**Optical Emission Spectroscopy of Elemental Composition and Bioactivity of As-Razzoq**

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**Abstract.** In this study, the macro- and microelement composition of the food supplement “AS-RAZZOQ” was determined using an inductively coupled plasma optical emission spectrometer (iCAP PRO X Duo ICP-OES) and the antiradical properties were determined using the DPPH method. In order to analyse Antimicrobial properties we used modified agar diffusion method against Staphylococcus aureus and Pseudomonas spp. using a with Buffered Peptone Water (BPW) as the nutrient medium, and the resulting inhibition zones were quantitatively analyzed. Additionally, mucolytic activity was examined by monitoring viscosity variations in sputum samples in vitro.

**INTRODUCTION**

Natural food supplements are an essential source of amino acids, fatty acids, vitamins, minerals, other necessary biologically active substances for the human body. They are mainly obtained from medicinal plants, fruits and vegetables, cereals and other natural raw materials. The main importance of such supplements is that they replenish nutrients that are insufficient in the daily diet, strengthen immunity, normalize metabolism and improve overall human health.

Rosa canina L. Is a plant belonging to the Rosaceae family, that includes more than 100 varieties, distributed in the Middle East, Europe, Africa, Asia, and North America [1]. This plant, also popular as briar rose or dog rose, it also resistant to severe weather conditions [2]. It has pseudo-fruits [3] that ripen in August-September [4] and are dark red also brick-red colors [5]. It is a rich resource from a medical and economic point of view [2]. It grows in walnut groves in Tashkent, Samarkand, Fergana, Andijan, Syrdarya, Jizzakh, Namangan, Surkhandarya Kashkadarya, regions [6]. Therefore, the plant has a complete reserve, and its fruit is especially used in medicine as a natural source of medicinal properties [7].

Glycyrrhiza Glabra is a plant belongs to the Leguminosae family and known as licorice. The stem is branched, rough, 40-150 cm tall, and grows upright. The leaves are complex, pinnate, and oblong. The flowers are purple, collected in a raceme. The fruit is an oblong pod. It blooms in April-June. It reproduces by seed and vegetatively from rhizomes. The root system is well-developed, penetrating up to 3 meters deep. Above-ground shoots are formed from the rhizomes. The seeds have a hard shell and germinate at a temperature of 30-35 °C [8]. It is spread among all regions of Central Asia. The first three regions have been considered the original areas of licorice cultivation in China [9]. Glycyrrhiza Glabra is also strewn across in Central Asia and Europe. [10, 11]

Biologically active compounds have been identified primarily as secondary metabolites and their derivatives, such as alkaloids [12], glycosides [13], flavonoids [14], phenolic compounds [15], saponins [12], tannins [14], terpenes [16], anthraquinones [17], essential oils [16], and steroids [18]. Chemical elements are the most important catalysts of various biochemical reactions, important and irreplaceable participants in the processes of growth and development of the organism, metabolism, adaptation to changing environmental conditions. It is literally known that biogenic elements in the human body have synergistic and antagonistic relationships. There are 105 bilateral and 455 triple interactions between the 15 known essential elements.All macro- and microelements have a significant effect on organisms to varying degrees and at different stages of their life cycle. They affect growth and development, the processes of fertilisation, respiration, hematopoiesis, immunogenesis, in short, the functioning of all morpho-physiological systems of the body. This interaction is carried out in the process of digestion, as well as in the process of tissue and cell metabolism, and again in the food itself. The interaction occurs depending on the type of synergism (joint action) or antagonism (resistance) between the elements [19]. The following table shows the synergistic and antagonistic effects of some important chemical elements:

**TABLE 1.** Interaction of Elements in the Human Body

|  |  |
| --- | --- |
| **Excess of element content** | **Element deficiency occurs** |
| **Mercury (Hg)** | Selenium (Se) |
| **Arsenic (As)** | Selenium (Se) |
| **Cadmium (Cd)** | Selenium (Se), Soul (Zn) |
| **Calcium (Ca)** | Zinc (Zn), (Pb) |
| **Ferrum (Fe)** | Copper (Cu), Zinc |
| **Manganese (Mn** | Mg, Copper (Cu) |
| **Molybdenum (Mo)** | Copper (Cu) |
| **Zn (Zn)** | Copper (Cu), Iron (Fe) |
| **Lead (Pb)** | Calcium (Ca), Soul (Zn) |
| **Copper (Cu)** | Zinc (Zn), molybdenum (Mo) |

Chemical elements are distributed and accumulated in different amounts among organs and tissues in the human body. Macro and microelements are unevenly distributed between different organs and tissues. The highest concentration of chemical elements is found in bone tissue, skin and its appendages, liver and muscles [20].

