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Ecotoxicological assessment of soil contaminated by military operations using earthworms *Eisenia fetida*

S. V. Khyzhnyak, V. I. Korniyenko, O. P. Samkova, S. V. Midyk, O. M. Iakubchak

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

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National University of Life and Environmental Sciences of Ukraine, Heroyiv Oborony st., 15, Kyiv, 03041, Ukraine.
Tel.: +38-050-696-92-23.
E-mail: khs2014@ukr.net

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Areas of soil cover of agricultural lands in Mykolaiv region, degraded due to military operations and characterized by complex pollution, were examined for ecotoxicity. Soil biotesting, according to ISO standards, was conducted under laboratory conditions (14-day exposure) using earthworms *Eisenia fetida*. High toxicity of the experimental (degraded) soil with respect to *E. fetida* (100% mortality of organisms) was established. However, with a twofold reduction in the proportion of contaminated soil in the earthworms' habitat, this index was reduced by approximately half. It is shown that possible effects of soil contamination, which disrupt the vital functions of earthworms, are accompanied by a redistribution of fatty acids (FAs) content of their total lipids. FAs analysis was performed on a Trace GC Ultra gas chromatograph with a Flame Ionization Detector, which allowed the identification of 19 fatty acids in the worms' body. A decrease in the content of unsaturated FAs by 24.3%, in particular monounsaturated FAs, mainly due to oleic acid (C18:1ω9), was revealed as a result of the impact of soil contamination on earthworms. In addition, an increase in the content of saturated FAs by 55.7%, especially such important FAs as palmitic acid (C16:0) by 53.7% and stearic acid (C18:0) by 50.1%, was detected, which may lead to structural and functional changes in cell membranes. An important fact is the decrease in the content of polyene FAs of the ω6 family, in particular arachidonic acid (C20:4ω6) by 39.2% and the ω3 family, which are involved in the regulation of a wide range of physiological processes. The reorganization of the profile of lipid FAs of *E. fetida* can be considered as an early marker of metabolic changes in the earthworm body. The obtained results contribute to the objective assessment of the ecotoxicological consequences of soil damage as a result of military operations and contribute to an effective strategy for managing contaminated areas of agricultural land.

Keywords: military actions; combined pollution; biotesting; terrestrial ecotoxicology; soil biota; biomarkers; fatty acids.

Introduction

Sustainable land management is a prerequisite for balanced development of the agricultural sector. Environmental protection and the production of eco-friendly products are therefore one of the major challenges in agriculture. War zones and military facilities are considered one of the main sources of pollution of soil ecosystems (Shukla et al., 2023). The use of heavy equipment, explosions and detonation of ammunition lead to the formation of toxic products that enter the soil, which can have long-term consequences for agricultural land. Over 20 million hectares of land in Ukraine require renewal of the soil cover (Dmytrukha et al., 2024), which necessitates monitoring studies and implementation of soil remediation measures (Fayiga, 2019). Chemical contamination of soils as a result of military actions is characterized by complexity due to the entry of explosives, petroleum hydrocarbons, heavy metals and other toxic substances. Among the explosives, nitroaromatic compounds predominate (Hill et al., 2012; Zulfiharov et al., 2023), which are usually quite stable in the soil (Certini et al., 2013). Chemical compounds, in particular those that are not biodegradable, can disrupt the structure of soil biocenoses, cause the death of microorganisms and invertebrates, which leads to soil degradation and a decrease in their fertility (Sakun & Kutkovy, 2025). To detect the presence of toxic chemical pollutants and determine their content in environmental objects (soil, water, biota), the leading role belongs to physicochemical methods (Khyzhnyak et al., 2025a).

Biotesting provides much more reliable information on the effects of environmental pollution on biota than soil chemical analysis alone, as it is based on reactions of living organisms to versatile factors that may act synergistically or antagonistically (Paustenbach, 2015). Assessment of the combined effects of toxicants reflects their real impact on the environment, unlike the effects of individual substances (Schnug et al., 2014). Ecotoxicological testing using soil-dwelling test organisms provides a comprehensive assessment of soil toxicity and environmental consequences (Wieczorek & Baran, 2022). It has been shown that contamination of the soil cover with heavy metals after military operations leads to their accumulation by plants, invertebrates and vertebrates (Skalny et al., 2021). Therefore, an objective and complete assessment of the ecological state of the soil cover is possible if physico-chemical methods and biotesting are combined in the comprehensive assessment of the ecological risk (Casado-Martinez et al., 2016).

