Analysis of the influence of lead (Pb) and soil pH on St. John's wort preparations - the case of the Lukavac industrial zone in Bosnia and Herzegovina

Azra Suljic[1], Ervina Becic[2], Maida Šljivić Husejnović[1], Nedim Suljic[3]

[1]Faculty of Pharmacy, University of Tuzla, Tuzla, Bosnia and Herzegovina

[2]Faculty of Pharmacy, University of Sarajevo, Sarajevo, Bosnia and Herzegovina

[3]Faculty of Mining, Geology and Civil Engineering, University of Tuzla, Tuzla, Bosnia and Herzegovina

corresponding author: Azra Suljić (sulji.azra@gmail.com)

Abstract

The industrial zone in Lukavac, a city in northeastern Bosnia and Herzegovina, is highly developed with large industrial complexes such as a cement factory, a soda factory, and a fertilizer factory. The location of the soil samples taken is next to the Tuzla-Doboj main road, near the cement factory and near the regulated Jala river. The impact of lead as a heavy metal in the surface layers of the soil usually ranges from 2 to 100 mg/kg, but lead is a strong pollutant of the environment and is toxic even in low concentrations. The population often uses medicinal plants near the location of the soil samples from which they make tea and other preparations. Also, near the location of the soil samples, there are artesian wells from which the population of Lukavac is supplied with water. The samples showed that there is a concentration of lead in the soil for a neutral to slightly basic pH ranging between 7.44 and 7.97. The paper analyzes the content of lead in oil and salve made from St. John's wort plants from the locality of soil sampling and the relationship between the concentration of lead in the soil and the mentioned St. John's wort preparations. The concentration of lead in oil and salve made from St. John's wort was tested using the method of atomic absorption spectrometry. The influence of lead on soil pollution and on the pollution of oil and salve made from St. John's wort plants are important from the aspect of human health, but also from the aspect of infiltration of rainwater into the deeper layers of the soil, which also pollutes the underground water at the location. The analyzes performed showed a higher concentration of lead in St. John's wort salve than in oil made from the same St. John's wort plants for lead concentrations in the soil between 4.60 mg/kg and 14.06 mg/kg.

Keywords: lead, St. John's wort, preparations of St. John's wort, soil pH, infiltration

Introduction

Lead is one of the most dangerous heavy metals due to its toxicity but also due to its wide distribution in the environment. Lead is found in soil, water and air, and therefore also in plants [1] [2]. The most common sources of lead pollution are the proximity of roads and polluted air in industrial areas [4] [16], but also the food we eat and the objects we use every day that may contain lead [8]. The share of lead from natural sources is small compared to the share of lead in the environment that is created by anthropogenic activity [9]. Due to rapid industrialization, lead exposure and its consequences are becoming a significant health problem in most developing countries [1] [14]. It is also important not to neglect the possible pollution of groundwater, due to the infiltration of rainwater into deeper layers of soil [10] [3].

Lead is classified as a group 2B carcinogen [11], which means that it is "possibly carcinogenic to humans", and this carcinogenicity can be caused by several mechanisms, such as: chronic nephropathy, mitogenesis, genotoxicity [13]. Its genotoxicity takes place mainly through indirect reactions with DNA, such as oxidative stress [12]. Lead is very quickly absorbed into the bloodstream, and mostly affects the central nervous system, cardiovascular system, immune system, bones and kidneys [18] [19]. Over time, lead accumulates in the human body, which absorbs up to 15% of swallowed or inhaled lead, and children's bodies absorb up to 50% of swallowed or inhaled lead. Given that it is divalent like calcium and zinc cations, it can interfere with many cellular mechanisms and biochemical reactions in the human body, changing these cations [8] [15].

Accumulation of lead in plants and even in St. John's wort depends on the form in which lead appears. The inorganic form of lead is difficult to accumulate and moves from the soil to the above-ground parts of plants, while the organic lead compounds accumulate quickly and moves from the surface layers of the soil to the above-ground parts of St. John's wort plants. Studies have shown that St. John's wort shows a high level of lead

absorption, however, this depends on several factors, including the pH value of the soil, but also the potential presence of other metals to which the plant is exposed [5]. It has been shown that metals usually have a higher availability in acidic soils, since acids help the breakdown of metals into bioavailable free ions, while in alkaline soils metals tend to form poorly soluble phosphates and carbonates [6] [7].