**METHODS AND MATERIALS**

Evaluation of the Antiradical Properties of the Sample Using the DPPH Method Rosa canina L. (rose hip) fruit and Glycyrrhiza glabra L. (licorice) root were break down to a particle variety of 0.1 to 1 mm, then three different samples were prepared from these materials.

The ratios of the plant used in every sample are provided in following table.

**TABLE 2.** Ratios of plant parts in preparation.

|  |  |  |
| --- | --- | --- |
| **Samples** | **Ratio of parts of plants, %** | |
| ***Rosa canina* L fruit** | ***Glycyrrhiza glabra* L root** |
| **№ 1** | 1 | 3 |
| **№ 2** | 3 | 1 |
| **№ 3** | 1 | 1 |

Extracts prepared in two different ways.

1. Preparation of hydrous extract. 25 ml of water 10 minutes in a conical flask which equipped with a reflux condenser. The gained extract was filtered using a 0.45 μm syringe filter.

2. Preparation of alcoholic extract. 25 ml of 96% ethanol was taken and one gramm of plant sample was put in ultrasonic extraxtor for 20 minutes at 60 °C. The gained extract was filtered using a 0.45 μm syringe filter.

The antiradical properties of the samples were calculated using the following formula:

(1)

The content of macroelements and microelements of the extract was analyzed by the dry ashing method of 1 g of the sample, previously dried, ground, and measured on a balance with an exact accuracy of 0.001 g (Navigator, OHAUS), and heated to 500 °C in a muffle oven (Nabertherm, Germany). Initially, it was heated up to 550 °C, for 100 oC/h and held at 550 °C for five hours. 6 ml of 70% nitric acid (Sigma Aldrich, USA) and 60% H2O2 of 2 ml of ICP-MS purity were added.

The analysis was performed according the following order. For this purpose, the iCAP PRO X Duo ICP-OES manufactured by Thermo Fisher Scientific (USA), The analysis parameters are listed in Table 3.

Buffered Peptone Water (BPW) was chosen as the nutrient medium. Staphylococcus aureus and Pseudomonas spp. Suspensions were prepared in physiological saline and added to 250 ml vials with BPW. 20 ml of nutrient media, cooled to 45°C, were poured into Petri dishes. 0.5 ml of “AS-RAZZOQ” food additive and standard were put to the upper surface. For 24 hours at 35°C dishes were incubated. The results were evaluated by zone diameter using a calliper [21-25].

**TABLE 3.** Analysis method parameters

|  |  |  |
| --- | --- | --- |
| **Parameters** | **Settings** | |
| **Pump tube** | Tygon® yellow/white for sample | Tygon ® white/white for drainage |
| **Speed of the pump** | 45 Rounds per minute | |
| **Spray chamber** | The cyclonic glass | |
| **Nebulizer** | The concentric glass | |
| **Coolant gas flow** | 12.5 L·min-1 | |
| **Nebulizer gas flow** | 0.6 L·min-1 | |
| **Auxiliary gas flow** | 0.5 L·min-1 | |
| **Center tube** | 2 mm | |
| **power of RF** | 1150W | |
| **Repeatability** | x3 | |
| **Analysis time** | Axial | Radial |
| 15 seconds | 15 seconds |

To research themucolytic effect of the food additive "AS-RAZZOQ" on the viscosity of sputum from patients with acute or chronic bronchitis, samples were collected. Sputum samples were collected in a 20 ml vial and kept in a water bath at 37°C for 60 minutes. Then, the viscosity was measured for 5 minutes at a speed of 60 rpm using a rotational viscometer (ATAGO VISCO 6800 (Russia)). After the measurement, 60 mg of the food additive "AS-RAZZOQ" in powder form was added to the sample, mixed, and hold at 37°C xin a water bath for 60 minutes, and the viscosity of the sample was measured using a rotational viscometer.

