

## INVESTIGATION OF Pb AND Ni LEVELS IN SUNFLOWER PLANT AGRICULTURE TWO DIFFERENT pH ENVIRONMENTS

Cihan Torlak<sup>1</sup>, Cemile OZCAN<sup>2\*</sup>

<sup>1,2</sup>Department of Chemistry, Science and Art Faculty, Kırklareli University, Kırklareli, Turkey

### Abstract

In this study, in the soil samples collected from farmland with two different pH environments located in Kırklareli Province, heavy metals (Pb and Ni) were aimed to investigate the relationship. The working pH values are took in 3 different periods (after the formation of the plant, week of 1 and 6, harvest time) from the farm land in the town of Kırklareli and Kavaklı. In order to determine the concentration of Pb and Ni metals, the reading process was performed by the Flame Atomic Absorption Spectrophotometer (FAAS). The concentrations of Pb and Ni metals in soils in the first region were 36.3-43.7 mg/kg and 12.6-14.9 mg/kg, in the second region soil concentrations were 11.7-17.1 mg/kg, 0.09-0.24 mg/kg, 3.12-4.353 mg/kg, 7.89-11.2 mg/kg, 56.3-9.54 mg/kg, respectively. The concentrations of Pb and Ni metals in first region plant components were 1.93-12.1 mg/kg, and 0.56-11.8 mg/kg, the second region was 6.92-14.8 mg/kg and 0.58-9.37 mg/kg, respectively. The relative standard deviation (RSD) was found to be less than 10%. The RSD of analysis of samples have satisfying precision.

**Keywords:** FAAS, heavy metal, soil, sunflower.

### AYÇİÇEĞİ BİTKİSİ TARIMINDA İKİ FARKLI pH ORTAMINDA Pb ve Ni DÜZEYLERİNİN İNCELENMESİ

#### Öz

Bu çalışmada Kırklareli İlinde bulunan iki farklı pH ortamına sahip tarım arazilerinden toplanan toprak örneklerinde ağır metallerin (Pb ve Ni) ilişkisinin incelenmesi amaçlanmıştır. Kırklareli ve Kavaklı ilçesindeki çiftlik arazisinden çalışma pH değerleri 3 farklı dönemde (bitkinin oluşumundan sonra 1. ve 6. hafta, hasat zamanı) bitki örnekleri alındı. Pb ve Ni metallerinin konsantrasyonunu belirlemek için tayin işlemi Alevli Atomik Absorpsiyon Spektrofotometresi (FAAS) ile gerçekleştirildi. Birinci bölgedeki topraklardaki Pb ve Ni metal konsantrasyonları 36.3-43.7 mg / kg ve 12.6-14.9 mg / kg, ikinci bölgede ise sırasıyla 11.7-17.1 mg/kg, 0.09-0.24 mg/kg, 3.12-4.353 mg/kg, 7.89-11.2 mg/kg, 56.3-9.54 mg/kg bulundu. Birinci bölge bitki bileşenlerindeki Pb ve Ni metal konsantrasyonları sırasıyla 1.93-12.1 mg / kg ve 0.56-11.8 mg / kg, ikinci bölge ise sırasıyla 6.92-14.8 mg / kg ve 0.58-9.37 mg / kg idi. Bağlı standart sapmanın (RSD) % 10'dan az bulundu. Numunelerin analizinin RSD'si tatmin edici bir hassasiyete sahip olduğu belirlenmiştir.

**Anahtar Kelimeler:** FAAS, ağır metal, toprak, ayçiçeği.

Corresponding author: cemilebal.ozcan@klu.edu.tr

## INTRODUCTION

In the world, there has been a great progress in the industrial and industrial fields with the technology to meet the basic needs of the people [1-3]. As a result of this progression, it should not be ignored that pollution poses a serious problem to the environment, and that pollution, especially to the agricultural land, air layer and the water resources used for irrigation purposes, cause serious damage. In this context, there is a significant decrease or extinction of living species and basic food products. The most important environmental problem in the world is heavy metal pollution. Heavy metals are an important area for the most widely used and monitored pollutants due to their common uses [3-9].