Thus, research on the identification soil cover pollutants, assessment of its ecological state as a result of military operations, is important from a scientific and practical point of view, which makes it possible to determine the safety of soil use in agriculture. Earthworms are typical invertebrate organisms found in soil ecosystems and play a crucial role in soil-forming processes. For them, soil is the environment for existence, growth and development. Earthworms as bioindicators of soil pollution are considered key organisms for assessing environmental risk (Song et al., 2009).

Ecotoxicological studies using earthworms is becoming increasingly popular. Traditionally, typical tests on earthworms have included whole-organism measurements such as survival, growth, reproduction, and behavioral changes. However, the use of molecular biomarkers is also proposed, such as detection of DNA damage, changes in enzyme activities and gene expression, etc. (Uwizeyimana et al., 2017). In ecotoxicological studies, earthworms, in particular the species *Eisenia fetida* (Savigny, 1826), are considered the best biological indicators of soil pollution due to their sensitivity to soil pollutants (Fourie et al., 2007).

The aim of the study was to conduct ecological and toxicological assessment of soil pollution caused by military operations of agricultural lands using earthworms *Eisenia fetida*.

Materials and methods

The paper examines soil samples (experimental material) from an agricultural plot with damage to the soil cover from artillery and mortar shells, which were taken in 2025 (Snihurivka Territorial Community, Mykolaiv region) (Fig. 1).

Soil sampling was carried out in accordance with: ISO 10381-1:2002 Soil quality. – Part 1: Guidance on the design of sampling programmes; ISO 10381-2:2002 Soil quality. – Part 2: Guidance on sampling methods; ISO 10381-4:2002 Soil quality. – Part 4: Guidance on the procedure for investigatic natural, near-natural and cultivated sites. Soil material was collected from 5 points at a depth of 0–20 cm and combined. Background (uncontaminated) soil samples were also similarly collected in this area. Combined soil samples were cleared of any foreign impurities, crushed, and sieved through a sieve with pores with a diameter of 2 mm. The average weight of background (uncontaminated) and contaminated soil samples was 4 kg each. Agrochemical analysis indicates that the background soil can be classified as heavy loamy southern chernozem, and after hostilities degradation and depletion of the humus of the soil cover occurs (Khyzhnyak et al., 2025b).

Fig. 1. View of the area with soil cover after military actions (Snihurivka Territorial Community, Mykolaiv region, June 2025)

Ecotoxicological studies were performed in acute toxicity tests for earthworms *Eisenia fetida* according to the OECD recommended method (ISO 11268-1:1993 Soil quality – Effects of pollutants on earthworms (*Eisenia fetida*) – Part 1: Determination of acute toxicity using artificial soil substrate). The toxicity of uncontaminated (background) soil and contaminated soil was determined. Additional control was artificial soil of the following composition: peat – 10%, clay – 20%, quartz sand –70% of the content per dry mass, recommended by ISO 11268-1: 1993.

The following studied soil groups were formed: control (artificial) soil, control (background) soil, contaminated soil, contaminated + artificial soil (ratio 50:50), contaminated + background soil (ratio 50:50). The containers were pre-filled with soil in an amount of 500 g by dry weight, which was thoroughly mixed with water to create humidity at a level close to 55% of its total water holding capacity (full moisture capacity), the pH of the soil in the containers was from 5.9 to 6.2.

The biological object was represented by adult sexually mature individuals of the earthworms *Eisenia fetida* (Savigny, 1826), aged from 2 months, weighing from 300 to 500 mg. Test organisms, washed with distilled water, dried on filter paper and weighed in batches, were placed in glass containers (2 L volume). For each studied soil group, 4 containers were used, each containing 10 worms, which had approximately the same weight. At the end of the experiment, the earthworms were removed from the soil, washed, dried, weighed and frozen for further research. Electronic laboratory scales PLS-510-3A with a resolution of 0.001 g (Kern & Sohn GmbH, Balingen, Germany, 2019) were used for weighing the worms.

At the end of the study, mortality, biomass and behavioral activity of the test organisms were determined. The change in earthworm body weight was calculated from the average body weight at the beginning and end of the experiment. The exposure period of the earthworms in the soil was 14 days. The number of repetitions for each soil group was 4 times.