Norming the content of heavy metals in soil from the aspect of environmental protection is quite difficult due to the problems related to taking into account all environmental factors. The standards do not take into account the climatic characteristics of individual areas and regions. According to the World Health Organization, the maximum permissible concentration of lead in soil is 85 mg/kg of soil, while agricultural soils can contain on average about 10 mg/kg [17].

The soil at the analyzed location has a complex heterogeneous composition, so the development of a single maximum permissible concentration is a rather difficult task. The assessment of the threat to soil from heavy metals is carried out according to the purpose of the territory. Changes in soil acidity can significantly affect the phytotoxicity of metals. The dependence of the generally permissible concentrations on the granulometric composition of the soil is very important. The soil at the location of the samples taken is sandy soil.

The location of the soil samples taken and the St. John's wort samples from which the oil and salve were made is located near the Lukavac cement factory, which is about 450 m as the crow flies, and from the Sisecam factory, which produces soda ash, light soda and bicarbonate of soda, which are used as raw materials. for the industrial production of glass and detergents, as well as for the production of chemical substances and textiles, the location of the soil and St. John's wort samples taken is about 1000 m as the crow flies. The main road Tuzla-Doboj is about 12 m away from the location of the soil samples and St. John's wort plants.

St. John's wort and its properties

St. John's wort is a perennial herb with an upright, branched stem up to 1 m high, which blooms throughout the summer [20]. It is a very widespread plant species, growing wild in Europe and Asia, and has also spread to Australia and North America. It has been present in the traditional medicine of many nations since ancient times [2], including in traditional Bosnian and Herzegovina medicine. It can grow in groups or singly in sunny parts of meadows, along roads, along forest edges and on uncultivated land. It is widely distributed in Bosnia and Herzegovina. It is rarely cultivated in plantations, and it is mainly collected from nature on the market, so there is a high probability of potential contamination with heavy metals, including lead. For this reason, quality control of raw plant material and preparations based on St. John's wort is of great importance[21] [24].

Although St. John's wort is best known as an antidepressant that is most often prescribed in mild depressive states [23], it is also used as a sedative, diuretic, analgesic, astringent and antipyretic [19] [20]. The most famous St. John's wort preparation is St. John's wort oil, which has multiple uses in folk medicine. St. John's wort oil is used for wound healing, for skin health, relief of burns, cuts and rheumatic pain [22].

According to some research, St. John's wort preparations can help with neurodegenerative diseases, such as Parkinson's disease [25] and in the treatment of AIDS [24], and it is assumed that hypericin and hyperforin from St. John's wort can increase the level of serotonin in the brain, and people suffering from depression often have low levels of serotonin [26].

St. John's wort's medicinal value is quite extensive, and preparations and preparations from St. John's wort are used in the form of tablets and teas, as well as in the form of oils, vagitories and creams. For the purposes of this research, preparations of St. John's wort oil and cream were analyzed as well as the properties of the land in the area of the industrial zone in Lukavac in Bosnia and Herzegovina, an area with highly polluted air and water.

Sampling of soil and St. John's wort

Soil sampling was done manually using a stainless steel knife on a field surface of 250 m^2 and at depths of 0 cm (field surface), 5 cm and 10 cm deep. St. John's wort plant samples are the above-ground part of the plant that was divided into St. John's wort leaf and flower, they were taken from the same location and place where the soil samples were taken.

Before sampling the land, a "recording" was carried out, that is, an inspection of the land based on which the size of the sampling network was determined, which depends on the plant culture and the homogeneity of the terrain surface. Based on this, three places on an area of 250 m^2 were selected, from which samples of St. John's wort and soil samples were taken. The analyzed experimental plot of 250 m^2 is only with St. John's wort, the plot is flat and without slope and of the same type of land, almost uniform soil color, without microdepressions, so three representative soils and St. John's wort plants were taken. For each analyzed sample, 500 g of soil was taken, in which the pH value and lead content were determined.