**RESULTS AND DISCUSSION**

To calculate the IC50 of the samples - the 50% inhibition concentration of solution of the DPPH, the following table was compiled based on the values of absorbance (D2) and antiradical activity (ARA%) in each experiment at 30 minutes and calculated based on the line of trend function applied to it.

**FIGURE 1.** Graph of the sample A determined at 10 minutes of relationship between ARA% and volumes

y=mx+b formula was used to calculate ARA% (IC50):

(2)

**TABLE 4.** ARA% values of samples with different ratios

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Ratio** | **Volume, μl** | **Aqueous** | | **Alcoholic** | |
| **D2** | **ARA, %** | **D2** | **ARA, %** |
| **1:1** | 25 | 0,764 | 10,85 | 0,5 | 41,25 |
| 50 | 0,645 | 24,74 | 0,221 | 74,03 |
| 75 | 0,588 | 31,39 | 0,187 | 78,03 |
| 100 | 0,536 | 37,46 | 0,096 | 88,72 |
| **1:3** | 25 | 0,735 | 14,24 | 0,24 | 72,00 |
| 50 | 0,59 | 31,16 | 0,178 | 79,23 |
| 75 | 0,47 | 45,16 | 0,096 | 88,80 |
| 100 | 0,326 | 61,96 | 0,034 | 96,03 |
| **3:1** | 25 | 0,724 | 15,52 | 0,223 | 73,98 |
| 50 | 0,672 | 21,59 | 0,166 | 80,63 |
| 75 | 0,567 | 33,84 | 0,116 | 86,46 |
| 100 | 0,505 | 41,07 | 0,112 | 86,93 |

**TABLE 5.** Results of the ICP-OES method in determination of chemical elements in the sample μg/100 g.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Analyte** | **Emission wavelength, nm (detection method)** | **Amount in 100 g of sample, μg** | **Analyte** | **Emission wavelength, nm (detection method)** | **Amount in 100 g of sample, μg** |
| **Ag** | 338.289 | 0,11864±0,011 | Nb | 316.340 | 0,02355±0,003 |
| **Al** | 396.152 | 14,88908±0,016 | Nd | 378.425 | 0,18772±0,025 |
| **As** | 189.042 | 0,02037±0,002 | Ni | 221.647 | 0,1592±0,003 |
| **B** | 249.773 | 2,62225±0,014 | Tl | 190.856 | 0,0207±0,023 |
| **Ir** | 224.268 | 0,05262±0,002 | Pb | 220.353 | 0,0183±0,003 |
| **Cr** | 283.563 | 1,80651±0,003 |
| **Cs** | 852.113 | 3,02922±0,039 | Sb | 206.833 | 0,06018±0,109 |
| **Cu** | 324.754 | 2,70135±0,023 | Sc | 361.384 | 0,05054±0 |
| **Er** | 323.058 | 0,03812±0,002 | Si | 251.611 | 14,41839±0,254 |
| **Eu** | 381.967 | 0,04578±0,001 | Sm | 363.429 | 0,04661±0,014 |
| **In** | 325.609 | 0,22864±0,009 | Th | 283.231 | 0,36764±0,003 |
| **Ho** | 345.600 | 0,05531±0,001 | Mo | 202.030 | 0,23246±0,008 |
| **Mg** | 285.213 | 458,8338±7,08 | Na | 589.592 (Radial) | 395,88882±2,287 |
| **Fe** | 259.940 | 28,16389±0,132 | Zn | 213.856 | 3,79275±0,029 |
| **Hg** | 184.950 | 0,00508±0,001 | P | 185.942 | 169,17473±0,678 |
| **K** | 766.490 (Radial) | 2780,05587±0,037246 | Ti | 334.941 | 0,19404±0,003 |
| **Ba** | 455.403 | 2,519±0,023 | Mn | 257.610 | 8,95527±0,069 |
| **Sn** | 189.989 | 3,94345±0,041 | Tm | 342.508 | 0,00201±0 |
| **Y** | 371.030 | 0,05019±0,001 | U | 264.547 | 0,74313±0,457 |
| **La** | 333.749 | 0,05162±0 | V | 309.311 | 0,39373±0,004 |
| **Li** | 670.776 | 1,33232±0,015 |
| **Lu** | 261.542 | 0,04575±0 | Yb | 328.937 | 0,05159±0 |
| **Zr** | 343.823 | 0,0236±0 |  |  |  |

\* Note: <LOQ – result is less than the minimum of detection

Macroelements are found in large quantities in the studied grease samples. The quantitative value of macroelements decreases in the order K˃ Ca ˃ Mg ˃ Na ˃ P according to the Table 6.

