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IDENTIFICATION OF THE TRACE AND TOXIC ELEMENTS OF WHEAT CULTIVATED IN DIFFERENT REGIONS OF UZBEKISTAN

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Abstract
In Uzbekistan, the main food product is bread, which is produced in various forms from local and imported wheat. Most of it is not studied by microelement analysis. The objective of this study is to quantify trace elements in locally produced wheat from various cultivated regions of Uzbekistan. Four different varieties of wheat are cultivated in Sirdarya, Tashkent region, Kashkadarya and Fergana were selected for analysis. Next, a comparative analysis of the elemental composition like iron, zinc, potassium, calcium etc., in the selected samples were studied. To carry out the above aims, an innovative research method was chosen, which is called improved instrumental neutron activation analysis. This method differs from others in its high accuracy in determining trace elements and its multi-element nature. Using this method, we can understand how insufficient nutrients are in baked goods made from wheat grain grown in Uzbekistan. Based on the results of the study, it can be recommended to enrich the soil with fertilizers high in iron and zinc. As a result, in turn, will have a positive effect on the health of the population, since today there is a shortage of the above listed elements. It was also revealed that the concentration of potassium and calcium in wheat cultivated in Uzbekistan is in a high state than other elements. Using the chosen method, two toxic elements were determined and according to the analysis for toxic elements, all four samples are recommended for consumption, as they do not exceed the permissible level for food safety.
Key words: wheat, elemental analysis, food insecurity, neutron activation method, food supply.

Introduction

In Uzbekistan, the main food product is bread, which is produced in various forms from local and imported wheat. Most of it is not subject to microelement analysis. The objective of this study is to quantify trace elements in locally produced wheat grain samples from various cultivated regions of Uzbekistan.

Trace elements (TE) are chemical elements that occur in low concentrations in nature (El-Youssi *et al.*, 2023). The source of microelements for grain is soil and water, from which it is extracted through the root system and transferred to vegetative organs. Human-made activities can notably elevate the levels of trace elements in the environment, ultimately integrating them into the food chain. Heavy trace elements like cadmium, arsenic, lead, and mercury have no beneficial role in biological systems and can adversely affect human health. Consuming contaminated food is a common pathway for these toxic elements to infiltrate the human body.

Rashid Ben Akame and Naima Makhin reviewed data for presence of toxic elements in food products available in Mediterranean countries (El Youssi *et al.*, 2023). To determine trace elements, the instrumental neutron activation analysis (INAA) method was chosen. The results of this work proved the presence of high levels of toxic element contamination in vegetables and fish from some Mediterranean countries (El-Youssi *et al*., 2023).

In recent years, many authors around the world have reported several studies on the importance of the elemental composition of plant-based foods. The majority of these studies found that essential metals can lead to harmful effects when consumed in elevated amounts, whereas non-essential metals can be detrimental to human health even at minimal concentrations (Messaoudi & Begaa, 2018). Also, a decrease in the intake of essential metals in the human diet can, in turn, cause big problems both for the growing body and for senior people.

Plant foods can make a significant contribution to human nutrition and health as they contain almost all mineral and organic nutrients recognized as essential for human nutrition. Toxic elements are very important for food security and are defined as those that are present in food in quantities that may be potentially hazardous to human health. In turn, the lack of essential metals will negatively affect the vitality of the human body. Recommended Dietary Allowance is defined as intake level of essential nutrients that are considered sufficient to meet the known nutritional requirements of all healthy individuals based on scientific knowledge (Resolution, 1999). In addition, recommended intakes are classified according to the requirements for certain nutrients based on age group (e.g., infants, children, adolescents and adults), gender (male/female), and physiological needs (e.g., pregnancy, lactation) (Recommended Dietary Values) (Resolution, 1999).

Commercially available varieties of cereals are an important part of the diet of preschool children, schoolchildren and adults, due to their mineral and vitamin content that meets the dietary needs of these target groups. The presence of impurities such as arsenic and mercury in wheat grains can pose a health hazard to people, especially children, since their bodies are most vulnerable to various toxic substances. Using the recommended dietary allowance (RDA), it is possible to control the intake of essential metals and, in cases of deficiency, make adjustments to the diet (Ghuniem *et al*., 2020).