The maximum permissible concentration of heavy metals in soil has a number of shortcomings, such as the lack of a unified approach to establishing the maximum permissible concentration of heavy metals, including lead, in soil, the complex interaction of heavy metals in soil is not taken into account, and there is a significant difference in the standards for the maximum permissible concentration and the generally permissible concentrations of heavy metals in soil.

Today, maximum permitted concentrations and tentatively permitted concentrations are used. Therefore, it is advisable to compare the level of soil pollution with the approximate permissible concentration and maximum permissible concentration of heavy metals, i.e. lead, in this research.

Figure 1. Location of soli and St John's wort sampling

St. John's wort was sampled at the beginning of August 2023 at the location of the industrial zone in the town of Lukavac in Bosnia and Herzegovina. At the same time, from the same points, soil samples were taken in which the pH value and lead concentration were tested. The aerial parts of the plant with flowers were taken from St. John's wort, from which oil and cream were made.

Figure 2. St. John's wort at the sampling location

Results of lead concentration in soil, St. John's wort plants and St. John's wort preparations

On the basis of soil samples at the location of the industrial zone in Lukavac in the northeastern part of Bosnia and Herzegovina, the concentrations of lead in the soil, the concentration of lead in the St. John's wort plant and the pH value of the soil were determined. Three soil samples and three St. John's wort plant samples were analyzed from the same place where the first soil sample was taken.

Table 1: Results of lead (Pb) concentration in soil and St. John's wort samples

SAMPLE NUMBER	SOIL pH	Pb IN SOIL (mg/kg)	Pb IN ST. JOHN'S (mg/kg)
1	7.97	7.56	0.020
2	7.69	14.06	0.026
3	7.44	4.60	0.019

According to the pH value of the soil, the analysis found that it is a neutral to slightly basic soil, while the concentration of lead in the soil ranges from 4.60 mg/kg to 14.06 mg/kg. The concentration of lead (Pb) in St. John's wort plants ranges from 0.019 mg/kg to 0.026 mg/kg.

The graphical presentation of the obtained results can be used to define corresponding changes in certain values, i. e. changes in the concentration of lead in the soil and as a function of the concentration of lead in St. John's wort plants from the aspect of the slightly basic soil environment.

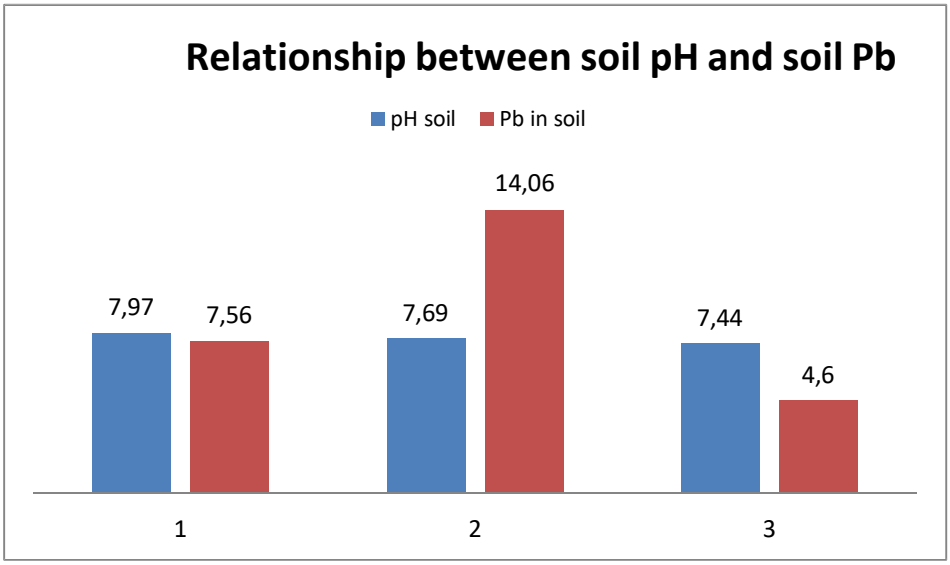

Figure 3. Diagram of the relationship between pH in soil samples and lead concentration in soil samples

