Influence of herbicides, insecticides and fungicides on food consumption and body weight of Rossiulus kessleri (Diplopoda, Julidae)

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Introduction

Contemporary agricultural production is impossible without pesticides. Monocultures (fields, gardens, forest plantations) are treated with various types of pesticides, each affecting living organisms in different ways (Reyera, 2004; Böhmke et al., 2016). Pesticides negatively affect living organisms of non-target taxonomic groups, often harming animals that are not harmful for plants cultivated by people (Ebring et al., 2005; Adamski et al., 2007). The general population and farmers in the territory of Ukraine most often use such insecticides, fungicides and herbicides as Roundup, Unahan Forte, Omite, Ridomil Gold, BI 58, Biotlin, Tilt, Horus, and fungicides (Ridomil Gold, Thiovit Jet, Penncozeb 80 WP, Falcon, Tilt, Horus) which are often used in agrocenoses of Ukraine. During a 20-day experiment we used herbicides (Roundup, Unahan Forte), insecticides (Omite, BI 58, Biotlin, Actellic, Nurelle D) and fungicides (Ridomil Gold, Tilt, Horus) which are used in agrocenoses of Ukraine. Under the impact of Roundup and Unahan, body weight of R. kessleri reliably did not change, but food consumption and production of excrement increased. Pesticide-treated litter did not digest in the intestine of millipedes, but they survived to the end of the experiment (20 days). In the conditions of treating litter with insecticides Omite, BI 58, Biotlin, Actellic and Nurelle D, the amount of consumed food and body weight reliably did not change; most of these insecticides slowed the formation of feaces in the millipedes. The highest studied concentrations of Actellic and Nurelle D preparations caused death to R. kessleri. Depending on the concentration in the litter, the studied fungicides Ridomil Gold, Tilt, Horus, and fungicides, had a varying effect on food consumption, body weight and the amount of excrement of R. kessleri. Thus, in agrocenoses and forest ecosystems adjacent to them (windbreaks, ravines and flood plain forests), R. kessleri can be significantly affected by the manufacturer-recommended doses of pesticides, as well as more than ten-fold lower doses.

Keywords: Myriapoda; glyphosate; Propargite; Mefenoxam; Mancozeb; Dimethoate; Imidacloprid; Pirimiphos-methyl; Chlorpyrifos; Cypermethrin; Tebuconazole; Triadimenol; Spiroxamine; Propiconazole; Cyprodinil.

Pesticides kill organisms harmful for the human organism, sometimes also harming beneficial ones. After treatment, pesticides remain on the soil surface in agrocenoses and adjacent plots for decades. For the laboratory experiment, we selected Rossiulus kessleri (Lochmander, 1927) – a species which lives 5–6 years on the soil surface and can dig in to soil to a depth of 30–40 cm. During a 20-day experiment we used herbicides (Roundup, Unahan Forte), insecticides (Omite, BI 58, Biotlin, Actellic, Nurelle D) and fungicides (Ridomil Gold, Tilt, Horus) which are often used in agrocenoses of Ukraine. Under the impact of Roundup and Unahan, body weight of R. kessleri reliably did not change, but food consumption and production of excrement increased. Pesticide-treated litter did not digest in the intestine of millipedes, but they survived to the end of the experiment (20 days). In the conditions of treating litter with insecticides Omite, BI 58, Biotlin, Actellic and Nurelle D, the amount of consumed food and body weight reliably did not change; most of these insecticides slowed the formation of feaces in the millipedes. The highest studied concentrations of Actellic and Nurelle D preparations caused death to R. kessleri. Depending on the concentration in the litter, the studied fungicides Ridomil Gold, Tilt, Horus, and fungicides, had a varying effect on food consumption, body weight and the amount of excrement of R. kessleri. Thus, in agrocenoses and forest ecosystems adjacent to them (windbreaks, ravines and flood plain forests), R. kessleri can be significantly affected by the manufacturer-recommended doses of pesticides, as well as more than ten-fold lower doses.

BI 58 is an insecticide with active substance dimethoate. In acidic environments, there forms O-dimethoate with heightened toxicity for warm-blooded animals and insects. However, this compound is unstable and quickly decomposes to phosphorous acid. Biotlin is an insecticide which is often used against aphids. Its main active agent is imidacloprid, which belongs to the class of neonicotinoids. The preparation blocks the functioning of the postsynaptic acetylcholine receptor of the central nervous system of arthropods, leading to paralysis and death. Tilt, Horus had a fungicide against downy mildew, etc. Its active agent is sulfur. After spraying, it remains on the surface of plants and foliage for a long time.

