



Productivity of bee families and biomonitoring of corbicular pollen and war-affected honeybee foraging sites with cultivated honey clover (*Melilotus albus*)

S. F. Razanov*, I. I. Ibatulin**, O. S. Razanov***, A. I. Dydiv****,
M. V. Voynalovich**, H. A. Lysak***, M. J. Lopotych***

*Vinnytsia National Agrarian University, Vinnytsia, Ukraine

**National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine

***National Scientific Center “Institute of Beekeeping named after P. I. Prokopovich”, Kyiv, Ukraine

****Lviv National Environmental University, Dublyany, Ukraine

Article info

Received 09.01.2024

Received in revised form 01.02.2024

Accepted 27.02.2024

Vinnytsia National Agrarian University,
Sonyachna st., 3, Vinnytsia, 21008, Ukraine.
Tel.: +38-096-353-25-12.
E-mail: razanovsergej65@gmail.com

National University of Life and Environmental
Sciences of Ukraine, Heroiv Oborony st.,
15, Kyiv, 03041, Ukraine. Tel.: +38-044-258-
42-63. E-mail: vstuf@mubip.edu.ua

National Scientific Center “Institute
of Beekeeping named after
P. I. Prokopovich”, Zabolotnogo st., 19, Kyiv,
03680, Ukraine. Tel.: +38-096-020-22-92.
E-mail: razanovoleksandr@gmail.com

Lviv National Environmental University,
V. Velikoho st., 1, Dublyany, 80381, Lviv
region, Ukraine. Tel.: +38-097-45-00-572.
E-mail: adydiv@gmail.com

Razanov, S. F., Ibatulin, I. I., Razanov, O. S., Dydiv, A. I., Voynalovich, M. V., Lysak, H. A., & Lopotych, M. J. (2024). Productivity of bee families and biomonitoring of corbicular pollen and war-affected honeybee foraging sites with cultivated honey clover (*Melilotus albus*). *Regulatory Mechanisms in Biosystems*, 15(1), 171–176. doi:10.15421/022425

As a result of military actions, some territories of Ukraine have suffered technogenic loading on the environment. Of special concern is disturbed agricultural soils that are significant for food safety of the country. Soils affected by military actions require control of contamination and restoration of fertility. The studies revealed the advantages of cultivating honey clover (*Melilotus albus*) in war-affected soils and controlling content of heavy metals in soils and corbicular pollen from honey clover cultivated in those soils. In gray forest average-loamy soils that had been affected by military actions, nitrogen content increased 6.3% thanks to cultivation of *M. albus*. High nitrogen content in soil improves its fertility. Cultivation of *M. albus* in affected soils promoted more intensive growth of the bee families, encouraging them to gather nectar more actively. As a result, this apiary produced greater amounts of honey, corbicular pollen, and bee pollen. The conducted studies revealed heightened levels of lead, cadmium, and zinc in the war-impacted agricultural land, compared with the territory beyond the impact zone. This caused increased content of heavy metals in corbicular pollen from *M. albus*. Contamination of corbicular pollen from *M. albus* manifested in excess over the allowable norms of cadmium, lead, and zinc. At the same time, the highest coefficient of transfer from soil into corbicular pollen was found for zinc. The high degree of contamination of the soil with heavy metals as a result of military action promoted a high level of ingress of lead and cadmium into the corbicular pollen, compared with the situation in clean territory. The study results expand the available data on biomonitoring of apiary products and foraging lands in contaminated areas. Analysis of the corbicular pollen and pollen can give information about the level of environmental contamination and help evaluate the ecological state of melliferous sites. Such a monitoring can be useful for decision making regarding the protection of the environment and healthcare.

Keywords: corbicular pollen; soil; *Melilotus albus*; lead; cadmium; zinc; bee families; honey; brood.

Introduction

Ecologically safe beekeeping products are becoming increasingly important in Ukraine and many countries around the world. Producers of products from apiaries emphasize the eco-friendliness of their products, thus attaining a competitive advantage on the market, because the consumers seek high-quality and safe products. Anthropogenic contamination of the air, including with toxic elements such as heavy metals, can negatively impact apiary products. This is especially relevant due to industrial processes, transport, military actions, and other contamination sources. Mutual efforts towards constant control of anthropogenic loading are directed at sustaining the quality and ecological pureness of beekeeping products.

The war in Ukraine has brought devastating ecological problems and inflicted damage on nature, causing large-scale and long-lasting environmental degradation. Military actions have caused rapid changes in the landscapes, raising doubts regarding safety of using the lands affected. All those problems can have serious implications for people's health, economics, and nature, and require attention and measures that would mitigate the harm and help the regions recover their ecological resilience. Controlling the negative impact of military action on the productivity of

soils is crucial. Those circumstances require complex scientific research focusing on the state of soils in war zones. Such studies can determine the criteria of land contamination and serve as groundwork for developing policy of managing war-ravaged territories. Military action imposes technogenic impacts on the environment, including mechanical, physical, and chemical effects, mostly on soil cover (Ali et al., 2014). According to some experts, the total of damage inflicted on Ukraine over the year has increased five-fold. As is known, explosions, fires, and detonations, caused by shell explosions and shelling, lead to accumulation of various contaminants in the soil (Barker et al., 2021). The radius of shell-explosion impacts is accompanied by a shock wave, spreading toxicants to large distances, depending on the mass of the explosive (Diaz & Massol-Deya, 2003; Cristaldi et al., 2013; Golubtsov et al., 2023).