The validity of the results was evaluated according to the criterion of the average loss of biomass in the control (artificial) soil, which should not exceed 20.0%, and the mortality of individuals, which should not exceed 10.0%. The following conditions were maintained during the experimental period: temperature 20 ± 2 °C, light intensity in the range of 400–800 Lux, illumination period 16 h:8 h.

To determine the content of the fatty acids (FAs) of total earthworm lipids, the homogenization and subsequent extraction of lipids with chloroform-methanol mixture was performed. Samples of earthworm tissues were taken from 4 containers of control (artificial) soil and 4 containers of contaminated + artificial soil (ratio 50:50), which were analyzed separately ($n = 4$). A mixture of chloroform and methanol (2:1) (v/v) in a volume of 20 mL and 1 mL of distilled water was added to a 1 g tissue sample, centrifuged at 3000 rpm for 10 minutes for clear phase separation. A Thermo Scientific refrigerated laboratory centrifuge (Thermo Fisher Scientific, Osterode am Harz, Germany, 2011) was used. Hydrolysis and methylation of lipid FAs was conducted, followed by gas chromatographic analysis according to (ISO 12966-2:2017. Animal and vegetable fats and oils – Gas chromatography of fatty acid methyl esters – Part 2: Preparation of methyl esters of fatty acids).

Gas chromatographic analysis of FAs methyl esters was performed on a Trace GC Ultra gas chromatograph (Thermo Fisher Scientific, Waltham, Massachusetts, USA, 2011) with Flame Ionization Detector (FID) using a Supelco SP-2560 capillary column (100 m long), Chromatography conditions: column temperature 140–240 ºC, detector temperature 260 ºC, analysis duration 65 min. FAs were identified using a Supelco 37 Component FAME Mix standard (Sigma-Aldrich, St. Louis, USA, 2024), which contains 37 fatty acids at a concentration of 10 mg/mL in dichloromethane. To quantify individual FAs the method of internal normalization was used and the relative content of FAs was presented as a percentage of their total amount. Moreover, the total quantities of saturated fatty acids (SFAs), unsaturated fatty acids (UFAs), monounsaturated fatty acids (MUFAs), polyunsaturated fatty acids (PUFAs), FAs of the ω3 and ω6 families were calculated.

Data are presented as mean ± standard deviation ($x \pm SD$). Mortality data were analyzed using Fisher's exact test ($P < 0.001$). Data of body weight changes were processed using one-factor analysis of variance (ANOVA), followed by post hoc Tukey testing. For parallel comparisons of the FAs specific indexes in the two groups: control and experimental, an independent two-sample t-test was used, but with Bonferroni correction for P-values separately for individual FAs (19 tests) and their subclasses (7 tests).

Results

According to the obtained results (Table 1), no mortality of earthworms was observed during the exposure period (14 days) in the control (artificial) and control (background) soils. The assessment of worm mortality rates indicates a high statistical significance of the differences between the studied groups (Fisher's exact test, $P < 0.001$). In the control, 100% survival was observed ($n = 40$). In cthe ontaminated soil, 100% death of worms is observed, mainly in the first 2–3 days of exposure. At the end of the study, the dead worms were at various stages of mummification (Fig. 2). When worms were exposed to a mixture of soils contaminated + artificial (ratio 50:50), the mortality of individuals was 47.5% (n = 21 dead out of 40), and in a mixture of soils contaminated + background (ratio 50:50) – 50.0% (n = 20 dead out of 40). This indicates a clear negative impact of contaminated soil on the viability of organisms.

The body weight of earthworms at the beginning and end of the study (14 days) did not significantly change in the research groups. There were also no significant weight changes between groups at the end of the experiment. In addition, the calculated worm body weight loss did not significantly differ between groups. An exception is the study group with contaminated soil, on which 100% death of worms is observed. Table 1 presents the average body weight of earthworms in groups and the average body weight loss. At the same time, it is worth noting the total change in biomass in the groups with contaminated + artificial soil (ratio 50:50) and contaminated + background soil (ratio 50:50) is 53.4±8.3% ($P < 0.01$) and 54.7±9.1% ($P < 0.01$), respectively, due to earthworm mortality.