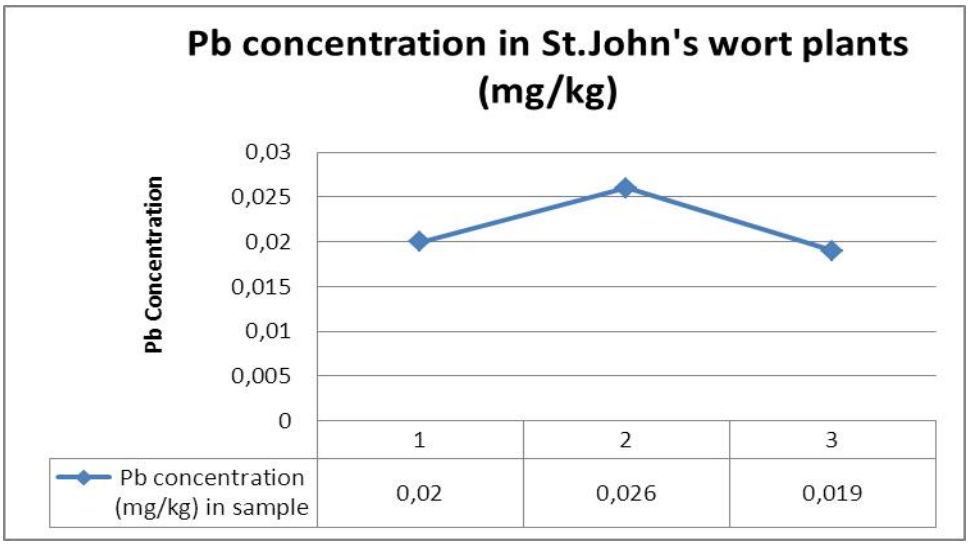

Figure 4. Lead concentration in St. John's wort plants (mg/kg)

St. John's wort plants in which the lead concentration was determined in values of 0.019 mg/kg to 0.026 mg/kg were made into oil and salve as St. John's wort preparations with determined concentrations of lead in the plants and in the soil. Based on the method of atomic absorption spectrometry (AAS) (BAS EN 13804:2015 and BAS EN 14084:2005), the following lead concentrations were determined in oil and salve made from St. John's wort plants, namely:

Table 2: Results of lead (Pb) concentration in oil and salve made from St. John's wort plants

SAMPLE NUMBER	OIL FROM ST. JOHN'S (concentration Pb in mg/kg)	SALVE FROM ST.JOHN'S (concentration Pb in mg/kg)	Pb IN ST. JOHN'S (mg/kg)
1	0.0023	0.0041	0.020
2	0.0025	0.0046	0.026
3	0.0022	0.0040	0.019

