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Determination of Iron, Zinc and Manganese in Nine Varieties of Wheat (*Triticum aestivum* L.) and Wheat Flour by using Atomic Absorption Spectrophotometer

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Abstract: Mineral elements compositions were estimated in local wheat and wheat flour samples by standard analytical methods coupled to the atomic absorption technique. The estimated significant quantities of three essential elements such as Iron (Fe), Zinc (Zn) and manganese (Mn) were determined by the atomic absorption spectrophotometer. The reference samples was obtained from the "Seed certification and Registration Department, Islamabad" as a reference material, while as representative samples were collected from different areas of Pakistan. In present work the essential elements iron, zinc and manganese were determined in different nine varieties of wheat grain and wheat flour, using atomic absorption spectrophotometer. It was observed that the level of Fe was high in Rohtas-90 (29.0-30.8) and low in Shahkar-91 (18.3-20.1) mg kg^{-1}. The level of Mn was high in Khyber-87 (41.8-48.4) and low in Rohtas-90 (25.0-27.4) mg kg^{-1}. The concentration of Zn was high in Khyber-87 (30.2-34.9) and low in Rohtas-90 (21.2-22.2) mg kg^{-1}.

Key words: *Triticum aestivum* L., minerals, atomic absorption technique

Introduction

Wheat *Triticum aestivum* L. is the world's most important crop, which provides carbohydrates, protein, vitamins and minerals. It is also staple food and constituents major portion of diet in Pakistan. It is obvious that all elements present in our body have been obtained mainly through food. So the daily intake of essential trace elements should be such that their lack does not lead to any disease . It will be necessary to study their presence in wheat, which is consumed by 98% population of Pakistan , because wheat alone can not meet the daily requirement of protein, thiamine and trace metals (Khan *et al.*, 1984). Trace elements are essential for the enzymatic processes of biological system (Katz *et al.*, 1980; Kazi *et al.*, 1997. These elements are made available to the human body by the vegetable kingdom and hence their presence is vital for the health of the body and for the cure of diseases. Historically, the increase in wheat yield as a result of genetic improvement efforts have largely been associated with improvements in harvest index (Sial *et at.*, 1988).

In Pakistan, the main emphasis is on yield, during breeding and production of wheat and unfortunately, very little attention is paid to its quality and nutritive value. Now a days many institute prepared different varieties of wheat through plant breeding, to enhance the quality and quantity of wheat. Minerals form a small part of the wheat, there are significant quantity of iron, manganese and zinc are present in different varieties of wheat. The amount of various elements making up plant tissue or seed vary with the species, the soil on which they grow and the fertility level of the soil. The role of these three elements are very important in plant and human beings. In human physiology the role of these elements are very important (Kazi *et al.*, 1999). The determination of trace and essential elements is therefore essential when working with new cultivars of wheat.

The aim and objective of the present research work was to determined the exact quantity in mg kg^{-1} of the iron, zinc and manganese within the wheat grain and wheat flour after the harvesting and compared with the permissible limit given by WHO.

Materials and Methods

Wheat samples: The different varieties were obtained from the "Federal Seed Certification and Registration Department Islamabad", as a reference sample and representative samples were collected from different areas of Punjab and NWFP.

Shahkar-91	(Punjab variety)	Pak-81	(Punjab)
Pasban-90	(Punjab)	Rohtas-90	(NWFP)
Faisalabad-85	(Punjab)	Pirsabak-85	(NWFP)
Punjab-85	(Punjab)	Khyber-87	(NWFP)
Inquilab-91	(Punjab)		

Reagents and calibration: The nitric acid (65% w/v) and hydrogen peroxide (35% w/v) were supra pure reagents (Merck), high-purity water (electrical resistivity >10m Ωcm) was produced with a Milli-Q system Millipore, MA, USA). Calibration was obtained with external standards. The standards solutions were prepared by diluting a 1000mg/l multi element solution (ICP Multi element standard IV, Merck, Darmstadt, FRG) with the same acid mixture used for sample dissolution. Glassware was cleaned by soaking with the contact over night in a 10% (w/v) nitric acid solution and then rinsed with deionized water.

Procedure: Sample digested with the HNO_3: 30% H_2O_2 (2:1) (Kazi *et al.*, 1999). Mineral elements analysis was carried out using atomic absorption spectrophotometer Hitachi Model 180-50 equipped with background correction and a data processor. All the parameters were set according to the manufacturer's instruction. iron, zinc and manganese hollow-cathode lamps (made by Mtiorika company) were operated at lamps current was set at 9.5. The flow-rate of acetylene for fuel 0.3, 0.2 and 0.2 kg/cm^2 and an air flow-rate 1.60 kg/cm^2 was used for Fe, Zn and Mn respectively to obtain a clear yellow flame (reducing condition). The spectrometer out put was connected to a Hitachi Model 180-50, (Hitachi Recorder 056) with a range of 5mV. The signals measured were the heights of the absorbance peaks. The detail is given in Table 1.