**TABLE 6.** The amount of toxic elements in the sample (mg/1 kg)

|  |  |  |
| --- | --- | --- |
| **Name of element** | **Quantity in 1kg sample** | **Maximum permitted amount according to hygiene standards** |
| **Pb** | 0,18\*10-3 | 1,0 |
| **As** | 0,2\*10-3 | 1,0 |
| **Cd** | 0,4\*10-3 | 0,2 |
| **Hg** | 0,5\*10-4 | 0,3 |

As can be seen from Table 6, the amount of toxic elements in the food additive "AS-RAZZOQ" was found to be significantly lower than the standard specified in the "Hygienic Standards for Food Safety" [26], approved by the order of the Chief State Sanitary Doctor of the Ministry of Health of the Republic of Uzbekistan dated 25.05.2019 No. 0366-19.

Additives used in the food industry plays essential role not only in improving product quality, and also in ensuring its safety. Identification of substances with antimicrobial properties is important in determining their beneficial or harmful aspects for health. The antimicrobial effect of the food additive "AS-RAZZOQ" against Staphylococcus aureus and Pseudomonas spp. was studied in vitro using a quantitative method. According to the results, the extract showed zone of inhibition 20 mm against S. aureus and showed 18 mm zone against Pseudomonas spp [27-28]. When compared with the standard antibiotic for comparative analysis erythromycin, the following was observed:

**TABLE 7.** Comparison of the inhibition zone of the sample with the inhibition zone of erythromycin.

|  |  |  |  |
| --- | --- | --- | --- |
| **Bacteria type** | **Inhibition zone of the sample (mm)** | **Erythromycin inhibition zone (mm)** | **Comparative assessment of the effect** |
| **Staphylococcus aureus** | 20 | 22 | Activity is close to erythromycin |
| **Pseudomonas spp** | 18 | 12 | Activity is higher than erythromycin |

It is well known that both natural and synthetic drugs with broncholytic (expectorant) properties work by reducing the viscosity of sputum. Initially, the viscosity index was measured at 1.98 mPa∙s at a speed of 60 rpm over a duration of 5 minutes. After adding 60 mg (4%) of an expectorant to the sputum sample, the viscosity index decreased to 1.36 mPa∙s. The effect of the food additive "*AS-RAZZOQ*," which is also regarded as an expectorant, on sputum viscosity can be calculated as a percentage using the following formula.

(3)

*VDP% – Viscosity value difference in percentage.*

*Vs – Viscosity of sputum sample.*

*Ve – Viscosity of sputum sample with expectorant added.*

(4)

**CONCLUSION**

In conclusion, it can be said that the sample exhibits antiradical activity, in particular, its IC50 value was found to be 93.59 μl. This leads to a scientifically based conclusion that the food additive "AS-RAZZOQ" eliminates radicals such as CFP, which cause inflammation in diseases of the upper and lower respiratory tract. In addition, the biologically active compounds contained in the natural remedy also have therapeutic effects, such as strengthening the immune system of patients and thinning sputum formed in the lower respiratory tract (bronchi).

According to the results of the study of the content of macro- and microelements in the sample, it was found that the content of toxic elements in the sample is significantly lower than the maximum permissible amount according to hygienic standards. It was found that the content of macroelements K, Ca, Mg, Na and P is high, and the content of microelements Fe, Zn, Sr and Cu is high. The elements W, Ru, Os, Pd, Pr, Pt, Rb, Re, Ga, Gd, Ge, Hf, Dy, Co were not detected in the sample.