Lead (Pb) is the metal that causes the most damage to our environment with human activities. Pb is one of the most important heavy metals emitted as a metal or organo-metallic compound into the atmosphere and creates ecological pollution due to its toxic properties [7-13]. It was revealed that the nickel (Ni) was a nutritional element needed for the growth and development of plants since 1987 [14, 15]. The need for nickel in plants is necessary in the germination period of the seed. In addition, nickel is a metal part of urease enzyme and many hydrogenase enzymes, which is a catalase enzyme that converts urea to ammonium and carbon dioxide [14-16].

*Helianthus annuus* L. (sunflower) is one of the most important oil plants produced and used today. Sunflower oil is among the most preferred vegetable oils in terms of food quality [8, 17-20]. For this reason, sunflower cultivation is also very common in the world. One of the factors affecting the transition of the necessary nutrients to the plant in plant development is undoubtedly the pH of the soil. As shown by the tolerance of the specific pH range of each plant, the sunflower plant can maintain its growth within the pH range of 6.0-7.2 [1, 21]. In addition to the upper or lower values of these limits, there is a decrease in plant growth, decline and decrease in product efficiency. In recent years, various breeding studies have been carried out to take control of weed in sunflower.

In this study, the sunflower (*Helianthus annuus* L.) grown or grown in two different pH environments in Kırklareli province, the possible residual contents of the soil and its components (root, leaf, stem and seed) grown in sunflower plants, possibly possible Pb and Ni metal levels are aimed to determine the relationship between.

## MATERIAL AND METHOD

### Study Area

This study was carried out in the farmland and the farmland in the Kavaklı District, which is located 10 km west of the province and in the agricultural land connected to the Atatürk Soil Water and Agricultural Metrology Research Institute, 4 km west of the city of Kırklareli on the Thrace side of the Marmara Region.

### Sample Collection

In this study, soil and sunflower-parts (root, stem, leaf, head and seed) samples which taken from TUBITAK-113Y529 project were used. These samples were two different pH values in Kırklareli Merkez (7.28) and Kavaklı (4.95). The periods in which the samples were taken and the plant components taken were shown in Table 1.

**Table 1:** Sample sampling planning and collection times.

Sampling Time	Sunflower plant components and growing soil					
	Soil	Root	Body	Leaf	Head	Seed
1 <sup>st</sup> Sowing period (week 1)	+	+	+	+	-	-
2 <sup>nd</sup> Plant development period (6th week)	+	+	+	+	+	-
3 <sup>rd</sup> Harvest period	+	+	+	+	+	+

The first location (Kırklareli City Center), Atatürk Soil Water and Agricultural Meteorology Research Institute, in the soil of the trial agricultural land has a clay soil structure and a neutral pH. As the second location, land information of Kavaklı agricultural land, in the soil structure is sand and has the property of acidity pH.

### Sample Preparation

Samples collected from specified points were brought to the laboratory and the sunflower plant was washed with tap water, then 3 times with pure water and then dried at 40 °C in the oven. The dried plant samples were ground by a titanium-coated blender and prepared for weighing. Soil samples were also dried at room temperature and passed through a 450 mesh sieve.

### Dissolution process in the microwave

For heavy metal analysis of the samples, CEM-MARS 6 brand microwave was used during the solubilization process. Optimization studies were conducted for the appropriate solubilization process. Power and temperature scans were performed for the most suitable dissolution for plant and soil samples (Table 2 and Table 3). The fourth method was chosen as the most suitable solubilization method for the plants and the third method was chosen for the soil.

**Table 2:** Microwave dissolution process optimization studies for plants.

Method	Power (W)	Time (min)	First temperature (°C)	Last temperature (°C)	Time to hold (min)	Total time (min)
1	450	20	100	220	5	40
2	450	20	100	180	5	40
3	550	20	100	150	5	40
4	600	20	100	180	5	40
5	600	20	100	200	5	40
6	600	20	100	220	5	40

**Table 3:** Microwave dissolution process optimization studies for soil.

Method	Power (W)	Time (min)	First temperature (°C)	Last temperature (°C)	Time to hold (min)	Total time (min)
1	600	20	100	180	5	40
2	600	20	100	200	5	40
3	600	20	100	220	5	40

Samples of plant components for microwave solubilization were weighed in an analytical precision balance of 0.5 g and transferred to 75 mL of teflon flasks. 1 mL of 35% H<sub>2</sub>O<sub>2</sub> (Merck) and 9 mL of 65% concentrated HNO<sub>3</sub> (Merck) were added to the vessels and digested (Table 2 fourth method) in the microwave, respectively. The dissolved samples were then taken to the falcon tubes and centrifuged at 15 °C at 5000 rpm for 7 minutes. The decanted supernatant was evaporated in a water bath at 40-45 °C until 2-3 mL remained and was completed with ultrapure water to a final volume of 50 mL. Soluble samples were prepared for reading by FAAS.

