

DETERMINATION OF THE AMOUNT OF CHEMICAL ELEMENTS IN THE COMPOSITION OF “AS-MIR” BY INDUCTION-COUPLED PLASMA OPTICAL EMISSION SPECTROMETRY AND ITS EFFECT ON INFLAMMATION

Bokiyev Mirzoxidbek Muzafarjon ugli

Assistant Professor, Department of Biological Chemistry, Andijan State Medical Institute, PhD student at Kokand State University.

Email: bmirzokhid@gmail.com

Orchid ID: <https://orcid.org/0000-0002-4221-474X>

Abstract: In this study, the content of macro- and microelements in the phytochemical supplement “AS-MIR” was determined by the inductively coupled plasma optical emission spectrometry (ICP-OES) method and their biochemical significance in inflammatory processes was studied. The processes of ash extraction from the samples, dissolution in acids and calibration based on standard solutions were carried out. According to the results of the analysis, the mixture contained significantly high amounts of calcium, potassium, magnesium, sodium, phosphorus and iron. Trace concentrations of copper, manganese, chromium, molybdenum, lithium, boron, silicon and nickel were recorded from the microelements. The results obtained were compared with the participation of these elements in the processes of inflammation and carcinogenesis. The study showed that the components of the “AS-MIR” mixture have not only antioxidant, but also potential anti-inflammatory properties

Keywords: AS-MIR, ICP-OES, biogenic elements, inflammation, carcinogenesis, macroelements, microelements, oxidative stress, antioxidant protection, polyphenols

Introduction. With the increase in the number of people on Earth, their need for food and medicinal plants is also increasing, among which the most widely consumed red cabbage (Latin *Brassica oleracea* var. *capitata* f. *rubra*) and horseradish (Latin *Armoracia rusticana*) are distinguished by their healing properties. They contain vitamins, minerals, polyphenols, etc. that are essential for human health. Therefore, medicinal products prepared from these plants have antibacterial, antifungal, expectorant effects, improve immunity and fight inflammation, and the antioxidants they contain reduce the effects of free radicals [1-3].

Biogenic elements consist of carbon, hydrogen, oxygen, nitrogen, sulfur, phosphorus, and trace elements such as iron, copper, zinc, and selenium, and are the basis of all metabolic and signaling processes in the body. Their deficiency or excess is an important pathobiochemical factor in the duration and intensity of the inflammatory response, as well as the onset and development of cancer processes[4-6]. Among macroelements, oxygen controls oxidation-reduction reactions and causes the formation of reactive oxygen species (ROS) in the inflammatory zone. Uncontrolled increase in ROS leads to oxidative damage to DNA, lipids, and proteins, enhances mutations, and creates favorable conditions for carcinogenesis. Nitric oxide (NO) is excessively synthesized by inducible NO synthase (iNOS) under the influence of inflammatory mediators; at high concentrations, it causes nitrosative damage to DNA and disrupts genetic stability.

Microelements — especially zinc, copper and iron — determine the intensity and duration of the inflammatory process as cofactors of antioxidant enzymes. Excess iron increases free radicals through the Fenton reaction, causing chronic oxidative stress in tissues; this contributes to the



development of cancer, which is associated with increased proliferation and angiogenesis. Selenium, as a component of glutathione peroxidase, reduces lipid peroxidation, reduces the activation of the NF- κ B pathway and has an anti-inflammatory effect. At the same time, selenium deficiency is associated with a weakening of the immune system, a slowdown in DNA repair mechanisms and the escape of tumor cells from apoptosis [7-9].