Hazardous toxic contaminants include heavy metals that enter the atmosphere and settle on the soil environment (Dydiv et al., 2023). Heavy metals are the most dangerous to nature, are highly toxic, and exert carcinogenic and mutagenic properties (Brygadyrenko & Ivanyshyn, 2015; Kozak & Brygadyrenko, 2018). In war-damaged territories, heavy metals have been tallied in large amounts (Jergovic et al., 2010; Jirau-Colon et al., 2019), and integrity of soils was observed to be deteriorated, which reduces fertility (Massol-Deyá et al., 2005). Compounds brought onto the soil

surface by military actions are characterized by various levels of water solvability. They intensively infiltrate the horizontal and vertical layers of the soil environment. The highest concentration of toxicants was found in soil at 15 cm depth. Intensity of migration of toxicants completely depends on duration of their stay in the soils, type of soil, characteristics of contaminants, and other factors (Mazur, 1997).

Studies by Rawtani et al. (2022) have confirmed a close relationship between the level of environmental contamination and heavy metals in people and level of morbidity in the conditions of military-technogenic loading. Heightened levels of lead, cadmium, aluminium, and rat poison have been found in urine, blood, and hair of the population living in the zone of war-caused technogenic impact.

Deterioration of soil qualities is long-lasting and affects its productive functions critically. Destruction of vegetation, damages to soil cover, loss of natural moisture are all common consequences of war and technogenic loading (Hu et al., 2013; Skalny et al., 2021; Omelchuk, 2022; Holubtsov et al., 2023). Those processes drastically decrease the biodiversity, which in turn affects the biological populations and species. Loss of biodiversity enhances alterations in the structure and functions of landscapes. Contaminants, heavy metals in particular, pose a great threat due to their ability to migrate to plants and accumulate in large concentrations. As is known, phytoindicator plants can accumulate in their vegetative mass hundreds- and even thousands-fold more toxicants than their concentrations in the soil, depending on their botanical origin (Razanov et al., 2018, 2020). Technogenic impact of military action drastically intensifies accumulation of heavy metals in vegetation (Pereira et al., 2022; Razanov et al., 2023). Shell explosions, burning, and detonations, accompanying military hostilities, release various contaminants, including heavy metals, into the soil, from where they travel into the plants. Studies (Robinson et al., 2008) suggest that repercussions of military action are expressed in high levels of heavy metals in plants that grow in the affected territories. From plants, heavy metals travel into food products, endangering human health.

Honey bees perfectly meet the criteria of bioindicators and, together with products of their activity, are unique objects for research (Celli & Maccagnani, 2003). Using them, one can expand the set of ecological characteristics to assess the state of the environment. Over the process of evolution, bees developed the adaptability to limited food availability. They provide themselves with all the necessary nutrients, such as proteins, fats, carbohydrates, mineral compounds, vitamins, and other, by consuming plant nectar and flower pollen. Bees closely interact with nectar-pollen plants, pollinating them and obtaining products they need for nutrition (Ayansola et al., 2012; Macukanovic-Jocic & Jaric, 2016; Bakour et al., 2021; Bezpalyi et al., 2021).

Production of honey bees such as honey, wax, corbicular pollen, are analyzed to detect toxicants, which can help in assessments of the quality of the environment. By collecting nectar and pollen from flowers, bees can accumulate heavy metals in their nests (Munir et al., 2022). Those metals enter the apiary products, which can be thus harmful to human health when consumed. High concentrations of heavy metals in apiary products also show the level of soil contamination and systematic presence in plants. Studies revealed that in zones of technogenic impacts, apiary products contain heavy metals in large quantities (Crane, 1984; Kovalchuk & Fedoruk, 2013; Dubin & Vasylenko, 2017). A greater degree of contamination was identified for pollen and bee pollen, which, according to some sources, had higher concentrations of heavy metals than honey. Kovalchuk & Fedoruk (2008) determined that concentration of cadmium in corbicular pollen made of pollen from dandelion growing in an industrial zone exceeded the level in a relatively clean area, ranging 1.2 to 3.2 times. Also, significant differences were found between concentrations of heavy metals in pollen from dandelion and buckwheat.

Richness of nectar-pollen area is determined by the level of production of nutrients for bees (nectar and flower pollen) and how actively bees harvest those products (Razanova et al., 2021). Those products affect the intensity of development of the bees and their productivity. The level of provision of bees with nectar and flower pollen depends on the presence of nectar-pollen flowers and their abundance. Dense populations of *M. albus* can provide production of up to 200 kg of nectar and 120 kg of flower pollen per 1 ha (Lavrinenko et al., 2019). This is important for providing bees with resources necessary for their health and activity.

To alleviate the effects of anthropogenic contamination on bee keeping and apiary products, it is important to take measures to reduce emissions of heavy metals into the atmosphere. This may include introduction of ecologically clean production technologies, restriction of using harmful compounds, and other measures to reduce emission of contaminants. In addition, it is important to monitor the levels of heavy metals in apiary products so as to ensure their safety for the consumers. This allows timely identification of possible contaminations and taking counter measures, promoting production of ecologically safe apiary goods. Such conditions require seeking ways and designing methods to effectively control technogenic loading on soil of agricultural lands and restore and sustain their fertility when subject to high risks.

The objective of the study was to analyze the productivity of honey bees and biomonitor corbicular pollen and foraging lands affected by military action in the conditions of cultivating *M. albus*.

Materials and methods

The study of the efficiency of using the method of controlling technogenically contaminated soils of agricultural lands and increasing their fertility in the period of rehabilitation was conducted in 2022–2023 in the territory of Vinnytsia Oblast that had been affected by a missile attack (GPS coordinates: 49.16725° N, 28.37065° E). Soils in the studied territories are gray forest average-loamy. The study objects were soils of foraging lands affected by military actions, and soils of foraging lands beyond the impact range, productivity of bee families, and intensity of accumulation of heavy metals in corbicular pollen from *M. albus*.