Behavioral reactions of test organisms that remained alive after 14 days of exposure in experimental soils: contaminated + artificial and contaminated + background, differed from control ones. They reacted sluggishly to both light and mechanical irritation, moved very slowly and, sometimes, convulsions of individual specimens were observed.

a *b*

Fig. 2. Assessment of soil toxicity for earthworms *Eisenia fetida* (14-day exposure): *a* – control (artificial) soil; *b* – contaminated soil

Table 1
Assessment of the ecotoxicity of soil in agricultural lands damaged by military operations using earthworms *Eisenia fetida* (x ± SD, n = 40)

Indicators, units	Control (artificial) soil	Control (background) soil	Contaminated soil	Contaminated + artificial soil (ratio 50:50)	Contaminated + background soil (ratio 50:50)
Total soil weight, g (dry mass)	500	500	500	250 + 250	250 + 250
[1]Mortality after 14 days, %	0.0 ± 0.0	0.0 ± 0.0	100 ± 0.0	47.5 ± 5.0$^{\#}$	50.0 ± 8.2$^{\#}$
[2]Mean body weights per earthworm, mg:	528 ± 40	487 ± 23	475 ± 11	499 ± 29	488 ± 23
beginning of experiment after 14 days	506 ± 24$^{n.s.}$	453 ± 27$^{n.s.}$	0.0 ± 0.0	445 ± 67$^{n.s.}$	430 ± 56$^{n.s.}$
[3]Body weight changes of earthworms, %	4.2 ± 3.0	7.0 ± 5.8	0.0 ± 0.0	10.8 ± 6.3$^{n.s.}$	11.9 ± 7.1$^{n.s.}$

Note: [1] Fisher's exact test ($^{\#}$ $P < 0.001$), statistically significant differences compared with the corresponding value in the control; [2] mean values do not differ significantly between the beginning and end of the experiment in the groups as determined by ANOVA, $^{n.s.}$ – not significantly different in groups between beginning and end of experiment and between groups at the end of the experiment; [3] the mean values are not significantly different between groups as determined by ANOVA, $^{n.s.}$ – not significantly different vs control.

Thus, contaminated soil, which is characterized as degraded (Fig. 1), shows high toxicity against soil earthworms *E. fetida*, which indicates high ecotoxicity of the soil cover due to military operations. When the proportion of contaminated soil in the habitat of worms is reduced, namely in a mixture of contaminated soil with artificial or background soil (at 50:50 ratio), its toxicity in terms of mortality is reduced by approximately half.

An important indicator that can reflect metabolic shifts in animal tissues in toxic exposures is the content of lipid fatty acids, which is dynamically related to the functional state during physiological and biochemical processes in the body. The results of the FAs profile of *E. fetida* total lipids after 14-day exposure in control (artificial) soil and contaminated + artificial soil (ratio 50:50) are shown in Table 2. By high-sensitivity gas chromatography, 19 fatty acids of earthworm lipids were identified and quantified in the control and experimental groups.

SFAs are mainly represented by palmitic (C16:0) and stearic (C18:0) acids. The UFAs pool is represented by MUFAs, in particular oleic (C18:1ω9) acid and by PUFAs, in particular linoleic (C18:2ω6), linolenic (C18:3ω3) and arachidonic (C20:4ω6) acids (Table 2). The results presented in Table 2 indicate an increase in total SFAs content (by 55.7%) and a decrease in UFAs (by 24.3%) of the earthworms in the contaminated + artificial soil compared to the control (artificial) soil. There was an increase in the content of such FAs as palmitic (C16:0) by 53.7% and stearic (C18:0) by 50.1%, which are able to raise rigidity and reduce the mobility of cell membranes, thereby affecting their structural and dynamic characteristics and functional activity. An increase in the total saturation index (SFAs/UFAs) may reflect lower elasticity of cell membranes.

Under these experimental conditions, the total content of MUFAs decreased by 24.9% compared to the control group. The most important is oleic (C18:1ω9) acid, the content of which decreased by 18.7%. The total content of PUFA decreased by 24.0 % versus the control group. Among long-chain FAs, arachidonic (C20:4ω6) acid prevails, the content of which decreased by 39.2 % in comparison with the control group.

It is important that in earthworms the content of ω6 FAs prevails over ω3, since it is known that cell membranes enriched with ω6 FAs are more resistant to exogenous factors. Among PUFAs, the ω3 and ω6 families are precursors of biologically active substances synthesized in many animal tissues in response to exogenous influences (Simopolos & Cleland 2003). As follows from Table 2, under contaminated soil conditions, the total content of ω3 and ω6 FAs in *E. fetida* decreased by 22.3% and 25.0%, respectively. The results indicate (Table 2) that the metabolism of lipid FAs in earthworms is sensitive to soil contamination due to military operations. Thus, using earthworms *E. fetida*, ecotoxicity was established for the biota of the soil cover damaged as a result of military operations.