Table 1: Instrumental conditions for the AAS measurement of Fe, Zn, and Mn

Elements	Wave length (nm)	Slit width (nm)	Lamp current (mA)	Burner height (mm)	Oxidant (Air) kg/cm^2	Fuel kg/cm^2 (Acetylene) kg/cm^2	Signal out put
Fe	248.3	0.2	9.5	7.5	1.60	0.3	100%
Zn	214.0	1.3	9.5	7.5	=	0.2	=
Mn	279.8	0.4	=	=	=	=	=

Determination of mineral elements: Standard solutions of Fe, Mn and Zn were prepared 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 fitting of straight line by least square method (Sahito *et al.*, 2001). These three elements were determined in different wheat varieties of Pakistan.

Results and Discussion

Analysis of different wheat varieties cultivated in Pakistan other then the calculation of percentage concentration of various elements, reporting

Table 2: Statistical data for standards of elements

Elements	Concentration range ppm (x)	Absorbance	Statistical calculation y = m x + c: m	c	r^2
Fe	0 - 1	0 - 0.096	0.0976	-0.0016	0.9989
Zn	0 - 0.5	0 - 0.138	0.2761	-0.0004	0.9999
Mn	0 -1	0 - 0.196	0.1962	0.0005	0.999

varieties has been already reported in literature (Bhanger *et al.*, 1993). A blank reading was also taken and necessary correction were made during

Table 3: Range of concentrations of metallic residues found in different varieties of wheat and its flour (mg kg^{-1})

	Fe in grains		Fe in flour		Mn in grains		Mn in flour		Zn in grains		Zn in flour	
Wheat varieties	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.
Shahkar-91	18.3	20.1	18.9	20.1	26.8	29.8	27.4	31.0	31.4	32.4	32.7	33.7
Inquilab-91	21.9	23.6	22.5	24.8	35.8	37.6	35.8	37.6	22.2	26.0	23.1	25.7
Pasban-90	23.0	24.8	22.5	24.2	30.4	33.4	30.4	34.6	23.8	26.0	25.0	27.3
Punjab-85	21.9	22.5	21.8	23.0	37.0	40.0	37.0	39.4	23.1	25.8	22.8	25.7
Faisalabad-85	20.7	22.5	21.3	22.5	29.2	32.2	29.2	33.4	23.7	26.0	23.8	26.6
Pak-81	24.2	25.4	24.2	26.0	37.0	38.2	36.4	38.2	21.5	24.1	24.7	27.3
Rohtas-90	29.0	30.8	29.0	31.4	25.0	27.4	25.0	26.8	21.2	22.2	21.2	23.4
Khyber-87	27.2	28.4	27.2	28.4	41.8	48.4	43.6	47.8	30.2	34.9	29.8	35.0
Pirsabak-85	27.8	29.0	28.8	29.0	43.6	46.0	43.6	45.4	21.5	23.1	20.6	22.8

When wheat consumed in adequate amount, the alone wheat provide the daily requirement of thiamine niacin and more than half of the daily needs of essential element i.e., Iron, zinc and manganese. Established varieties of wheat viz Shahkar-91, Punjab (PB)-85, Pasban-90, Faisalabad-85, Rohtas-90, PAK-81, Khyber-87, Pirsabak (PS)-85 were analyzed for zinc, iron and manganese. The standards of Fe, Zn and Mn were prepared in the range of 0-1ppm and their absorbance were noted 0-0.096, 0-0.138 and 0-0.196 ppm for Fe, Zn and Mn respectively. The detail of statistical calculations of the m, c and r^2 are given in Table 2. It was observed that the level of iron was high in Rohtas-90 (29.0-30.8 and 29.0-31.4) mg kg^{-1}, while as low was found in Shahkar-91 (18.3-20.1 and 18.9-20.1) mg kg^{-1} in whole wheat grains and their flour respectively. The level of manganese was high in Khyber-87 (41.8-48.4 and 43.6-47.8) mg kg^{-1} and Pirsabak-85 (43.6-46.0 and 43.6-45.4) mg kg^{-1} as compared to other varieties of wheat an its flour, while as the minimum value was found in Rohtas-90 (25.0-27.4 and 25.0-26.8) mg kg^{-1} in both wheat grains and wheat flour, respectively. The maximum concentration of zinc was observed in Shahkar-91 (31.4-32.4 and 32.7-33.7) mg kg^{-1} wheat grains and their flour respectively. However, the level of manganese was found in Rohtas-90 (21.2-22.2 and 21.2-23.4) mg kg^{-1} and Pirsabak-85 (21.5-23.8,20.6-22.8)mg kg^{-1} very low as compared other varieties in wheat grains and their flour respectively. The values of individual metal and the varieties are summarized in Table 3. The genetic variation for mineral uptake was observed in the different varieties of wheat. The variation in mineral content is mainly attributed to different climatic conditions, culture practice and level in soil. From the analytical results given in Table 3, the observed concentration range for trace essential elements in studied wheat samples and their flour is as follows: Fe, 18.3-31.4; Mn, 25.0-48.4 and Zn, 20.6-35.0 mg kg^{-1}. The found range of Fe, Mn and Zn in these wheat varieties and their flour samples are generally tolerable for human consumption (Brown *et al.*, 1987). From the analytical result it is also clear that accuracy and precision is satisfactory. Heating time of 45 minutes in one step and 45 minutes in second step is sufficient for the digestion procedure developed for the analysis of Fe, Mn and Zn in wheat grains and wheat flour. The method requires glass beaker of a heating arrangement on electric hot plate for digestion the samples.

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