Samples of soil components for microwave solubilization were weighed in an analytical precision balance of 0.5 g and transferred to teflon flasks. 1 mL of 35% H<sub>2</sub>O<sub>2</sub> (Merck) and 9 mL of 65%

concentrated HNO<sub>3</sub> (Merck) and 3 mL of %37 HCl were added to the vessels and digested (Table 3 third method) in the microwave, respectively. As in the plant sample, the soil samples were prepared for reading with same method by FAAS.

The same methods were made to blind samples and blind absorbance values were calculated by subtracting the actual values from the absorbance values.

### Preparation of standard solutions and reagents

For standard solutions of metals, 1000 mg/L NIST standard stock solutions were used. The standards of the metals for Pb and Ni to be analyzed were prepared for the appropriate working range of 0.025–2 mg/kg. Standard solutions were prepared by dissolving with 1 M HNO<sub>3</sub>.

Agilent 240 AA Duo model flame atomic absorption spectrophotometer (FAAS) was used for metal analyzes. Device parameters for metal analysis were given in Table 4.

**Table 4:** Parameters related to FAAS for metals.

	Wavelength (nm)	Lamp Current (mA)	Slit Width (nm)	Used Flame
<b>Pb</b>	217.0	10.0	0.2	Air/Acetylene
<b>Ni</b>	352.5	4.0		

The solutions of 0.025, 0.05, 0.1, 0.25, 0.5, 1 and 2 mg/kg prepared from standards of Pb and Ni were read in FAAS and RSD (relative standard deviation) (%) values were determined in the range of 0.5–9.8 and 0.2–1.9, respectively. The linear equation and the correlation coefficient value for the Pb and Ni, respectively obtained;  $y = 0.126x - 0.003$ ,  $R^2 = 0.9993$  and  $y = 0.1536x + 0.0005$ ;  $R^2 = 0.9998$ . The RSD of analysis of samples have satisfying precision.

## RESULTS

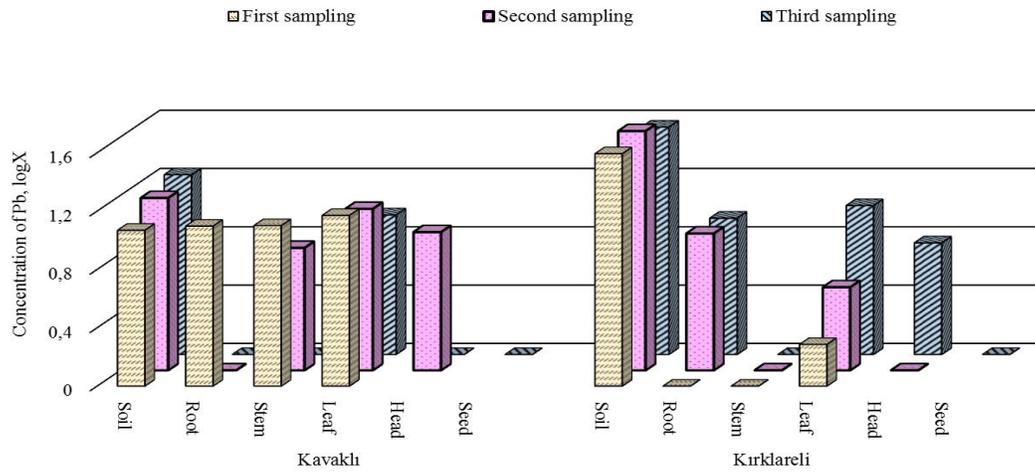
The heavy metal contents of samples, which sunflower plant and soil were collected of the samples taken from 3 different periods in two different pH environments in Kırklareli, were determined by FAAS. The obtained results were compared with the national and international quality control standard limit values such as Turkish Food Codex and WHO/FAO (World Health Organization/Food and Agriculture Organization) [19, 22, 23]. The heavy metal concentration

results which belonging to two different agricultural lands are given of soil samples and of the root and body part, leaf, head and seed samples of the sunflower plant, in Table 5.

**Table 5:** The amount of Pb and Ni in the soil where sunflower plants grow and the parts of sunflower plant (mg/kg, n=6).