Apiaries of five bee families in a group were located at the edge of each field at a 7 km distance from each other. Thus, we tried to limit the overlap of flying ranges of the honey bees in the fields with various melliferous plants so that pollen collected from each apiary could be considered independent. The bee families were kept in long hives. Bees of the experimental group used the foraging land that had been affected by the rocket strike. Bee families of this group were located in safe territory, 1 km away from the *M. albus* field, cultivated in the affected area. Bee families of the control group were located at 7 km distance from the war-disturbed zone.

Melilotus albus was sown in soils of foraging lands using agricultural drones in early spring during high soil moisture. Honey clover is a representative of melliferous plants in the legumes family and can reach the height of 90 to 150 cm (Lavrinenko et al., 2019; Zhang et al., 2019). Honey clover blossoms for quite a while, starting from mid June, lasting for 40–50 days, until mid August, and sometimes even longer. From the flowers of *M. albus*, the bees collected nectar and pollen. The plant adapts well to weather conditions and requires no special care and grows easily in non-cultivated lands (Zabala et al., 2018; Kintl et al., 2021). Honey clover is highly resistant to negative natural climatic conditions. The plant is highly productive, one plant can produce up to 10 thousand seeds. This allows it to quickly recover dense populations, displacing other plants. It is important to note that after forming dense populations, *M. albus* can continue to grow and develop with no need for repeated sowing.

Development of bee families was monitored by accounting sealed bee broods, assessed using the modified system of frame grid. For this purpose, from a honeycomb with a sealed bee brood, the bees were shaken off and using a grid frame, we measured its area. The grid frame was divided into 5 x 5 cm squares, each having the area of 25 cm². Afterwards, we estimated the general area of the honeycombs. The frame was attached to the honeycomb with bee brood, and first, we estimated the number of complete squares occupied by the brood, and then – the number of incomplete ones, in order to convert the latter squares into complete ones. One square of the grid frame could fit 100 bees and 75 drone cells with sealed brood. The count was conducted every 12 days.

The amounts of capped honey and bee pollen that were in the honeycombs of the bee nests were determined according to difference in weight of a honeycomb with the products and empty honeycomb (0.7 kg). The amount of centrifuged honey was determined on the CAS SW II-15 scales after pumping it (third decade of August) from the honeycombs. Full honeycombs with 1/3 sealed chambers were selected from each beehive. Then, the beeswax caps of the honeycombs were opened using a PROFI 220B electric uncapping knife. Then, the honeycombs were put in

an AISI 430 honey extractor and honey was pumped. Corbicular pollen was collected using pollen traps that were installed on the front wall of the beehives. Prior to sampling corbicular pollen, we prepared the bee families. For bees to adapt, the grid in the pollen traps was not used for the first two-three days. Then, on the 2–3rd days, we installed a tray and a grid. The pollen catchers were open to harvest on the 24th h. We gathered corbicular pollen from the trays of the pollen catchers every day and weighed it on the electronic scales BTU2100 AXIS. For the subsequent taxonomic identification, corbicular pollen was kept at -20°C . Botanical origin of the corbicular pollen was identified based on the morphological features of pollen grains. The selected corbicular pollen constituted a mixture of various plants, and therefore we performed studies to determine its species composition. When identifying pollen origins, we took into the account the color of the corbicular pollen, i.e. it was sorted by color that corresponded to *M. albus*. For an accurate classification of the samples of corbicular pollen, we identified pollen grains, shot on a videocamera with a Granum binocular microscope, determining the main parameters of pollen grain by comparing images with the standard samples. The studied samples of corbicular pollen were compared to the tables of colors of corbicular pollen from various plants (Kirk, 2006; Bartlet, 2021; Hodges, 2021). The sorting was carried out at electric light.

To measure the content of heavy metals in corbicular pollen, it was dried at the temperature of 41°C and stored in hermetically closed glass container. Samples for the laboratory study were taken using the method of point sampling from each bee family, 150 g from each.

Soil sampling was conducted using the envelope method, that is, in five places of one field (in the corners of a square and in its center), we sampled soil at 0–25 cm depth. Distance between the sampling points was 100 m. Nitrogen in the soil was measured in 4 repetitions. Soil for this purpose was sampled prior to sowing *M. albus* (third decade of March) and after its vegetation period – third decade of September.

Content of lead, cadmium, and zinc in the corbicular pollen and soil was identified using a Quant 2 AT atomic-absorption spectrometer (DSTU 4770.1-9:2007). Results of the study were subjected to statistical analysis using ANOVA. Statistically significant differences between the groups were identified at the threshold value of $P < 0.05$ taking into the account Bonferroni's Correction. The data in the tables are presented as $x \pm \text{SD}$ (mean and standard deviation).

Results

Improvement of the missile-struck honeybees' pasture by cultivating *M. albus* had an effect on the number of broods grown by the bee families during the blossom of the honey bearer. In particular, thanks to growing *M. albus*, bee families of the second group, located in the foraging site that had been affected by military actions, reared more brood in the period from June 1 to August 23 (Table 1).