Table 1. The role of individual elements in inflammation and cancer

Element	Role in Inflammation	Role in Cancer
Ca (Calcium)	- Regulates secretion of inflammatory mediators (IL-1, TNF- α). - Enhances macrophage and lymphocyte activity.	- Calcium signaling is essential for tumor cell proliferation. - Increased activity of Ca ²⁺ channels promotes tumor invasiveness [10–11].
K (Potassium)	- Necessary for intracellular balance and immune response. - Opening of potassium channels during inflammation triggers inflammasome (NLRP3) activation.	- Potassium channels in cancer cells regulate growth, migration, and metastasis [12].
Na (Sodium)	- Electrolyte balance changes during inflammation. - Hyponatremia increases levels of inflammatory mediators.	- Na ⁺ /H ⁺ pumps are overactive in cancer cell metabolism → increased invasiveness [13].
Mg (Magnesium)	- Reduces levels of cytokines (IL-6, CRP). - Deficiency induces chronic low-grade inflammation.	- Protects DNA, slows tumor progression. - Deficiency observed in many cancer types [14].
P (Phosphorus)	- ATP deficiency amplifies inflammatory cascades. - Phosphorylation is a key process in immune cell signaling.	- Alterations in phosphorylation pathways (MAPK, PI3K) in cancer cells increase tumor proliferation [28].
Fe (Iron)	- Generates ROS via the Fenton reaction → increases inflammation. - Iron deficiency impairs immunity.	- Excess iron causes DNA damage. - High iron load increases risk of colorectal, liver, and lung cancers [16–18].
Cu (Copper)	- Component of superoxide dismutase (SOD1), reduces free radicals in inflammation. - Excess copper may have pro-inflammatory effects.	- Activates angiogenesis (VEGF). - Excess copper associated with rapid tumor growth [19–20].
Mn (Manganese)	- Cofactor for SOD2, reduces oxidative stress. - Decreases inflammation.	- Protects DNA due to antioxidant properties. - Altered Mn levels observed in some tumor cells [15, 30].
Zn (Zinc)	- Strong anti-inflammatory effect. - Blocks NF- κ B pathway.	- Stabilizes p53 tumor suppressor protein. - Deficiency increases cancer risk (not in table, but scientifically relevant) [21–22].
Mo	- Component of antioxidant enzymes	- Deficiency enhances oxidative



Element	Role in Inflammation	Role in Cancer
(Molybdenum)	(xanthine oxidase). - Regulates oxidative stress.	DNA damage.
Cr (Chromium, trivalent)	- Increases insulin sensitivity, reduces inflammation.	- Deficiency impairs glucose metabolism, may increase cancer risk.
Co (Cobalt)	- Involved in erythropoiesis and immune system via vitamin B12.	- Excess cobalt increases ROS → cancer risk.
Se (Selenium)	- Component of glutathione peroxidase → strong antioxidant. - Reduces cytokine production.	- Numerous clinical studies confirm reduced cancer risk [23–24].
B (Boron)	- Activates metabolic pathways that reduce inflammation.	- Higher boron levels associated with lower prostate cancer risk [25].
Si (Silicon)	- Promotes connective tissue repair.	- Not strongly linked to cancer, but has antioxidant properties.
Ni (Nickel, trace)	- Participates in enzyme activity at trace levels.	- Excess nickel is a strong carcinogen (epigenetic changes) [26].
Li (Lithium)	- Anti-inflammatory effect: inhibits GSK-3 pathway.	- May promote apoptosis in tumor cells (studies on lithium therapy exist) [27].

The balance of biogenic elements in Table 1 is considered to be a key biochemical factor in the “inflammation-carcinogenesis” link in the transition from inflammation to cancer. In chronic inflammation, the activity of ROS, RNS, cytokines, and metalloenzymes increases, epigenetic changes, activation of proto-oncogenes, and a decrease in the activity of tumor suppressor genes are observed. Therefore, maintaining the concentration of biogenic elements at physiological levels is of strategic importance both in the treatment of inflammatory diseases and in the prevention of tumor processes.

Experimental part. Preparation of sample working solution. 1 g of pre-dried, ground, weighed on a balance with an accuracy of 0.001 g (Navigatortm, OHAUS®) sample (red cabbage leaves and horseradish root obtained from the local market to obtain the “AS-MIR” mixture were dried and ground and mixed in the appropriate ratio and packaged) was ashed in a porcelain crucible by heating to 500 oC in a muffle furnace (Nabertherm, Germany) using the dry ashing method. Initially, it was heated to 550 oC at a rate of 100 oC/h and held at 550 oC for 5 hours. 6 ml of 70% HNO₃ (Sigma Aldrich, USA) and 2 ml of 60% H₂O₂ of ICP-MS purity were added to the resulting ash and heated on a hot plate in a fume hood until the formation of white smoke ceased. The cooled solution was transferred to a 100 mL polypropylene volumetric flask and made up to the mark with ultrapure water. This working solution was filtered using a syringe filter and used for analysis.