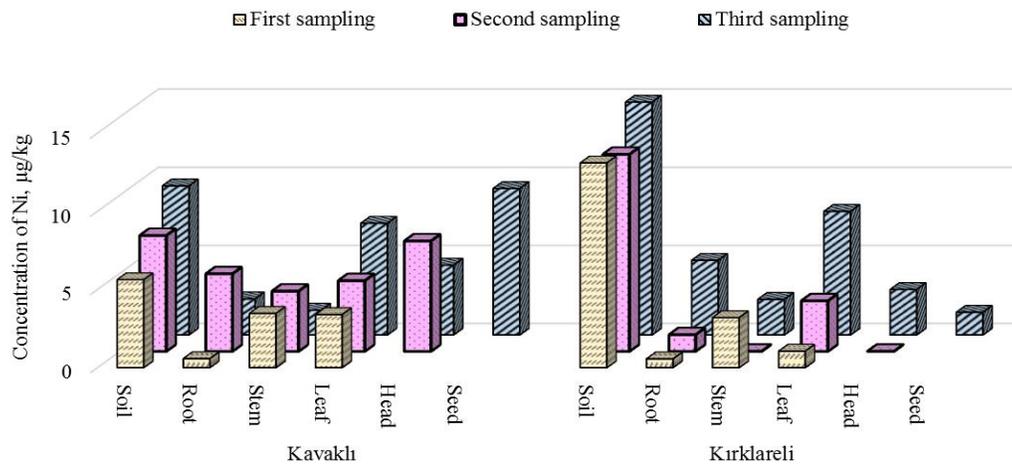
	Samples	Pb	Ni
Kavaklı Location	Soil 1	11,7 ± 1,21	5,63 ± 0,27
	Soil 2	15,2 ± 1,12	7,41 ± 0,34
	Soil 3	17,1 ± 0,97	9,54 ± 0,07
	Root 1	12,5 ± 1,17	0,58 ± 0,01
	Root 2	ND	4,96 ± 0,32
	Root 3	ND	2,31 ± 0,11
	Body 1	12,6 ± 1,91	3,47 ± 0,12
	Body 2	6,91 ± 1,36	3,85 ± 0,4
	Body 3	ND	1,59 ± 0,3
	Leaf 1	14,8 ± 1,31	3,4 ± 0,15
	Leaf 2	12,8 ± 1,31	4,52 ± 0,25
	Leaf 3	9,07 ± 0,33	7,15 ± 0,38
	Head 2	8,82 ± 1,24	7,06 ± 0,16
	Head 3	ND	4,47 ± 0,31
	Seed 3	ND	9,37 ± 0,17
Kırlareli Location	Soil 1	39,2 ± 3,95	13,1 ± 0,36
	Soil 2	43,7 ± 0,68	12,6 ± 0,7
	Soil 3	36,3 ± 0,75	14,9 ± 0,33
	Root 1	ND	0,56 ± 0,02
	Root 2	8,67 ± 1,22	1,06 ± 0,15
	Root 3	8,61 ± 1,31	4,78 ± 0,02
	Body 1	ND	3,2 ± 0,29
	Body 2	ND	ND
	Body 3	ND	2,28 ± 0,28
	Leaf 1	1,93 ± 0,08	1,06 ± 0,18
	Leaf 2	3,71 ± 0,34	3,23 ± 0,17
	Leaf 3	10,5 ± 1,25	7,91 ± 0,06
	Head 2	ND	ND
	Head 3	5,83 ± 0,07	2,91 ± 0,24
Seed 3	ND	1,45 ± 0,14	

ND: Not detection.



**Figure 1:** The distribution of Pb concentration in the sunflower plants of Kırklareli and Kavaklı.

According to the results of the graphical analysis, the Pb concentrations in the soil and plant parts of Kırklareli and Kavaklı locations were found to be amongst 1.93-39.2 mg/kg and 6.91-17.1 mg/kg, respectively (Figure 1). The Pb concentrations in two locations were exceeded the permissible limit values. When the addition 1 and addition 2 are examined, it is seen that the transfer factor of the lead in the Kırklareli and Kavaklı locations increases towards the soil from the plant components.



**Figure 2:** The distribution of nickel concentration in parts of sunflower plants from Kırklareli and Kavaklı location.