Table 2
Fatty acid content (%) in total lipids of earthworms *Eisenia fetida* after 14 days of exposure in the control (artificial) soil and contaminated+artificial soil (ratio 50:50, x ± SD, n = 4)

Fatty acids	Control (artificial) soil	Contaminated + artificial soil (ratio 50:50)	Compared to the control, %
Myristic acid, C14:0	1.71 ± 0.04	3.16 ± 0.04 #	184.8
Myristoleic acid, C14:1	0.74 ± 0.02	0.61 ± 0.03 #	82.4
Pentadecanoic acid, C15:0	1.20 ± 0.03	1.64 ± 0.04 #	136.7
Palmitic acid, C16:0	15.0 ± 0.28	23.06 ± 0.32 #	153.7
Palmitoleic acid, C16:1	4.60 ± 0.19	1.78 ± 0.08 #	38.7
Heptadecanoic acid, C17:0	0.76 ± 0.03	1.96 ± 0.06 #	257.9
Stearic acid, C18:0	10.26 ± 0.21	15.40 ± 0.33 #	150.1
Oleic acid, C18:1ω9	22.84 ± 1.02	18.57 ± 0.48 #	81.3
Linoleic acid, C18:2ω6	14.52 ± 0.90	12.21 ± 0.98 n.s.	84.1
Arachidic acid, C20:0	1.15 ± 0.09	1.72 ± 0.08 #	149.6
Gondoic acid, C20:1ω9	1.56 ± 0.12	1.38 ± 0.09 n.s.	88.5
γ-Linolenic acid, C18:3ω6	1.02 ± 0.05	0.68 ± 0.03 #	66.7
α-Linolenic acid, C18:3ω3	5.79 ± 0.15	4.48 ± 0.10 #	77.4
Eicosadienoic acid, C20:2ω6	0.97 ± 0.04	0.71 ± 0.03 #	73.2
Behenic acid, C22:0	0.35 ± 0.01	0.43 ± 0.02 #	122.9
Arachidonic acid, C20:4ω6	8.49 ± 0.45	5.16 ± 0.31 #	60.8
Eicosapentaenoic acid, C20:5ω3	6.50 ± 0.24	5.23 ± 0.21 #	80.5
Docosahexaenoic acid, C22:6ω3	2.54 ± 0.09	1.82 ± 0.08 #	71.7
Σ SFA	30.43 ± 0.32	47.37 ± 0.33 #	155.7
Σ UFAs	69.57 ± 0.48	52.63 ± 0.34 #	75.7
Σ MUFA	29.74 ± 0.13	22.34 ± 0.12 #	75.1
Σ PUFAs	39.83 ± 0.18	30.29 ± 0.15 #	76.0
Σ ω3	14.83 ± 0.09	11.53 ± 0.08 #	77.7
Σ ω6	25.00 ± 0.12	18.76 ± 0.16 #	75.0
SFAs / UFAs	0.44 ± 0.02	0.81 ± 0.05 #	184.1

Note: data are presented as mass fraction of fatty acid in % of total fatty acids. SFAs – saturated fatty acids, UFAs – unsaturated fatty acids, MUFAs – monounsaturated fatty acids, PUFAs – polyunsaturated fatty acids. # P<0.001 – mean value differ significantly vs control; n.s.– not significantly different vs control.

Discussion

Studying the impact of military operations on soils is important for assessing the extent of environmental pollution and developing effective methods for restoring agricultural land. Major threats include physical destruction of land cover, chemical contamination by heavy metals and toxic substances, which can have long-lasting consequences for soil biota (Hanyan et al., 2020). Changing the chemical composition of the soil leads to a decrease in fertility and complicates its use in agriculture (Chaika & Korotkova, 2023).

Our previous studies indicate combined soil contamination, which was characterized as degraded in the studied area of agricultural land in Mykolaiv region (Khyzhnyak et al., 2025b). An increase in the total content of such dangerous chemical elements as Zinc (Zn), Cadmium (Cd), Arsenum (As) was established. In particular, a 2–3 fold excess of Maximum Allowable Concentration (MAC) for the soil was found for Arsenum and Sulfur. In addition, an increase of nitroaromatic compounds was also established in the soil. In particular, the content of 4-amino-2,6-dinitrotoluene was 0.242 ± 0.012 mg/kg, which is also exceeds the MAC for this compound in the soil (Maximum Allowable Concentrations of hazardous substances in accordance with the Standards for Maximum Allowable Concentrations of Hazardous Substances in Soils, as well as the list of such substances approved by Resolution of the Cabinet Ministers of Ukraine dated December 15, 2021 No. 1325). At the same time, contamination with other toxic substances cannot be ruled out, which requires study.