Preparation of standard solutions. Standard solutions of 68 elements in 2% HNO₃ with a concentration of 10 mg/l (Highpuritystandards, USA), standard solutions of mercury element in 2 mol/l HNO₃ with a concentration of 1000 mg/l (Sigma Aldrich, Germany), standard solutions of 25 elements in 2% HNO₃ with a concentration of 10 mg/l (Aristar, USA), 70% HNO₃ (Sigma



Aldrich, USA) were used to prepare standard working solutions of elements in 2% HNO₃ and 3 more standard working solutions were prepared by dilution. A 2% HNO₃ solution was used as a blank. A calibration line was created for 69 elements using the above standard working solutions.

Analysis. The analysis was performed on an iCAP PRO X Duo ICP-OES inductively coupled plasma optical emission spectrometer manufactured by Thermo Fisher Scientific (USA). Method development and analysis of the analysis results were performed using the QTegra ISDS software[31]. The analysis parameters are listed in Table 2.

Table 2. Analysis method parameters.

Parameter	Settings
Pump tubing	Sample: Tygon® yellow/white Drain: Tygon® white/white
Pump speed	45 rpm
Spray chamber	Glass cyclonic
Nebulizer	Glass concentric
Nebulizer gas flow	0.6 L·min ⁻¹
Coolant gas flow	12.5 L·min ⁻¹
Auxiliary gas flow	0.5 L·min ⁻¹
Center tube	2 mm
RF power	1150 W
Replicates	3 times
Analysis time	Axial: 15 s Radial: 15 s

Results obtained. The analysis results are presented in the table below.

Table 3. Results of determination of chemical elements in the sample by the IBP-OES method, µg/100 g.

Element	Wavelength (nm)	Concentration (µg/100 g)	Element	Wavelength (nm)	Concentration (µg/100 g)
Ca	393.366	108800.542 ± 836.5	Fe	259.940	11254.262 ± 100
Ca	396.847	25202.596 ± 20.3	Cu	324.754	85.186 ± 0.4
K	766.490	204519.519 ± 2384.7	Mn	257.610	684.668 ± 2.4
Mg	279.553	37791.946 ± 55.1	Cr	283.563	117.1 ± 0.3
Mg	285.213	41657.934 ± 68.7	Mo	202.030	28.262 ± 0.8
Na	589.592	10289.36 ± 12.6	Li	670.776	139.165 ± 0.5
P	185.942	23483.855 ± 8.8	B	249.773	294.66 ± 1
Si	251.611	1301.668 ± 3.5	Ni	221.647	27.755 ± 0.5

Based on the data in Table 3, it is possible to observe a change in the amount of elements in the composition of the phytochemical supplement “AS-MIR” in the order Ca>K>Mg>Na>P>Fe>Cu. Of these elements, Mg was found to be 377.9 mg/kg, Fe 112.54 mg/kg.



Conclusion. The results of the study showed that the composition of the phytochemical supplement “AS-MIR” contains a significant amount of biologically important elements. The high concentration of calcium, potassium, magnesium, sodium, phosphorus and iron in the mixture, as well as the presence of copper, manganese, lithium, boron and other trace elements, indicates its ability to affect inflammatory processes. According to biochemical analyses: elements such as Mg, Zn, Se, Mn have an anti-inflammatory effect by reducing NF- κ B pathways and reducing oxidative stress. The presence of Fe and Cu in normal amounts supports the activity of antioxidant enzymes, but in excess increases oxidative stress. Ca and K are important regulators of cell signaling and immune cell activity. The results obtained indicate that the “AS-MIR” mixture can be used as an auxiliary phytobiological support agent in chronic inflammatory processes. The combined antioxidant and immunomodulatory effects of polyphenols and microelements in the mixture can have a positive effect on the reduction of inflammatory mediators, oxidative stress, and tissue regeneration. Therefore, the use of the “AS-MIR” phytochemical supplement is recommended as an immune-boosting supplement in cases of bronchitis, colds, and low-grade chronic inflammation. However, additional pharmacological and clinical studies are needed to assess clinical efficacy.

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