When the graphs obtained from the Ni contents were examined, the Ni concentrations in the soil and plant components in Kırklareli and Kavaklı locations were determined as 0.56-14.9 mg/kg and 0.58-9.54 mg/kg, respectively (Figure 2). Ni concentrations in Kırklareli and Kavaklı locations are transferred from soil to plant components (Transfer factor addition 1 and 2). Moreover, Ni concentrations in two locations were determined to be within the limit values.

The comprises a group of technics in which plants absorb heavy metals, including lead, through their roots and relocate them to harvestable parts of the plant, such as stems, shoots, and leaves, being called of plants [1, 12]. Therefore, the correlation between sunflower grown in Kırklareli and Kavaklı and its environment was examined. Positive correlation (0.80 and above) in soil and stem and negative correlation (= 0.98 and above) in root and leaf were determined for Pb and Ni in Kavaklı. Negative correlation in soil and stem, positive correlation (= 0.59 and above) in root and leaf were determined for Pb and Ni in Kırklareli. No correlation was determined in the head and seed.

## CONCLUSION

At the end of the study, three different periods (first week, sixth week and harvest period) were performed in the Kırklareli city center and Kavaklı town. Transitions of plant components of soil elements can be evaluated by means of transfer factor. It is known that some elements accumulate in their components as a result of accumulation within the body by absorbing the plants depending on the environmental factors that are not present in the soil structure. At the beginning of the elements that adhere to the tissues of the plants with environmental impact, lead and cadmium are the most common.

In the study, the results of metal analysis did not exceed the limit values determined by Turkish Food Codex and WHO/FAO except lead. The most important factors in the dissemination of heavy metals to the environment are industrial works, motor vehicle exhausts, mining operations and fertilizers used in agriculture. The Pb concentrations in the soil and plant samples of Kırklareli location were found between 36.3-43.7 mg/kg and 1.93-12.1 mg/kg. The Pb concentrations in the soil and plant samples of Kavaklı town were found to be between 11.7-17.1 mg/kg and 6.92-14.8 mg/kg. Ni concentrations in the soil samples of Kırklareli and Kavaklı locations were determined

between 12.6-14.9 mg/kg and 5.63-9.54 mg/kg, while, plant parts were found between 0.56-11.8 mg / kg and 0.58-9.37 mg/kg, respectively.

As a result of metal analysis were investigated in contents of Pb and Ni. When soil and plant samples were compared according to European Union standard, Turkish Food Codex and WHO limit values, only, Pb concentrations were found to be above the limit values.

## ACKNOWLEDGEMENTS

The authors thank the Kırklareli University Research Foundation for their support (KLUBAP/121).

## REFERENCES

- [1] Sessitsch, A., Kuffner, M., Kidd, P., Vangronsveld, J., Wenzel, W.W., Fallmann, K., The role of plant-associated bacteria in the mobilization and phytoextraction of trace elements in contaminated soils. *Soil Biol Biochem*, C 60, S 182-94, 2013.
- [2] Mendil, D., Uluozlu, O.D., Tuzen, M., Soylak, M., Investigation of the levels of some element in edible oil samples produced in Turkey by atomic absorption spectrometry *J. Hazard. Mater.*, C 165, S 724–728, 2009.
- [3] Bakircioglu, D., Kurtulus, Y.B., İbar, H., Comparison of extraction procedures for assessing soil metal bioavailability of to wheat grains. *Clean–Soil, Air, Water*, C 39 (8), S 728-734, 2011.
- [4] Amin H, Arain BA, Jahangir TM, Abbasi MS, Amin F. Accumulation and distribution of lead (Pb) in plant tissues of guar (*Cyamopsis tetragonoloba* L.) and sesame (*Sesamum indicum* L.): profitable phytoremediation with biofuel crops. *Geol Ecol Landscapes*, C 2, S 51-60, 2018.
- [5] Okçu, M., Tozlu, E., Kumlay, M., Pehlivan, M., Ağır Metallerin Bitkiler Üzerine Etkileri. *Doğu Anadolu Tarımsal Araştırma Enstitüsü Müdürlüğü Erzurum*. C 17 (B), S 14-26, 2009.
- [6] Azevedo, R. A. and Lea P.J., Toxic Metals in Plants, *Brasil Journal Plant Physiology*, 17-1, 2005.
- [7] Yörük, O., *Ergene Havzasında Yetiştirilen Ayçiçek Bitkisinde (Helianthus annuus L.) Bazı Eser Element İçeriklerinin ICP-OES ile Tayini* (Yüksek Lisans Tezi), Trakya Üniversitesi Fen Bilimleri Enstitüsü, Kimya Anabilim Dalı Edirne, 2008.
- [8] Mehmet Aycicek, Olcay Kaplan, Mehmet Yaman, Effect of Cadmium on Germination, Early Seedling Growth and Metal Contents of Sunflower (*Helianthus annus* L.), *Asian J. Chem*, 20(4), S 2663-2672, 2008.