This requires the use of biotesting methods to assess the ecotoxicity of the soil of agricultural land as a result of military operations. Earthworms are reliable indicators for ecotoxicological assessment of soil contamination due to their ecological significance, high biomass in the soil, and sensitivity to relatively low concentrations of toxic substances in the environment (Singh & Fatima, 2022). They can accumulate a wide range of organic as well as inorganic pollutants from the soil (Morrison et al., 2000).

Biotesting, according to standardized method (ISO 11268-1:1993), was carried out in laboratory conditions using *E. fetida*. The obtained results are valid, since in the control (artificial) soil mortality was less than 10% (namely absent) and body weight loss was less than 20%, namely 4.2% (Table 1). In the test for acute toxicity (14-day exposure), according to the mortality rate, the high toxicity of the soil contaminated as a result of military operations of the studied area (Mykolaiv region) was established for *E. fetida* (100% death). It is known that mixtures of toxic elements (substances) lead to more unpredictable and more active toxic reactions compared to their individual effects (Pueyo et al., 2004), which can occur in soil contaminated by military operations. However, the reduced proportion of contaminated soil in the environment of earthworms (contaminated / artificial in a ratio of 50:50) resulted in a sublethal effect, its toxicity decreased two times. In this case, one of the ecotoxicological approaches in assessing the bioavailability of pollutants and sublethal effects is the use of molecular and biochemical indicators.

To identify possible pathways of toxicity of contaminated soil on the body of earthworms, in addition to the dermal effect, absorption processes and the effect on metabolism were taken into account (Ye et al. 2016). In this context, the study of the biochemical response of *E. fetida* to contaminated soil is based on the determination of the lipid profile of FAs as a labile indicator of metabolic shifts under toxic actions.

It is known that FAs, as important structural and energy components of cells, play a significant role not only in metabolic processes, but also mediate the body's defense response under the influence of exogenous factors (Wang et al., 2012). The work compared the FAs content of total lipids of earthworms that were in contaminated + artificial soil compared to control (artificial) soil.

Changes in the FAs content in the body are shown: an increase in the content of saturated and a decrease of unsaturated FAs, which leads to an increase in the saturation level. The detected composition of FAs of *E. fetida* is consistent with our previously obtained data (Khyzhnyak et al., 2020), as well as the data of other researchers (Gunya et al., 2016).

This characterizes an increase in rigidity and a decrease in the mobility of cell membranes, which affects their functional activity. Along with this, a decrease in the content of unsaturated FAs is likely associated with the activation of oxidative processes. It is known that MUFAs (the content of which decreases under experimental conditions) are characterized by antioxidant properties due to their ability to neutralize active forms of oxygen (Perona et al., 2005). At the same time, a decrease in the content of polyene FAs, in particular the ω6 and ω3 families, which are involved in the regulation of a wide range of physiological processes, was detected. Regarding PUFAs of the ω3 family, a decrease in their total content including the content of long-chain biologically active acids (C20:5ω3 and C22:6ω3), which are characterized by the lipid bilayer stabilizing functions, has been observed (Simopolos & Cleland, 2003).

The obtained results indicate a reorganization of the FAs profile of the total lipids of earthworms when the vital functions of *E. fetida* are disrupted (sublethal effect) due to exposure to contaminated soil. In turn, the modulation of the FAs profile of soil invertebrates is a sensitive indicator of changes in tissue metabolism, which is also manifested under the influence of toxic agents (Khyzhnyak et al., 2020).

Conclusion

Ecotoxicity of soil resulting from combined contamination following military actions using *E. fetida* has been established. The possibility of assessing the ecotoxicity of soil depending on the level of its contamination is shown. The complex effect of pollutants on the earthworms under sublethal exposure is associated with metabolic disorders, which is manifested in a change in the fatty acid profile of total lipids. The combination of physicochemical and ecological-toxicological soil studies contributes to a more accurate assessment of environmental risks and will allow reliable predictions of the environmental consequences of soil contamination. This indicates the importance of integrating ecotoxicological studies in monitoring agricultural lands that have been exposed to military influence.

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