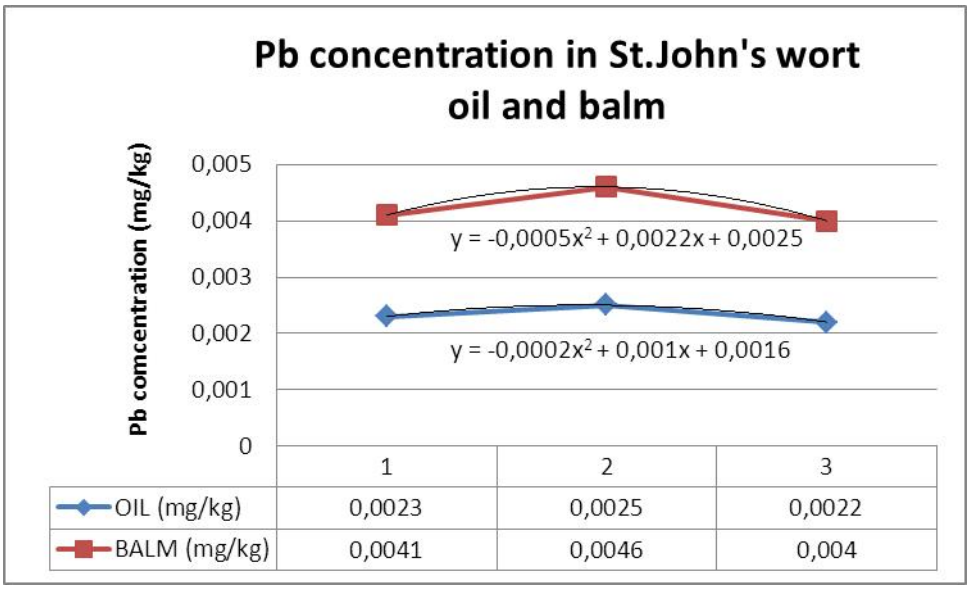

Figure 5. Lead concentration in oil and salve made from St. John's wort (mg/kg)

From Figure 5, it can be seen that the change in lead concentration in the oil and salve made from St. John's wort plants, which in their natural state at the site of the industrial zone in Lukavac had a lead concentration ranging from 0.019 mg/kg to 0.026 mg/kg, can be described by a mathematical quadratic function which quite realistically describes the given change in lead concentration.

Tests showed that there was a higher concentration of lead in the salve made from St. John's wort than in the oil according to the AAS method. The reason for the increased concentration of lead in the salve is due to the presence of heavy metals, including lead, in traces (in permitted concentrations) in the wax that was used to make the salve. The wax (Cera alba) contained 0.0080 mg/kg of lead, which was also measured by the AAS method (BAS EN 13804:2015 and BAS EN 14084:2005).

It is especially important to emphasize that the soil at the location of the soil samples and St. John's wort plants is mostly loamy soil with a low content of loam, so that the maximum allowable amounts of lead in such soil are from 0 mg/kg to 50 mg/kg. The soil samples had a lead concentration of 4.60 mg/kg to 14.06 mg/kg. During rainfall, the amount of rain quickly infiltrates this type of soil, and this leads to lead concentration in the subsoil and underground water. The level of underground water in the locality is at about 1.30 m during significant water saturation of the soil and up to 3.00 m during the dry period in the summer days. On the basis of these characteristics of the soil of the first level of groundwater, it can be concluded that there is a high possibility of contamination of groundwater with lead concentrations. Nearby there is a regulated bed of the Jala river as well as artesian wells for water supply to the population of Lukavac.

Conclusion

Based on the conducted analyses, it can be concluded that the concentration of lead in the soil at the sampling location in the industrial zone of Lukavac influenced the presence of lead in the oil and salve made from St. John's wort, however, very little lead was absorbed by St. John's wort. The reason for this is most likely the slightly basic character of the soil, since it is known that heavy metals, including lead, have better biological availability in more acidic soil. The concentration of lead in the soil could be lower compared to acidic soils, so

there is still a risk of its transfer to the plant species, in the context of this study St. John's wort, and further into products such as oil and salve, especially if it is taken into account its potentially greater bioavailability in acidic soils. The results showed that the concentration of lead in the salve is higher than in the oil, which indicates the possibility of a greater transfer of lead into these preparations, and potential risks for the health of people who use these products, in the event that St. John's wort comes from a more polluted environment with a higher lead content. Lead, as a highly toxic heavy metal, represents a serious environmental and health problem, especially in industrial zones such as Lukavac, where soil, water and vegetation pollution is obvious. St. John's wort is a widespread plant species, which is often used in Bosnian folk medicine, and St. John's wort is often used in home-made herbal preparations, which are not controlled for the content of heavy metals. These results emphasize the need for careful monitoring and quality control of medicinal plants used for the preparation of pharmaceutical preparations, in order to reduce as much as possible the potential risks of the presence of toxic metals in human nutrition and health. Of particular importance is the possible infiltration of rainwater through permeable soil with the presence of lead, which further contaminates groundwater, the level of which is high on days of higher precipitation. It is important to emphasize that at a distance of about 400 m as the crow flies, there are 17 artesian wells that supply most of the population of the city of Lukavac with water, and in this way the groundwater is additionally polluted.

There are no conflicting interests in this paper.