- [9] Asri, F., Sönmez, S., Ağır Metal Toksisitesinin Bitki Metabolizması Üzerine Etkileri. *Akdeniz Üniversitesi Ziraat Fakültesi Toprak Bölümü*, Antalya, 2006.
- [10] Pourrut B, Shahid M, Dumat C, Winterton P, Lead Uptake, Toxicity, and Detoxification in Plants. Springer Verlag 213, S 113–36, 2012.
- [11] Zengin, F. ve Munzuroğlu K., Effects of lead (Pb<sup>2+</sup>) and Copper (Cu<sup>2+</sup>) on the growth of root, Shoot and Leaf of Bean (*Phaseolus vulgaris* L.) Seedlings, *Gazi University, Journal Of Science* 17(3), S 1-10, 2004.
- [12] Ashraf U, Kanu AS, Mo Z, Hussain S, Anjum SA, Khan I., Lead toxicity in rice: effects, mechanisms, and mitigation strategies—a mini review. *Environ Sci Pollut Res*, 22, S 18318-32, 2015.
- [13] Brown, P., H., Welch, R., M., Cary, E., E. Nickel: A micronutrient essential for higher plants. *Plant Physiol.* 85, S 801–803, 1987.
- [14] Gerendas, J., Polacco, J., C., Freyermuth, S., K., Sattelmacher, B., Significance of nickel for plant growth and metabolism. *J. Plant Nutr. Soil Sc.* 162 (3), S 241–256, 1999.
- [15] Tokay, F., Bağdat, S. Extraction of nickel from edible oils with a complexing agent prior to determination by FAAS. *Food Chemistry*, 197, S 445-449, 2016.
- [16] Bakircioglu Kurtulus, Y., Bakircioglu, D., Babac A.C., Yurtsever, S., Topraksever, N., Extraction of Cu, Fe, Mn and Ni from Margarine Samples using Extraction Induced by Emulsion Breaking Procedure prior to Graphite Furnace Atomic Absorption Spectrometry and Comparison of Results to Provisional Tolerable Daily Intake Values, *Journal of AOAC International*, 103(5), S 1256–1263, 2020.
- [17] Konyalı, S., Sunflower Production and Agricultural Policies In Turkey. *Sosyal Bilimler Araştırma Dergisi*, 6(4), S 11-19, 2017.
- [18] Anonim, 2002, Official Gazette, Ministry of Agriculture and Rural Affairs and Ministry of Health (2002) Turkish Food Codex Communique' on Determination of Maximum Levels of Specific Contaminants in Foodstuffs (Communique'No: 2002/63) 1–13. <http://www.resmigazete.gov.tr/eskiler/2002/09/20020923.htm>. date of access: 18.09.2020.
- [19] Anonim 2016, TÜİK (2016). Türkiye İstatistik Kurumu (TÜİK). (web sayfası: <http://www.tuik.gov.tr/UstMenu.do?metod=kategorist> ), alındığı tarihi: 04.09.2017.
- [20] USDA, (2016), United States Department of Agriculture Economic Research Service (USDA).



- [21] Bilen, S., Sezen, Y., Toprak Reaksiyonunun Bitki Besin Elementleri El Verişliliği Üzerine Etkisi. Atatürk Ü. Zir. Fak. Der., 24 (2), 156-166, Erzurum, Türkiye, 1993.
- [22] JECFA (1983) Evaluation of Certain Food Additives and Contaminants. Twenty-seventh Report of the joint FAO/WHO Expert Committee on Food Additives. Technical Report Series 696, 52. <https://www.who.int/ipcs/publications/jecfa/reports/trs940.pdf>
- [23] FAO (1996) Trace Elements in Human Nutrition and Health. World Health Organization. <https://www.who.int/nutrition/publications/micronutrients/9241561734/en/> date of access: 18.09.2020.