At the beginning of the account period (1.06), productivity of the bee families by number of reared brood was almost equal in groups. The start of blossom of *M. albus* in lands affected by military actions stimulated the

bees to intensive brood rearing and already on the third day of the account (6/24) the advantage according to this parameter in the experimental group equaled 13.3% ($P < 0.001$). Increase in the amount of flowering plants encouraged bees in both groups to more intensive rear brood up to June 18, although in the experimental group this parameter was 24.9% higher ($P < 0.001$) than in the control. Already in late July, the blossom intensity of the honey clover declined and therefore the bees started to rear much smaller broods. Decrease in brood size in this period occurred in both groups, measuring 21.4% in the control and 16.3% in the experimental group, compared with the previous date. Bees of the experimental group maintained their activity in brood rearing, having a 33.0% advantage ($P < 0.001$), and did not lose their potential until the end of the account period. At the end of the account period and blossom of *M. albus*, the bee families in the war-affected territory reared 8.0% ($P < 0.05$) more brood.

Table 1

Dynamics of rearing brood by the bee families in the war-affected territory with *M. albus* cultivated (cm^2 , $x \pm \text{SD}$, $n = 5$)

Date of account of <i>M. albus</i> blossom	Foraging area beyond war impact	Foraging area affected by military actions
June 01	3555 \pm 47	3588 \pm 43
June 12	4211 \pm 39	4233 \pm 47
June 24	5277 \pm 52	5981 \pm 66***
July 06	6266 \pm 84	7646 \pm 91***
July 18	7283 \pm 91	9096 \pm 104***
July 30	5725 \pm 74	7617 \pm 81***
August 11	3909 \pm 83	5630 \pm 105***
August 23	3555 \pm 78	3840 \pm 60***

Note: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ within each row according to ANOVA.

Cultivation of *M. albus* in foraging lands affected by military action promoted increase in the productivity of the bee families in brood rearing, which led to increase in the number of bees and the amounts of honey they produced, corbicular pollen, and bee pollen (Table 2).

During the account period, bees of the experimental group that had been living in the war-affected territory, from 6/01 to 8/23 reared 20.2% ($P < 0.001$) more brood. Bee families of the experimental group produced 15.3% ($P < 0.01$) more total honey and 43.2% more centrifuged honey ($P < 0.001$), compared with bee families of the control group. For the winter period, the bees of the experimental group prepared a somewhat smaller amount of capped honey – by 5.4%. Growing a larger brood stimulated bees of the experimental-group families to collect a larger amount of corbicular pollen and prepare more bee pollen. Therefore, bee families that had been in the lands affected by military actions and used pollen from *M. albus* produced 21.9% ($P < 0.05$) more corbicular pollen and 32.1% ($P < 0.01$) more bee pollen.

Military technogenic impact contaminated the soil with heavy metals. Soil of the missile-hit foraging land contained 3.7 times more lead ($P < 0.001$), 2.7 times more cadmium ($P < 0.001$), and 1.9 times more zinc ($P < 0.001$), compared with soil beyond the impact range (Table 3).

Table 2

Productivity of bee families in war-affected foraging area with *M. albus* ($x \pm \text{SD}$, $n = 5$)

Parameter	Foraging lands beyond war impact	Foraging area affected by military actions
Brood grown in the account period, cm^2	39781 \pm 1445	47814 \pm 608***
Capped honey in honeycombs, kg	7.02 \pm 0.53	6.28 \pm 0.59
Centrifuged honey, kg	6.48 \pm 0.11	9.28 \pm 0.31***
Total production of honey, kg	13.50 \pm 0.24	15.56 \pm 0.41***
Corbicular honey, kg	0.639 \pm 0.034	0.782 \pm 0.047***
Bee pollen, kg	0.811 \pm 0.013	1.067 \pm 0.038***

Note: see Table 1.

Table 3

Intensity of accumulation of heavy metals in corbicular pollen and soils of foraging area affected by war ($x \pm \text{SD}$, $n = 4$)

Heavy metals	Concentrations of heavy metals					
	foraging area outside the war-impact zone			foraging area affected by military actions		
	corbicular pollen, mg/kg	soil, mg/kg	accumulation coefficient	corbicular pollen, mg/kg	soil, mg/kg	accumulation coefficient
Lead	0.0714 \pm 0.0046	2.47 \pm 0.13	0.0289 \pm 0.0036	1.114 \pm 0.078***	9.24 \pm 0.17***	0.0577 \pm 0.0012***
Cadmium	0.0423 \pm 0.0028	0.633 \pm 0.024	0.0671 \pm 0.0035	0.281 \pm 0.026***	1.730 \pm 0.067***	0.0753 \pm 0.0021***
Zinc	1.244 \pm 0.092	2.32 \pm 0.10	0.536 \pm 0.014	2.25 \pm 0.17***	4.41 \pm 0.38***	0.510 \pm 0.017***

Note: see Table 1.

Keeping bee families in war-impacted lands and cultivating *M. albus* in contaminated soil led to a considerable contamination of corbicular pollen from *M. albus* with heavy metals. Lead concentration in corbicular pollen from *M. albus* was 1.114 mg/kg, which was 15.6-fold higher ($P < 0.001$) compared with such from the foraging area beyond the missile impact. Cadmium content in the contaminated territory was 0.281 mg/kg against 0.0423 mg/kg, which was 6.6-fold higher ($P < 0.001$). Concentration of zinc in corbicular pollen from the territory affected by military actions equaled 2.25 mg/kg, 1.8-fold higher ($P < 0.01$) than such from the unaffected foraging land. Inferring from the data regarding the coefficient of accumulation of heavy metals in corbicular pollen, the highest transfer coefficient from the war-impacted territory was seen for zinc (0.510) and the lowest for lead – 0.0577. From the clean foraging site, the accumulation coefficient was the highest for zinc (0.536) and the lowest for lead (0.0289). However, the coefficients of transfer of lead and cadmium from soil into corbicular pollen were 2.0-fold ($P < 0.001$) and 12.2% higher ($P < 0.05$), respectively, from the foraging area affected by military action. The transfer coefficient of zinc was, by contrast, higher from foraging land beyond the impact of military-action.

