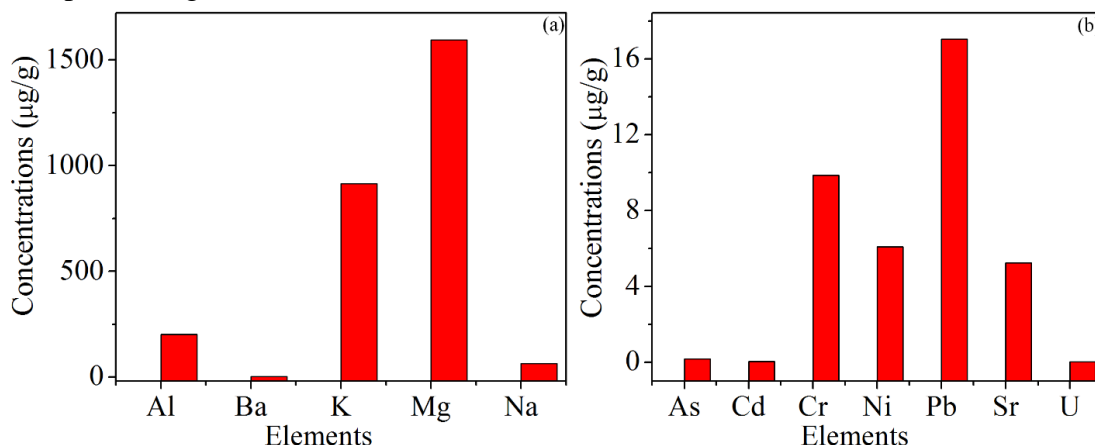

Figure 3: (a) Concentration of the probably essential elements determined by ICP-MS in the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan (b) Concentration of toxic elements determined by ICP-MS in the yellow maize flour powder grown at research area of the Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan.

For the first time, inductively coupled plasma mass spectroscopy (ICP-MS) was used to rapidly quantify inorganic trace elements in yellow maize flour. A large number (20) of trace essential, probably essential, and toxic elements were identified by ICP-MS. Excellent detection limits and limits of quantitation were obtained at both low and high concentration levels. The procedure presented here is also time- and cost-efficient, and thus suitable for routine analysis of maize and other crops and vegetables as part of the assessment of food quality and safety.

4. Acknowledgements

References

1. M.F. RODRÍGUEZ, C.A. ASCHERO. Archaeological evidence of zea mays L.(poaceae) in the southern argentinean puna (antofagasta de la sierra, catamarca). *J. Ethnobiol.* 256: 27 (2007).
2. P. RENGASAMY. Transient salinity and subsoil constraints to dryland farming in australian sodic soils: An overview. *Aust. J. Exp. Agric.* 351: 42 (2002).
3. B. ESTRADA, R. AROCA, J.M. BAREA, J.M. RUIZ-LOZANO. Native arbuscular mycorrhizal fungi isolated from a saline habitat improved maize antioxidant systems and plant tolerance to salinity. *Plant Sci.* 42: 201 (2013).
4. P.G. JONES, P.K.THORNTON. The potential impacts of climate change on maize production in africa and latin america in 2055. *Glob. Environ. Chang.* 51: 13 (2003).
5. R. SHUKLA, CHERYAN, M. ZEIN. The industrial protein from corn. *Ind. Crops. Prod.* 171: 13 (2001).

6. C. BRERA, C. CATANO, B. DE SANTIS, F. DEBEGNACH, M. DE GIACOMO, E. PANNUNZI, M. MIRAGLIA. Effect of industrial processing on the distribution of aflatoxins and zearalenone in corn-milling fractions. *J. Agric. Food Chem.* 5014: 54 (2006).

7. M. ASIEDU, R. NILSEN, Ø. LIE, E. LIED. Effect of processing (sprouting and/or fermentation) on sorghum and maize. I: Proximate composition, minerals and fatty acids. *Food Chem.* 35: 46 (1993).

8. A. WALTER, G. RJMBACH, E. MOST, J. PALLAUF. Effect of citric acid supplements to a maize-soya diet on the in vitro availability of minerals, trace elements, and heavy metals. *J. VET. MED. A.* 517: 45 (1998).

9. S.B. GOLDHABER, Trace element risk assessment: Essentiality vs. Toxicity. *REGUL. TOXICOL. PHARM.* 232: 38 (2003).

10. C. MILLS. Dietary interactions involving the trace elements. *ANNU. REV. NUTR.* 173: 5 (1985).

11. A. KABATA-PENDIAS, *Trace elements in soils and plants*. CRC press: 2010.

12. M. OLIVARES, R. UAUY. Copper as an essential nutrient1, 2. *AM. J. CLIN. NUTR.* 791: 63 (1996).

13. I.-M. CHUNG, J.-K. KIM, J.-K. LEE, S.-H. KIM, Discrimination of geographical origin of rice (oryza sativa l.) by multielement analysis using inductively coupled plasma atomic emission spectroscopy and multivariate analysis. *J. CEREAL. SCI.* 252: 65 (2015).

14. A.B. HASSAN, G.A. OSMAN, M.A. RUSHDI, M.M. ELTAYEB, E. DIAB. Effect of gamma irradiation on the nutritional quality of maize cultivars (zea mays) and sorghum (sorghum bicolor) grains. *PAK. J. NUTR.* 167: 8 (2009).

15. I. ULLAH, M. ALI, A. FAROOQI. Chemical and nutritional properties of some maize (zea mays L.) varieties grown in nwfp, pakistan. *PAK. J. NUTR.* 1113: 9 (2010).

16. B. FEIL, S. MOSER, S. JAMPATONG, P. STAMP. Mineral composition of the grains of tropical maize varieties as affected by pre-anthesis drought and rate of nitrogen fertilization. *Crop Sci.* 516: 45 (2005).

17. P. PFAHLER, H. LINSKENS. Ash percentage and mineral content of maize (zea mays L.) pollen and style. *THEOR. APPL. GENET.* 32: 45 (1974).

18. E. SANTOS, D. LAURIA, C.P. DA SILVEIRA. Assessment of daily intake of trace elements due to consumption of foodstuffs by adult inhabitants of rio de janeiro city. *Sci. Total Environ.* 69: 327 (2004).

19. A. SZYMCZYCHA-MADEJA. Rapid method of element determination in rye crispbread by icp oes. *ARAB. J. CHEM.* 3913: 10 (2014).

20. C. CUADRADO, J. KUMPULAINEN, A. CARBAJAL, O. MOREIRAS. Cereals contribution to the total dietary intake of heavy metals in madrid, spain. *J. FOOD COMPOS. ANAL.* 495: 13 (2000).

21. B. DEMIRÖZÜ, I. SALDAMLI, B. GÜRSEL, A. UÇAK, F. ÇETINYOKUŞ, N. YÜZBAŞI, Determination of some metals which are important for food quality control in bread. *J. CEREAL. SCI.* 171: 37 (2003).

22. R.B. KHOUZAM, R. LOBINSKI, P. POHL, Multi-element analysis of bread, cheese, fruit and vegetables by double-focusing sector-field inductively coupled plasma mass spectrometry. *ANAL. METHODS.* 2115: 3 (2011).

23. H. ZHAO, B. GUO, Y. WEI, B. ZHANG. Multi-element composition of wheat grain and provenance soil and their potentialities as fingerprints of geographical origin. *J. CEREAL. SCI.* 391: 57 (2013).