Penccozeb 80 WP is a contact fungicide against diseases of vegetables, fruit-bearing plants and vineyards. Its active agent is Mancozeb. Manganese and zinc in the active substance provide simultaneous effect against the diseases and extra-root nutrition with the mentioned microelements, activating photosynthesis, therefore allowing higher yield to be obtained. Actellic is an insecticide of the group of phosphor-organic compounds. It is an intestinal contact fungicide with killing power against many insect pests of fruit-bearing plants and plant-feeding mites, including some spider mites. Nurelle D is an insecticide the main active substances of which are Cypermethrin and Chlorpyrifos. The preparation is considered low-toxic for birds, bees and aquatic organisms.

Falcon is a fungicide the active agents of which are tebuconazole, triadimenol, and spiroxamine. All the agents included in the content of fungicide Falcon (two substances of the azole group – triadimenol and tebuconazole, and a representative of class of spirotetralazines – spiroxamine) contribute to biosynthesis of sterols without participation of methyl at different levels, impair selectivity of cellular walls of the fungus regarding allowing access of various compounds to the cytoplasm, ultimately leading to death of pathogenic microorganisms. Tilt is a systemic
fungicide for protection of cereal crops against a complex of leaf and ear diseases, and also protection of berry plants and fodder herbs. The main active agent is propiconazole. Horus is a systemic fungicide for protection of fruit-bearing plants against a complex of diseases, particularly protection of grapevines against fungal pathogens. The main active agent of the preparation is cyprodinil. Cyprodinil impairs biosynthesis of methionine in fungal cells.

To determine how strong the effects of these substances on invertebrates are, it is practical to test the commonest of them. Millipedes of the Julidae family are phytosaprophages which consume mostly foliage of grasses, shrubs and trees (Bzyzov, 2006). The Julidae fauna of Ukraine and the European part of the former USSR is studied unevenly (Striganova & Prisnyi, 1990; Prisnyi, 2002; Golovatch, 2008; Golovatch & Karne, 2009; Evsyukov & Golovatch, 2013; Kokoška & Golovatch, 2020).

Julidae play an important role (Gere, 1956; Brygadyrenko, 2015, 2016) in functioning of both agroecosystems and adjacent natural ecosystems (forests, steppes, meadow areas). The effect of chemical pollutants on the organisms of Julidae is not studied sufficiently (Brygadyrenko & Ivanyushyn, 2015; Douglas et al., 2017, 2019; Kozak & Brygadyrenko, 2018; De Souza et al., 2019; Tóth & Hornung, 2019), while the effect of agrochemicals is practically unstudied. The most abundant diploid in Central and Southern Ukraine is Rossiades kessleri (Lochman, 1927). The effect of agrochemicals on this species still remains unstudied (Syvyrychkeno & Brygadyrenko, 2014).

The objective of the article was to evaluate the effects of the most common pesticides in Ukraine on the food consumption, changes in body weight, formation of feces and mortality of R. kessleri.

Materials and methods

The experiment started with collection of the material for the study – R. kessleri millipedes. They were collected 10 km away from the city, near Dnipropetrovsk Airport. Specimens of R. kessleri were collected manually during several days in late June (over 2,000 individuals of R. kessleri were collected). The millipedes were captured manually in the litter of a ravine forest area. Litter, the trophic resource of millipedes, was collected manually using special sieves which helped remove twigs and garbage from the leaves. We used sieves with the cells of 12.0 and 3.5 mm. The collected plant material was transferred to the laboratory of the Department of Zoology and Ecology of Dnipropetrovsk National University and was dried for two weeks. Dry litter was equally (2 g in each) distributed in plastic cups of 200 mL capacity. The number of cups equaled the number of millipedes. The millipedes were weighed with the accuracy of up to 0.1 mg, their sex was determined, and they were divided to cups. Pesticides (Table 1) were dissolved in distilled water. Using a pipette with a solution of pesticides (3.5 mL) or distilled water (in the control), the litter was moistened. Each variant of the experiment was performed in 10-fold replication (for each experiment, we used 5 males and 5 females with average body weight, the largest and smallest individuals according to the body weight were not taken into the experiment). The experiment lasted for 20 days. To maintain optimum moisture during the experiment, 2 mL of distilled water was added to the substrates in the cups each three days. After the experiment, the millipedes were weighed again and the litter was dried, excrement was removed and weighed as well. All weighing procedures were performed with the accuracy of up to 0.1 mg.