According to the results, having cultivated *M. albus* in the soil of the rocket-hit foraging ground had an effect on soil fertility. Nitrogen content was 2.4% higher in the first year of crop cultivation and 3.8% higher ($P < 0.05$) in the second year as compared with the soil samples gathered prior to cultivating the plant (Table 4).

Table 4
Effects of *M. albus* on content of nitrogen in soils of foraging site affected by military actions ($x \pm SD$, $n = 5$)

Soil	Nitrogen content, mg/kg
Prior to sowing <i>M. albus</i>	127.2 ± 1.4 ^a
At the end of the first year of cultivating <i>M. albus</i>	130.4 ± 1.3 ^b
At the end of the second year of cultivating <i>M. albus</i>	135.1 ± 1.6 ^c

Note: see Table 1.

Over the two years of cultivating *M. albus* in the affected soils, the amount of nitrogen increased by 6.3% ($P < 0.01$).

Discussion

In modern conditions, where the main priority is the principle of ecological rationality, there is ongoing active development and implementation of systems, technologies, and methods oriented at producing organic goods in the spheres of crop farming and bee keeping (Bayir & Aygun, 2022). Heavy metals are considered dangerous contaminants, because their spread and migration in the environment can negatively impact on ecosystems and human health. In this context, it is necessary to come up with strategies of regulating their concentrations in the system “soil–plant–products”, based on interrelated and interdependent processes of this circulation (Kastrati et al., 2021). Soils constitute natural accumulators of heavy metals in the environment and are the main source of contamination of plants. Around 90% of heavy metals that occur in the environment are concentrated in soil, and then migrate into natural waters, are absorbed by plants and become included into the trophic chains. The end link of those chains is the human organism.

The objective of our studies was cultivating *M. albus* in territory disturbed by military actions, which was a bioindicator of soil pollution, estimated through its pollen collected by the bees. Vegetative mass of *M. albus* and its nitrogen-fixating properties were used to recover soil fertility (Zhang et al., 2019; Kintl et al., 2021). Growing *M. albus* had positive effects on the soil, in particular thanks to nitrogen fixation. This process is important for enrichment of soil with nitrogen, which in turn promotes fertility and supports a sustainable agroecosystem. Honey clover is a good variant of green manure for nitrogen accumulation in the soil. This plant has a powerful system of roots that stabilizes the soil and improves its structure. Moreover, *M. albus* can fixate nitrogen from the air with nitrogen-fixating bacteria living in its roots. After decomposition of *M. albus* plant mass, nitrogen that it had accumulated becomes available to other plants growing in the soil. Thus, cultivation of *M. albus* can substantially increase the nitrogen level and improve the structure and fertility of soil.

Over the two years of *M. albus* cultivation in war-affected land, the nitrogen content there increased by 6.3%.

Over the process of evolution, bees had adapted to having limited resources to provide themselves with food. By gathering all necessary nutrients, such as proteins, fats, carbohydrates, minerals, and vitamins, from nectar and flower pollen, bees acquire essential nutrition elements. Therefore, bees live in close interaction with nectar-pollen plants, pollinating them and at the same time obtaining necessary products for vitality of their families. One of the indicators of substantial nectar-pollen resources is their development, expressed in increased amount of reared brood and strengthened families. This relationship indicates a successful interaction between bees and plants, where both sides mutually benefit. Identification of the amount of melliferous resources and how effectively bee families utilize them is not limited to assessing development, i.e. brood rearing. An important parameter is also production of various products such as honey, corbicular pollen, and other. Hladun et al. (2016) believe that apiaries that are close to territories contaminated with heavy metals can be subject to certain harm and suffer a negative impact due to changes in the dynamics and survival of the bee families. Use of honey clover in the foraging ground in war-impacted territory improved the productive parameters of the bee families. Accordingly, bee reared 20.2% more brood, prepared 15.3% more honey, 21.9% more corbicular pollen, and 32.1% more bee pollen.

Heavy metals negatively affect vitality of honey bees, development of brood, and species diversity. According to Verma et al. (2023) and Zajdel et al. (2023), *Apis mellifera* can be a marker for monitoring environmental contamination with heavy metals, because it reflects presence and number of those metals in plants around apiaries. Honeybees' pastures are essential to the quality and safety of apiary products, and their current state is determined by the presence of technogenic contamination, including with heavy metals (Verma et al., 2023). Quality and safety of flower pollen and products from it processed by bees, such as corbicular pollen, bee pollen, and homogenate to a high degree depend on ecological state of the melliferous lands, in particular level of soil contamination (Rewiev et al., 2023).

Different species of plants exert various abilities to absorb lead. Cereals and buckwheat have a tendency towards high lead accumulation. Among wild-growing plants, high absorption of this metal was seen for *Mentha piperita*, *Matricaria recutita*, leaves of *Convallaria majalis*, *Leonurus*, *Melilotus officinalis*, and *Tussilago farfara*. Kovalchuk & Fedoruk (2013) believe that agroecological conditions of agricultural lands remote from an industrial center provide the lowest content of heavy metals in apiary products (corbicular pollen and honey), whereas corbicular pollen and bee pollen from sunflower were noted to contain high amounts of lead and cadmium. Increases in iron, zinc, cuprum, chromium, nickel, lead and cadmium depending on location of beehive were confirmed by the studies of Klym (2017).