There was no funding for this paper.

Reference

1. Tong, S., von Schirnding, Y. E., & Prapamontol, T. (2000). Environmental lead exposure: a public health problem of global dimensions. Bulletin of the World Health Organization, 78(9), 1068–1077.
2. Šljivić Husejnović, M., Janković, S., Nikolić, D. i Antonijević, B. (2021). Human health risk assessment of lead, cadmium, and mercury co-exposure from agricultural soils in the Tuzla Canton (Bosnia and Herzegovina). *Arhiv za higijenu rada i toksikologiju, 72* (4), 268-279. https://doi.org/10.2478/aiht-2021-72-3533
3. Azra Suljić, & Nedim Suljić. (2021). Degradation of the Pharmaceutical and Medical Waste on Landfills Due to Rainwater Infiltration. International Journal for Research in Applied Sciences and Biotechnology, 8(1), 138–143. Retrieved from https://ijrasb.com/index.php/ijrasb/article/view/103
4. Suljić, A., Bečić, E., Husejnović, M.Š., Ademović, Z. (2024). St John's Wort as a Possible Tool for Remediation of the Soil Contaminated with Heavy Metals. In: Arslanagić-Kalajdžić, M., Ademović, N., Tufek-Memišević, T. (eds) Interdisciplinary Advances in Sustainable Development II. BHAAAS 2023. Lecture Notes in Networks and Systems, vol 804. Springer, Cham. https://doi.org/10.1007/978-3-031-46692-2_6
5. Glavač, N. K., Djogo, S., Ražić, S., Kreft, S., & Veber, M. (2017). Accumulation of heavy metals from soil in medicinal plants. *Arhiv za higijenu rada i toksikologiju*, *68*(3), 236–244. https://doi.org/10.1515/aiht-2017-68-2990
6. Kashem, M., Singh, B. Metal availability in contaminated soils: I. Effects of floodingand organic matter on changes in Eh, pH and solubility of Cd, Ni andZn. *Nutrient Cycling in Agroecosystems* **61**, 247–255 (2001). https://doi.org/10.1023/A:1013762204510
7. Adamczyk-Szabela, D., & Wolf, W. M. (2022). The Impact of Soil pH on Heavy Metals Uptake and Photosynthesis Efficiency in *Melissa officinalis*, *Taraxacum officinalis*, *Ocimum basilicum*. *Molecules (Basel, Switzerland)*, *27*(15), 4671. https://doi.org/10.3390/molecules27154671
8. Charkiewicz, A. E., & Backstrand, J. R. (2020). Lead Toxicity and Pollution in Poland. *International journal of environmental research and public health*, *17*(12), 4385. https://doi.org/10.3390/ijerph17124385
9. Sljivic Husejnovic, M., Bergant, M., Jankovic, S. *et al.* (2018). Assessment of Pb, Cd and Hg soil contamination and its potential to cause cytotoxic and genotoxic effects in human cell lines (CaCo-2 and HaCaT). *Environ Geochem Health* **40**, 1557–1572 (2018). https://doi.org/10.1007/s10653-018-0071-6
10. Suljić, N. (2022). Determination of Change in Coefficiency of Flow and Coefficient of Rainwater Infiltration into Body of Waste Landfill by Suljic Method. In: Karabegović, I., Kovačević, A., Mandžuka, S. (eds) New Technologies, Development and Application V. NT 2022. Lecture Notes in Networks and Systems, vol 472. Springer, Cham. https://doi.org/10.1007/978-3-031-05230-9_103
11. Overall evaluations of carcinogenicity: an updating of IARC Monographs volumes 1 to 42 (1987) IARC Monogr Eval Carcinog Risks Hum Suppl, 7:1-440
12. Dautović, E., Šljivić Husejnović, M., Bergant, M., Sabitović, D., Srabović, N., Smajlović, A., Begić, L., & Softić, A. (2019). Lead and cadmium induced cytotoxic and genotoxic effects on HL-60 and Jurkat cell lines. *Genetics & Applications*, *3*(1), 57–64. https://doi.org/10.31383/ga.vol3iss1pp57-64
13. Barciszewska, M. Z., Szymanski, M., Wyszko, E., Pas, J., Rychlewski, L., & Barciszewski, J. (2005). Lead toxicity through the leadzyme. *Mutation research*, *589*(2), 103–110.