In their work, authors determined Co, Cr, Fe, Se and Zn in cassava from several cities in Java using the Nuclear Analytical Technique (NAT). Cassava

(*Manihot esculenta* Crantz) is a major food crop grown in subtropical and tropical regions (Kurniawati *et al*., 2021). In Indonesia, cassava ranks as the third most significant carbohydrate source following corn and rice. Hence, the researchers deemed it crucial to assess the nutritional content of cassava, focusing on its trace micronutrients that are indispensable for human health and pivotal in human metabolism. Instrumental neutron activation analysis (INAA), as one of the NAT methods, was used in this study due to its accuracy and sensitivity for the determination of trace elements. Counting and elemental measurements were carried out using an HPGe detector. Data analysis quality control was applied using SRM NIST (Standart Reference Materials) 1567a wheat flour and yielded good results with recovery percentages ranging from 94.2 to 113.5%. The concentration range of Co, Cr, Fe, Se and Zn was 0.002-0.098, 0.023-0.158, 1.65-18.9, 0.008-0.228 and 2.06-7.68 mg/kg, respectively (Kurniawati *et al*., 2021).

Microelement (Co, Cr, Cs, Fe, Sc, Se and Zn) composition of Algerian wheat was studied using instrumental neutron activation analysis (INAA). The results showed that the content of trace elements in the studied samples in the province of El Harrach was within the basic safety level of all analyzed elements recommended by the WHA/FAO, with the exception of Co (Beladel *et al*., 2022).

For monitoring and assessing the health risk of individual trace elements & heavy metals in wheat, rice and soil samples, a study was carried out to determine eight trace elements: cadmium, copper, cobalt, chromium, zinc, lead, nickel manganese in wheat, rice and soil samples collected from different cities of Punjab (India) (Hamid *et al*., 2020). The results showed a general tendency for the accumulation of trace elements & heavy metals in the samples in the form of Cd > Cu > Zn > Co > Mn. All wheat samples and nine rice samples exceeded the permissible limit for Cd content. Copper levels were high in eight wheat and rice samples, and Zn levels were high in three wheat and one rice samples. However, only one Multan soil sample exceeded the maximum permissible limit for copper content set by WHO in 2007 and EU 2000. The concentration of manganese and cobalt was within the permissible limits, and Ni, Cr and Pb were not detected in any sample. The health risk index was greater than 1 for Cu, Cd, Co and Mn, indicating a potential health risk to consumers (Hamid *et al*., 2020).

In the work (Mansouri *et al.*, 2021), zinc levels were measured in scalp hair and nails to identify potential risk factors for breast cancer. Zinc level was analyzed using instrumental neutron activation analysis (INAA). In scalp hair samples, it was found in the range of 119–792 $\mu g\ g^{-1}$ in healthy individuals and in the range of 82–806 $\mu g\ g^{-1}$ in patients.

A research was undertaken to examine the microelement profile of Fe, Zn, Mn, Cr and Co in food products frequently eaten by the people (Mulyaningsih *et al*., 2021). Sampling was conducted at four traditional markets located in Pandeglang, Cianjur, Magelang, and Bangkalan. The samples collected encompassed dietary items such as carbohydrates, vegetables, fish, legumes, tofu, and tempeh. Prior to analyzing their trace mineral content, the food samples underwent preparation using neutron activation analysis techniques (Mulyaningsih *et al*., 2021).

Determination of Fe in food products was performed using instrumental neutron activation analysis (INAA). In this study, iron content of 73 local foods from Mamuju District, West Sulawesi Province was determined. The results showed that green vegetables such as kangkong (*Ipomoea Aquatica* Forsk), bayam (*Amaranthus* spp), kemangi (*Ocimum citriodorum*), daun pakis (*Diplazium esculentum*), daun katuk (*Sauropus androgynous*), daun seledri (*Apium graveolens*), daun singkong (cassava leaves), cesim (*Brassica rapa*) and buncis (*Phaseolus vulgaris*) have an elemental content of Fe > 100 mg/kg. Kangkung, bayam, and kemangi are green vegetables that are rich in the element Fe, the concentration of Fe in the vegetables is more than 280 mg kg^{-1}. Concentrations of the element Fe in vegetables are higher compared to ones in beef or other protein sources (Yusuf *et al*., 2019).

Materials and Methods

Materials: To assess the microelement composition, samples were collected from 4 regions, with different geographical, soil, climatic and sea conditions. The selected regions are: Sirdarya, Kashkadarya, Tashkent and Fergana regions.