The results of the study by Rivis et al. (2022) suggest a significant increase in the level of cadmium in corbicular pollen and pollen from *Taraxacum officinale* and apple tree compared with relatively clean mountainous environment. Cadmium accumulated 2.5–4.0-fold more in those products. Pollen from *T. officinale* was found to be an active accumulator of heavy metals, concentrating twice the amount present in pollen from apple trees. Rivis et al. (2022) reported that zinc had a high coefficient of transfer from tilled soil into corbicular pollen and pollen from dandelion and apple tree, while lead had somewhat lower coefficients. Heavy metals that are in available form in soil, become included in the process of plants' nutrition and amass in their products, leading to their poorer quality and safety (Butsiak & Pecha, 2007; Toth et al., 2016). In the studied territories, we found a negative impact of military actions, where content of heavy metals (lead, cadmium, and zinc) increased, in turn contaminating corbicular pollen. The study results indicate significant excess of heavy metals in corbicular pollen from *M. albus* cultivated in soils in melliferous lands affected by military actions. The highest intensity of transfer from soil into corbicular pollen was observed for zinc. High concentrations of lead and cadmium in corbicular pollen exceeded the allowable thresholds and concentrations for most plant products outlined in the International Food Standard in the Codex Alimentarius of 1995, amended as of 2019 (Codex Alimentarius 1995 Amended, www.fao.org/fao-who-codexalimentarius). Higher coefficient of transfer into corbicular pollen was seen for lead and

cadmium from soil in melliferous lands that had been affected by military actions.

Conclusions

Cultivation of *M. albus* in soils of war-affected melliferous lands promoted improvement of soil fertility, increase in productivity of the bee families, but at the same time an increase occurred in the content of heavy metals in corbicular pollen from *M. albus*. During the beekeeping season, the bees reared 20.2% more brood, prepared 15.3% more honey, and 21.9% more corbicular pollen, and 32.1% more bee pollen. The coefficients of transfer of cadmium and lead from soil into corbicular pollen were higher from war-affected melliferous lands. At the same time, transfer of zinc was the highest among those metals. In soil, the nitrogen content increased 6.3% as a result of cultivating *M. albus*. Further studies will focus on analyzing quality of honey from war-affected areas contaminated with heavy metals according to concentrations of heavy metals.

The authors declare no conflict of interests regarding their report and results.