https://doi.org/10.1016/j.mrrev.2004.11.002

14. Ali AS, Bayih AA, Gari SR. Meta-analysis of public health risks of lead accumulation in wastewater, irrigated soil, and crops nexus. Front Public Health. 2022 Oct 18;10:977721. doi: 10.3389/fpubh.2022.977721. PMID: 36330130; PMCID: PMC9623109.
15. Halmo, L., & Nappe, T. M. (2023). Lead Toxicity. In *StatPearls*. StatPearls Publishing.
16. Wani AL, Ara A, Usmani JA. Lead toxicity: a review. Interdiscip Toxicol. 2015 Jun;8(2):55-64. doi: 10.1515/intox-2015-0009. PMID: 27486361; PMCID: PMC4961898.
17. Begić A., Šljivić-Husejnović M., Aščerić M., Brekalo-Lazarević S. (2019) Uvod u toksikologiju: eksperimentalna toksikologija s teoretskim osnovama, Tuzla. ISBN 978-9958-894-67-1
18. Kladar N., Srđenović B., Grujić N., Bokić B., Rat M., Anačkov G., et al. (2015) Ecologically and ontogenetically induced variations in phenolic compounds and biological activities of Hypericum maculatum subsp. maculatum, Hypericaceae. Brazilian Journal of Botany, 38(4):703-715.
19. Lukić P. (1985). Farmakognozija, 3. dopunjeno izdanje. Farmaceutski fakultet Beograd.
20. Barnes, J., Anderson, L. A., & Phillipson, J. D. (2001). St John's wort (Hypericum perforatum L.): a review of its chemistry, pharmacology and clinical properties. The Journal of pharmacy and pharmacology, 53(5), 583–600. https://doi.org/10.1211/0022357011775910
21. Klemow, K. M., Bartlow, A., Crawford, J., Kocher, N., Shah, J., Ritsick, M. (2011) Medical Attributes of St. John's Wort (Hypericum perforatum). In I. Benzie (Eds.) et. al., Herbal Medicine: Biomolecular and Clinical Aspects. (2nd ed.). CRC Press/Taylor & Francis.
22. Wölfle, U., Seelinger, G., & Schempp, C. M. (2014). Topical application of St. John's wort (Hypericum perforatum). Planta medica, 80(2-3), 109–120. https://doi.org/10.1055/s-0033-1351019
23. Wurglics, M., & Schubert-Zsilavecz, M. (2006). Hypericum perforatum: a 'modern' herbal antidepressant: pharmacokinetics of active ingredients. *Clinical pharmacokinetics*, *45*(5), 449–468. https://doi.org/10.2165/00003088-200645050-00002
24. Maury, W., Price, J. P., Brindley, M. A., Oh, C., Neighbors, J. D., Wiemer, D. F., Wills, N., Carpenter, S., Hauck, C., Murphy, P., Widrlechner, M. P., Delate, K., Kumar, G., Kraus, G. A., Rizshsky, L., & Nikolau, B. (2009). Identification of light-independent inhibition of human immunodeficiency virus-1 infection through bioguided fractionation of Hypericum perforatum. Virology journal, 6, 101. https://doi.org/10.1186/1743-422X-6-101
25. Suryawanshi, M. V., Gujarathi, P. P., Mulla, T., & Bagban, I. (2024). Hypericum perforatum: a comprehensive review on pharmacognosy, preclinical studies, putative molecular mechanism, and clinical studies in neurodegenerative diseases. *Naunyn-Schmiedeberg's archives of pharmacology*, *397*(6), 3803–3818. https://doi.org/10.1007/s00210-023-02915-6
26. Yu P. H. (2000). Effect of the Hypericum perforatum extract on serotonin turnover in the mouse brain. Pharmacopsychiatry, 33(2), 60–65. https://doi.org/10.1055/s-2000-7970