The wheat variety 'Asr' was selected from Sirdarya, the plant height is 95-105 cm, and it is resistant to lodging. The head is cylindrical, large. Plowing the land was turned to the axis. Potassium and phosphorus fertilizers were added to the soil. During the growing season, wheat was abundantly treated with nitrogen for further accumulation in leaves and stems. Humidity during the wheat growing season was 70-75%. During storage, the temperature was kept no lower than +10 and no higher than +15 degrees, and the humidity of the grain itself was 14%. Harvest was in 2023.

The 'Turon' wheat variety was selected from Kashkadarya. Plowing the land was turned to the axis. Potassium and phosphorus fertilizers were added to the soil. During the growing season, wheat was abundantly treated with nitrogen. Humidity during the wheat growing season was 70-75%. During storage, the temperature was kept no lower than +10 and no higher than +15 degrees, and the humidity of the grain itself was 14%. Harvest was in 2023.

The wheat variety 'Alekseich' was selected from the Tashkent region. The wheat is mid-season, frost-resistant. A yield of 40 centners per hectare was obtained. The advantage of the variety should be considered its resistance to many diseases.

It is semi-dwarf, the plant height is 81 cm, and it is highly resistant to lodging. Seeding rate is 5 million viable seeds per hectare. Harvest was in 2023. The soil was fertilized with potassium and phosphorus. During the growing season, wheat was abundantly treated with nitrogen and potassium. Humidity during the wheat growing season was 70-75%. During storage, the temperature was kept no lower than +10 and no higher than +15 degrees, and the humidity of the grain itself was 14.2%.

The variety 'Alekseich' was selected from Fergana. Plowing the land was turned to the axis. Potassium and phosphorus fertilizers were added to the soil. During the growing season, wheat was abundantly treated with nitrogen and potassium. Humidity during the wheat growing season was 70-75%. During storage, the temperature was kept no lower than +10 and no higher than +15 degrees, and the humidity of the grain itself was 14.1%. Harvest was in 2023.

Equipment: The measuring equipment for instrumental neutron activation analysis consists of a WWR-SM nuclear reactor of the Institute of Nuclear Physics of the Academy of Sciences of Uzbekistan, a semiconductor detector made of pure germanium, and a multichannel pulse analyzer. For the analysis, IAEA international standard samples were used - IAEA-36 Lichen, NIST SRFM 1572-Citrus leaves, produced by the US Institute of Standards and Technology.

Software: Gamma spectra measured using a semiconductor germanium detector are processed with the Genie-2000 computer program.

Methods:

Iron, potassium, zinc, calcium and etc. elements concentrations were calculated using instrumental methods of neutron activation analysis. Analytical work to determine the trace element composition of selected wheat samples was carried out in the Ecology and Biotechnology laboratory of the Institute of Nuclear Physics of the Academy of Sciences of the Republic of Uzbekistan (INP AS Uz). The neutron sources were a research atomic reactor of the WWR-SM type at the INP AS Uz. In this study, trace and toxic elements in four wheat varieties cultivated in different regions of Uzbekistan were identified by using instrumental neutron activation analysis (INAA).

The selected samples were dried in a special drying cabinet at a temperature of 60˚C to constant weight and hermetically packaged in a plastic bag, then placed in a special aluminum container for irradiation with a neutron flux with a flux density of $3 \cdot 10^{13}$ neutron·cm^{-2}·s^{-1}. Based on the gamma spectrum of radionuclides that are formed in the samples under study during irradiation with a neutron flux, the content of microelements present in these samples was determined. The measurement of the spectra of gamma radiation emitted from the sample under study was carried out using a gamma spectrometer based on a high-resolution semiconductor detector. The concentrations of all elements in wheat were calculated using INAA- in comparison with reference samples. In this study, we used standard (SRM) material for the quality control/QA method of the INAA (Bode & Blaauw, 2011).

Results and Discussion

The risk associated with the intake of toxic elements from food was assessed by taking into account consumer exposure and threshold values, such as the lower confidence limit of the reference dose and the preliminary tolerable weekly intake established by the resolution of the Cabinet of Ministers of the Republic of Uzbekistan 'On approval of the General Technical Regulations on the Safety of Grain' (Resolution, 1999) food additives, respectively.