According to the results of the microbiological analysis of the new “AS-RAZZOQ” food additive based on medicinal plants, which was carried out in the microbiological analysis laboratory of the “Andijan Branch of the Center for the Safety of Pharmaceutical Products” using the method of quantitative determination of the antimicrobial activity in vitro, it was noted that the sample showed a 20 mm inhibition zone against S. aureus. This natural remedy contains biologically active substances effective against gram-positive bacteria. A noteworthy result is that the synthetic drug erythromycin, taken for comparison, showed a 12 mm inhibition zone against Pseudomonas spp., while the sample had a significantly higher inhibition zone of 18 mm. This means that the extract has a stronger antimicrobial effect than this antibiotic. In particular, the scientific conclusion is that the food supplement "AS-RAZZOQ" is effective against Pseudomonas spp., which is highly resistant to antibiotics, and can be used as a natural remedy for the treatment of respiratory tract diseases.

It is known that the effect of all natural and synthetic drugs with broncholytic (expectorant) activity is based on reducing the viscosity of sputum. The sputum-expectorant properties of the new natural food supplement AS-RAZZOQ were studied in vitro on sputum obtained from patients with acute or chronic bronchitis, and according to the results of the study, it was experimentally determined that this food supplement reduces the patient's sputum by 31.3%. Therefore, the natural food supplement AS-RAZZOQ is considered to have sputum-expectorant (broncholytic) properties.