References

- Ali, A., Zinnert, J. C., Muthukumar, B., Kumar, M., & Peng, Y. (2014). Physiological and transcriptional responses of *Baccharis halimifolia* to the explosive "composition B" (RDX/TNT) in amended soil. *Environmental Science and Pollution Research*, 21, 8261–8270.
- Ayansola, A. A., Davies, B. A., & Davies, B. A. (2012). Honeybee floral resources in Southwestern Nigeria. *Journal of Biology and Life Science*, 3(1), 127–139.
- Bakour, M., Laaroussi, H., Menyiy, N. E., Elaraj, T., Ghouzi, A. E., & Lyoussi, B. (2021). The beekeeping state and inventory of mellifero-medicinal plants in the north-central of Morocco. *Scientific World Journal*, 2021, 9039726.
- Barker, A. J., Clausen, J. L., Douglas, T. A., Bednar, A. J., Griggs, C. S., & Martin, W. A. (2021). Environmental impact of metals resulting from military training activities: A review. *Chemosphere*, 265, 129110.
- Bartlett, J. (2021). *The pollen landscape*. Northern Bee Books, West Yorkshire.
- Bayir, H., & Aygun, A. (2022). Heavy metal in honey bees, honey, and pollen produced in rural and urban areas of Konya province in Turkey. *Environmental Science and Pollution Research*, 29, 74569–74578.
- Bezpalyy, I. F., Postoienko, V. O., & Polishchuk, A. A. (2021). Biotechnological factors of bees' ethology during nectar collection. *Bulletin of Poltava State Agrarian Academy*, 2, 188–193.
- Brygadyrenko, V., & Ivanyshyn, V. (2015). Changes in the body mass of *Megaphyllium kievense* (Diplopoda, Julidae) and the granulometric composition of leaf litter subject to different concentrations of copper. *Journal of Forest Science*, 61(9), 369–376.
- Butsiak, V. I., & Pechar, N. P. (2007). Vazhki metaly v grunti ta kharchovykh produktakh PP "Berezhyntsia" [Heavy metals in soil and foodstuff in private agricultural enterprise "Berezhyntsia"]. *Naukovyi Visnyk L'vivs'koho Natsional'nogo Universytetu Veterynarnoyi Medytsyny ta Biotekhnolohiyi imeni S. Z. Hzhyskoho*, 3, 121–125 (in Ukrainian).
- Celli, G., & Maccagnani, B. (2003). Honey bees as bioindicators of environmental pollution. *Bulletin of Insectology*, 56(1), 137–139.
- Crane, E. (1984). Bees, honey and pollen as indicators of metals in the environment. *Bee World*, 55, 47–49.
- Cristaldi, M., Foschi, C., Szpunar, G., Brini, C., Marinelli, F., & Triolo, L. (2013). Toxic emissions from a military test site in the territory of Sardinia, Italy. *International Journal of Environmental Research and Public Health*, 10(4), 1631–1646.
- Diaz, E., & Massol-Deya, A. (2003). Trace element composition in forage samples from a military target range, three agricultural areas, and one natural area in Puerto Rico. *Caribbean Journal of Science*, 39, 215–220.
- Dubin, O. M., & Vasylenko, O. V. (2017). Otsinka yakosti produktsii bdzhilnytstva v suchasnykh ekolohichnykh umovakh Cherkaskoyi oblasti [Assessment of the quality of beekeeping products in modern ecological conditions of the Cherkasy region]. *Visnyk Umans'koho Natsional'nogo Universytetu Sadivnytstva*, 1, 12–17 (in Ukrainian).
- Dydiv, A., Piddubna, A., Gucol, G., Vradii, O., Zhylyshchych, Y., Titarenko, O., Razzanova, A., Odnosum, H., Postoienko, D., & Kerek, S. (2023). Accumulation of lead and cadmium by vegetables at different levels of gray forest soil moistening in the conditions of the right bank forest steppe of Ukraine. *Journal of Ecological Engineering*, 24(10), 198–204.
- Hladun, K. R., Di, N., Liu, T. X., & Trumble, J. T. (2016). Metal contaminant accumulation in the hive: Consequences for whole-colony health and brood production in the honey bee (*Apis mellifera* L.). *Environmental Toxicology and Chemistry*, 35(2), 322–329.
- Hodges, D. (2021). The pollen grain drawings of dorothy hodges: Taken from the pollen loads of the honeybee. *International Bee Research Association*, Monmouth.
- Holubtsov, O., Sorokina, L., Splodytel, A., & Chumachenko, S. (2023). Vplyv viiny rosiyi proty Ukrainy na stan ukrainskykh gruntiv. Rezultaty analizu [The impact of Russia's war against Ukraine on the state of Ukrainian soils. Analysis results]. *Tsentr Ekolohichnykh Initsiatyv "Ekodiia"*, Kyiv (in Ukrainian).
- Hu, Y., Liu, X., Bai, J., Shih, K., Zeng, E. Y., & Cheng, H. (2013). Assessing heavy metal pollution in the surface soils of a region that had undergone three decades of intense industrialization and urbanization. *Environmental Science and Pollution Research*, 20, 6150–6159.
- Jergovic, M., Miskulin, M., Puntaric, D., Gmajnic, R., Milas, J., & Sipos, L. (2010). Cross-sectional biomonitoring of metals in adult populations in post-war Eastern Croatia: Differences between areas of moderate and heavy combat. *Croatian Medical Journal*, 51(5), 451–460.
- Jirau-Colon, H., Cosme, A., Marcial-Vega, V., & Jimenez-Vrelez, B. (2019). Toxic metals depuration profiles from a population adjacent to a military target range (Vieques) and Main Island Puerto Rico. *International Journal of Environmental Research and Public Health*, 17(1), 264.
- Kastrati, G., Paçarizi, M., Sopaj, F., Tašev, K., Stafilov, T., & Mustafa, M. K. (2021). Investigation of concentration and distribution of elements in three environmental compartments in the region of Mitrovica, Kosovo: Soil, honey and bee pollen. *International Journal of Environmental Research and Public Health*, 18(5), 2269.
- Kintl, A., Huňady, I., Vymyslický, T., Ondrisková, V., Hammerschmidt, T., Brtnický, M., & Elbl, J. (2021). Effect of seed coating and peg-induced drought on the germination capacity of five clover crops. *Plants*, 10(4), 724.
- Kirk, W. (2006). *A colour guide to the pollen loads of the honey bee*. 2nd ed. International Bee Research Association. Monmouth.
- Klym, O. Y. (2017). Vmist zhymykh kyslot i vazhkykh metaliv u pylku z kulbaby likarskoyi u riznykh pryrodno-ekolohichnykh zonakh Karpatskoho rehionu [Fatty acids and heavy metals in the dandelion pollen in different natural environmental areas of the Carpathian region]. *Naukovo-Tekhnichniy Biuletyn Derzhavnoho Naukovo-Doslidnoho Kontrol'noho Instytutu Veterynarnykh Preparativ ta Kormovykh Dobavok i Instytutu Biolohii Tvaryn*, 18(1), 55–65 (in Ukrainian).
- Kovalchuk, I. I., & Fedoruk, R. S. (2008). Medonosni bdzholy ta med – bioindykatory zabrudnennia navkolyshn'oho seredovyscha vazhkykh metalamy [Melliferous bees and honey are bioindicator contamination of environment by heavy metals]. *Biolohiia Tvaryn*, 10(1), 24–32 (in Ukrainian).
- Kovalchuk, I. I., & Fedoruk, R. S. (2013). Vmist vazhkykh metaliv u tkanyakh bdzhil u yikh produktsii zalezno vid ahrekolohichnykh umov Karpats'koho rehionu [Content of heavy metals in the bees tissues and products depending on agroecological conditions of carpathians region]. *Biolohiia Tvaryn*, 15(4), 54–65 (in Ukrainian).
- Kozak, V. M., & Brygadyrenko, V. V. (2018). Impact of cadmium and lead on *Megaphyllum kievense* (Diplopoda, Julidae) in a laboratory experiment. *Biosystems Diversity*, 26(2), 128–131.
- Lavrinenko, Y. A., Vlaschuk, A. H., Drobit, A. S., & Vlaschuk, O. A. (2019). Nasinieva produktyvnist' sortiv burkunu biloho odnorichnoho na pivdni Ukrainy [Seed productivity of white one-year white clover varieties in the south of Ukraine]. *Naukovi Dopovidi NUBiP Ukrainy*, 2, 78 (in Ukrainian).
- Macukanovic-Jocic, M., & Jaric, S. (2016). The melliferous potential of apiflora of Southwestern Vojvodina (Serbia). *Archives of Biological Sciences*, 68(1), 81–91.
- Massol-Deyá, A., Pérez, D., Pérez, E., Berrios, M., & Díaz, E. (2005). Trace elements analysis in forage samples from a US Navy bombing range (Vieques, Puerto Rico). *International Journal of Environmental Research and Public Health*, 2(2), 263–266.
- Mazur, T. (1997). Konstantni metody matematychnoi obrobky kilkinsnykh pokaznykiv [Constant methods of mathematical processing of quantitative indicators]. *Veterinary Medicine of Ukraine*, 9, 35–37 (in Ukrainian).
- Munir, N., Jahangeer, M., Bouyahya, A., El Omani, N., Ghchime, R., Balahbib, A., Aboulghras, S., Mahmood, Z., Akram, M., & Shah, S. M. A. (2022). Heavy metal contamination of natural foods is a serious health issue: A review. *Sustainability*, 14(1), 161.
- Pereira, P., Bašić, F., Bogunovic, I., & Barcelo, D. (2022). Russian-Ukrainian war impacts the total environment. *Science of the Total Environment*, 837, 155865.
- Polishchuk, V. P., & Lokutova, O. A. (2002). Biolohichni osoblyvosti zhyvlennia bdzhil i zbyrannia kvitkovoho pylku v umovakh poliflornoho vziatku [Biological features of bee feeding and pollen collection under conditions of polyfloral bribery]. *Biolohiia Tvaryn*, 4(1), 236–242 (in Ukrainian).
- Rawtani, D., Gupta, G., Khatri, N., Rao, P. K., & Hussain, C. M. (2022). Environmental damages due to war in Ukraine: A perspective. *Science of the Total Environment*, 850, 157932.
- Razanov, S. F., Razzanova, A. M., Razzanov, O. S., & Kutsenko, M. I. (2023). Sposib vykorystannia gruntiv, porushenykh vnaslidok boiovykh dii pid chas reabilitatsii [The method of using soils disturbed as a result of hostilities during their rehabilitation]. *Utility model patent No. 154051*. Ukraine. *Industrial property. Bulletin* 40 (in Ukrainian).