Table 1 shows the experimental results on the neutron activation determination of toxic elements: arsenic and mercury and comparison with the permissible levels of these elements in food products (Resolution, 1999).

Table 1

Experimental results on neutron activation determination of toxic elements: arsenic and mercury and comparison with permissible levels of these elements in food products

Indicators	Arsenic, As	Mercury, Hg
'Alekseich' Tashkent region	<0.01 mg/kg	<0.001 mg/kg
'Asr' Sirdarya	<0.01 mg/kg	<0.001 mg/kg
'Turon' Kashkadarya	<0.01 mg/kg	0.0053 mg/kg
'Alekseich' Fergana	<0.01 mg/kg	<0.001 mg/kg
Permissible levels, mg/kg, no more	0.2 mg/kg	0.03 mg/kg

All selected grains were results below the recommended tolerable levels and acceptable guideline values. The results were compared with the maximum permissible rate of daily intake of toxic elements and identified as being below the permissible limits.

Table 2 shows the results of instrumental neutron activation analysis of wheat grain samples.

Table 2

Neutron activation analysis of wheat grain samples mg/kg

Ele-ments	'Alekseich' Tashkent region	'Asr' Sir-darya	'Turon' Kashka-darya	'Alekseich' Fergana
Cu	3.4	3.9	<1.0	<1.0
Mn	20.6	36.4	37.2	26.5
Na	17.6	17	21	23.8
K	3400	3600	3560	4500
Br	0.59	0.22	0.19	0.27
Ca	390	490	410	470
Zn	24.5	47.9	24.4	29.8
Fe	41.4	44.6	49.9	42.2
Se	<0.01	0.066	0.047	0.13

Eleven elements were assessed and we found three categories: major, minor, and traces of elements that existed in all samples.

The results of the analysis showed that the concentration of copper in wheat growing in the Tashkent region and Sirdarya is much higher than in the other two samples. And the content of the element manganese is significantly lower in wheat growing in the Tashkent region and Fergana, and the Asr and Turon grains predominate in this element. Sodium concentration values vary slightly among regions.

The potassium content in three wheat samples Asr, Alekseich and Turon almost does not change its value, and the Alekseich wheat variety grown in the Fergana region contains potassium by 27.8% more than the average value of this element than in other regions of Uzbekistan.

Calcium is contained in large quantities in the samples Asr, cultivated in Sirdarya and Alekseich, cultivated in Fergana; regarding samples from the Tashkent region and Kashkadarya, it may be seen that the calcium content is 100 mg kg^{-1} less. A very important element for human life, zinc is found in a stable value in the samples of the Tashkent region, Kashkadarya and Fergana, and only the ‘Asr’ sample from Sirdarya has a double value of zinc. The content of iron and selenium in all samples is almost the same.

Table 2 shows the difference between the values of zinc and iron.

Despite the fact that in the Republic of Uzbekistan flour is enriched with B group vitamins and microelements iron and zinc, there is a deficiency of essential elements. And the lack of essential elements such as iron in the human body can cause growth retardation in children, iron anemia in pregnant women, lactating women and children under two years of age (Ursova, 2015). This information may be useful in preventing growth retardation, especially growth retardation caused by glandular deficiency (Erin Farah et. Aa.). Selenium has a positive impact on psoriasis. Selenium is crucial for safeguarding against free radical damage due to its presence in the glutathione peroxidase enzyme.

In Table 2, a large difference between the two main elements potassium and calcium is displayed. Compared to other elements, they exceed the daily human need for these elements.

Potassium and calcium perform very important functions in the human body. For example, calcium helps in the formation of bones and teeth in young children. And potassium is an auxiliary element of the cardiovascular system.

Conclusions

1. Based on the results in the studied wheat, it was established that the concentration of toxic elements such as arsenic and mercury does not exceed the maximum permissible concentrations prescribed by standards.
2. The data obtained in this study complements the database on the microelement composition of grain varieties grown in Uzbekistan and allows for preferred varieties for cultivation. Thus, it is possible to select wheat varieties richer in biological components.
3. Based on the experimental results obtained, we can conclude that the Alekseich wheat variety, cultivated in the Tashkent region, is the poorest in microelements such as iron, zinc and calcium. Based on this, it is recommended to use fertilizers containing these elements to compensate the deficiency of these microelements.

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