# **REFERENCES**

1. Askarov I.R. Abdulloyev O.Sh. Otakhonov Q.Q. Razzakov Z.N. (2025) STUDY OF ANTI-RADICAL ACTIVITY OF MIXTURES OF CHAMOMILE (Matricaria chamomilla) AND OAK BARK (Corticis quercus) IN VARIOUS RATIO. (9(135)). <https://7universum.com/ru/nature/archive/item/20727>
2. Demir, N., Yildiz, O., Alpaslan, M., & Hayaloglu, A. (2014). Evaluation of volatiles, phenolic compounds and antioxidant activities of rose hip (Rosa L.) fruits in Turkey. LWT, 57(1), 126–133. <https://doi.org/10.1016/j.lwt.2013.12.038>
3. Ercisli, S. (2007). Chemical composition of fruits in some rose (Rosa spp.) species. Food Chemistry, 104(4), 1379–1384. <https://doi.org/10.1016/j.foodchem.2007.01.053>
4. Chrubasik, C., Roufogalis, B. D., Müller-Ladner, U., & Chrubasik, S. (2008). A systematic review on the Rosa canina effect and efficacy profiles. Phytotherapy Research, 22(6), 725–733. <https://doi.org/10.1002/ptr.2400>
5. Turkben, C., Barut, E., Copur, O. U., Durgut, E., & Himelrick, D. G. (2005). Evaluation of rose hips (Rosa spp.) selections. International Journal of Fruit Science, 5(2), 113–121. <https://doi.org/10.1300/J492v05n02_09>
6. Iancu, P., Soare, R., Dinu, M., Soare, M., Bonea, D., & Popescu, M. (2020). Analysis of the existing research regarding the use of the species Rosa canina L. Scientific Papers. Series B, Horticulture, 64(1), 325–331.
7. Gyamfi, M. A., Yonamine, M., & Aniya, Y. (1999). Free-radical scavenging action of medicinal herbs from Ghana: Thonningia sanguinea on experimentally-induced liver injuries. General Pharmacology: The Vascular System, 32(6), 661–667. <https://doi.org/10.1016/S0306-3623(98)00238-9>
8. Yang, R., Wang, L. Q., Yuan, B. C., & Liu, Y. (2015). The pharmacological activities of licorice. Planta Medica, 81(18), 1654–1669. <https://doi.org/10.1055/s-0035-1557893>
9. Hayashi, H., Hattori, S., Inoue, K., Sarsenbaev, K., Ito, M., & Honda, G. (2003). Field survey of Glycyrrhiza plants in Central Asia (3). Chemical characterization of G. glabra collected in Uzbekistan. Chemical and Pharmaceutical Bulletin, 51(11), 1338–1340. <https://doi.org/10.1248/cpb.51.1338>
10. Wenbin, L., Lin, L., Yidan, Z., Yanhua, C., Huaxu, Z., & Ming, N. (2019). Research status and trends of three kinds of medical radix glycyrrhizae based on bibliometric analysis (1992-2018). World Chinese Medicine, 14(3), 624–632. <https://doi.org/10.3969/j.issn.1673-7202.2019.03.029>
11. Sarker, S. D., & Nahar, L. (2007). Chemistry for pharmacy students: General, organic and natural product chemistry. John Wiley & Sons.
12. Firn, R. (2010). Nature’s chemicals: The natural products that shaped our world. Oxford University Press.
13. Kar, A. (2007). Pharmacognosy and pharmacobiotechnology (2nd rev. ed.). New Age International.
14. Cai, Y., Luo, Q., Sun, M., & Corke, H. (2004). Antioxidant activity and phenolic compounds of 112 traditional Chinese medicinal plants associated with anticancer. Life Sciences, 74(17), 2157–2184. <https://doi.org/10.1016/j.lfs.2003.09.047>
15. Martinez, M. J. A., Lazaro, R. M., del Olmo, L. M. B., & Benito, P. B. (2008). Anti-infectious activity in the Anthemideae tribe. In A. Rahman (Ed.), Studies in natural products chemistry (Vol. 35, pp. 445–516). Elsevier. <https://doi.org/10.1016/S1572-5995(08)80013-9>
16. Maurya, R., Singh, G., & Yadav, P. P. (2008). Antiosteoporotic agents from natural sources. In A. Rahman (Ed.), Studies in natural products chemistry (Vol. 35, pp. 517–545). Elsevier. <https://doi.org/10.1016/S1572-5995(08)80014-0>
17. Madziga, H. A., Sanni, S., & Sandabe, U. K. (2010). Phytochemical and elemental analysis of Acalypha wilkesiana leaf. Journal of American Science, 6(11), 510–514.
18. Gulcin, I., Beydemir, S., Sat, I. G., & Kufrevioglu, O. I. (2005). Evaluation of antioxidant activity of cornelian cherry (Cornus mas L.). Acta Alimentaria, 34(2), 193–202. <https://doi.org/10.1556/AAlim.34.2005.2.10>
19. Blois, M. S. (1958). Antioxidant determinations by the use of a stable free radical. Nature, 181(4617), 1199–1200. <https://doi.org/10.1038/1811199a0>
20. Abdullaev, S. S. (2024). Antiradical activity of food additives "Askarun" and "Askarufen". Journal of Chemistry of Goods and Traditional Medicine, 3(4), 116–133. <https://doi.org/10.55475/jcgtm/vol3.iss4.2024.349>
21. Balouiri, M., Sadiki, M., & Ibnsouda, S. K. (2016). Methods for in vitro evaluating antimicrobial activity: A review. Journal of Pharmaceutical Analysis, 6(2), 71–79. <https://doi.org/10.1016/j.jpha.2015.11.005>
22. Hausdorfer, J., Sompek, E., Allerberger, F., Dierich, M. P., & Rüsch-Gerdes, S. (1998). E-test for susceptibility testing of Mycobacterium tuberculosis. The International Journal of Tuberculosis and Lung Disease, 2(9), 751–755.
23. Wikipedia contributors. (n.d.). Glycyrrhiza. In Wikipedia. Retrieved [Month Day, Year], from <https://en.wikipedia.org/wiki/Glycyrrhiza>
24. Razzakov, N. A. (2022). Chemical composition of Rosa canina and Berberis plants and development of new food additives based on them [Doctoral dissertation, Andijan State University].
25. Ministry of Health of the Republic of Uzbekistan. (2019, May 25). The hygienic standards for food safety (Order No. 0366-19). Lex.uz. <https://lex.uz/docs/-4951750>
26. Bonitenko, E. Y., Skalny, A. V., & Kiseleva, M. F. (2005). Elemental status of the population of Russia. Part 1. General issues and modern methodological approaches to assessing the elemental status of an individual and a population. ELBI-SPb.
27. Kostrov, S. V. (2012). Evaluation of the effectiveness of cobalt complex compounds in purulent-inflammatory processes [Abstract of Doctoral dissertation]. Kursk State Medical University.
28. Radysh, I. V., Skalny, A. V., Notova, S. V., Marshinskaya, O. V., & Kazakova, T. V. (2017). Introduction to elementology. Orenburg State University Press.