- Razanov, S. F., Tkachuk, O. P., Mazur, V. A., & Didur, I. M. (2018). Effect of bean perennial plants growing on soil heavy metal concentrations. *Ukrainian Journal of Ecology*, 8(2), 294–300.
- Razanov, S. F., Tkachuk, O. P., Razanova, A. M., Bakhmat, M. I., & Bakhmat, O. M. (2020). Intensity of heavy metal accumulation in plants of *Silybum marianum* L. in conditions of field rotation. *Ukrainian Journal of Ecology*, 10(2), 131–136.
- Razanova, O., Kucheriavy, V., Tsaruk, L., Lotka, H., & Novgorodska, N. (2021). Productive flight activity of bees in the active period in the conditions of Vinnytsia region. *Journal of Animal Behaviour and Biometeorology*, 9(4), 21038.
- Rewiev, A., Erhan, S., & Matur, E. (2023). Honey can be used as an indicator of environmental pollution. *Ahranyi Visnyk Prychomomoria*, 108, 171–175.
- Robinson, B. H., Bischofberger, S., Stoll, A., Schroer, D., Furrer, G., Roulier, S., Gruenwald, A., Attinger, W., & Schulin, R. (2008). Plant uptake of trace elements on a Swiss military shooting range: Uptake pathways and land management implications. *Environmental Pollution*, 153(3), 668–676.
- Rivis, Y. F., Postoienko, V. O., Stasiv, O. F., Saranchuk, I. I., Klym, O. Y., Diachenko, O. B., Stadnytska, O. I., Fedak, V. D., & Hopanenko, O. O. (2022). Koeffitsiienty perekhodu vazhkykh metaliv i vmist anionnykh zhymykh kyslot u bdzholynomu obnizhzhii (pylku roslin) u riznykh pryrodnykh zonakh Karpatskoho rehionu [Heavy metals transition coefficients and anionic fatty acid content in bee pollen (plant pollen) in different natural areas of the carpathian region]. *Perezhime ta Hirske Zemlerobstvo i Tvarynyystvo*, 72(2), 164–185 (in Ukrainian).
- Skalny, A. V., Aschner, M., Bobrovniksky, I. P., Chen, P., Tsatsakis, A., Paoliello, M. M. B., Djordjevic, A. B., & Tinkov, A. A. (2021). Environmental and health hazards of military metal pollution. *Environmental Research*, 201, 111568.
- Toth, G., Da Hermann, T., Silva, M. R., & Montanarella, L. (2016). Heavy metals in agricultural soils of the European Union with implications for food safety. *Environment International*, 88, 299–309.
- Verma, V., Singh, R., & Kumar, V. (2023). Bioaccumulation and analysis of heavy metals, minerals, and trace elements in the soil, pollen, nectar, and honey samples from Majha region, Punjab, India. *The Pharma Innovation Journal*, 12(12), 4034–4044.
- Zabala, J. M., Marinoni, L., Giavedoni, J. A., & Schrauf, G. E. (2018). Breeding strategies in *Melilotus albus* Desr., a salt-tolerant forage legume. *Euphytica*, 214(2), 22.
- Zajdel, B., Migdał, P., Murawska, A., Murawska, A., Jójczyk, A., Berbec, E., & Kucharska, K. J. (2023). Gąbka. Concentration of heavy metals in pollen and bees *Osmia bicornis* L. in three different habitats in the lowicz district in Central Poland. *Agriculture*, 13(12), 2209.
- Zhang, H., Bai, R., Wu, F., Guo, W., Yan, Z., Yan, Q., Zhang, Y., Ma, J., & Zhang, J. (2019). Genetic diversity, phylogenetic structure and development of core collections in *Melilotus* accessions from a Chinese gene bank. *Scientific Reports*, 9(1), 13017.