The results were statistically analyzed, determining the mean value, median, standard deviation (SD), and the first and the third quartiles for each characteristic in all the variants of the experiment. The selections were compared using single-factor dispersion analysis (ANOVA) with Bonferroni correction, considering differences between the selections reliable at the level of $P < 0.05$. All calculations and developments of diagrams were made in Statistica 8.0 software (StatSoft, USA, 2012).

Results

Impact of herbicides Roundup and Unahan Forte on R. kessleri. Under the effect of the preparation, Roundup, R. kessleri reliably increased their food consumption (Fig. 1a) in three variants of the experiment with highest doses of the herbicide (2.5 • 10–1, 2.5 • 10–2 and 2.5 • 10–3 mg of isopropylamine salt of glyphosate/g of litter). This was an unexpected result, because the opposite seemed more likely, namely slowing of food consumption due to impact of the pesticide Roundup. For other variants of the experiment, the median of the amount of consumed food ranged within 9–15 mg/day. No reliable changes in the body weight of R. kessleri under the influence of Roundup were observed (Fig. 1b). The median weight gain of R. kessleri for all the variants of the experiment ranged at the level of 0.1–0.6 mg/24 h. The rates of formation of excrement (Fig. 1c) increased in both variants of the experiment with maximum concentrations of pesticide (2.8 • 10–2 and 2.8 • 10–3 mg of isopropylamine salt of glyphosate/g of litter). In the other variants of the experiment, the median of the tempi of formation of excrement ranged within 9–13 mg/24 h.

Table 1

| Pesticides            | Active substance                  | Recommended dose in agroecosystems, kg/ha | Doses of preparations in our survey, mg/g of litter |
|-----------------------|-----------------------------------|-------------------------------------------|---------------------------------------------------|
| Roundup               | Isopropylamine salt of glyphosate, 450 g/L | 0.5 - 6.4                                | 2.8•10–3 – 2.8•102                                 |
| Unahan Forte          | Potassium salt of glyphosate, 500 g/L | 1.5 - 4.0                                | 3•10–3 – 3•102                                    |
| Omite                 | Propargyl, 570 g/L                | 0.9–2.2                                  | 3.6•10–3 – 3.6•101                                |
| Ridoral Gold          | Mancozeb, 640 g/kg; Mefanoxam, 40 g/kg | 2.5                                       | 4.0•103 – 4•103                                   |
| BI 58 EC              | Dimethoxate, 380 g/L             | 0.5–2.8                                  | 2.4•103 – 2.4•104                                 |
| Biofin                 | Imidacloprid, 200 g/L            | 0.3–0.5                                  | 1.2•103 – 1.2•104                                 |
| Thiovit Jet           | Sulfur, 800 g/3                   | 2.6–8.0                                  | 4.8•103 – 4.8•104                                 |
| Porocozeb 80 WP       | Mancozeb, 800 g/kg              | 1.7–1.8                                  | 4.8•103 – 4.8•104                                 |
| Actellic              | Pirimphos-methyl, 500 g/L        | 0.4–0.8                                  | 3•103 – 3•104                                     |
| Nurelle D             | Chlorpyrifos, 500 g/L; Cypermethrin, 50 g/L | 0.5–1.0                                | 3•103 – 3•104                                     |
| Falcon                | Tebuzonoxol, 167 g/L; triadimenol, 43 g/L; spiroxamine, 250 g/L | 0.4–0.6                                 | 1.1•103 – 1.1•104                                |
| Tilt                  | Propiconazole, 250 g/L           | 2.5–5.0                                  | 1.5•103 – 1.5•104                                 |
| Horus                 | Cyprodinil, 250 g/L              | 0.45                                     | 4.5•103 – 4.5•104                                 |

Influence of insecticides Omite, BI 58, Biotin, Actellic, Nurelle D on R. kessleri. The preparation Omite reliably did not increase the tempi of food consumption by specimens of R. kessleri (Fig. 3a), though the differences between the mass of the litter at the beginning and the end of the experiment in the variants with and without millipedes declined with increase in the concentration of pesticide compared to the control, suggesting cessation of nutrition by most specimens of millipedes. In the tested range of the values, during increase in the concentration of Omite preparation, body weight of R. kessleri reliably did not change (Fig. 3b), and the amount of feces reliably decreased (Fig. 3c). At maximum concentration of this pesticide (3.6 • 10–1 mg of propargyl/e of litter) the median of the amount of excrement increased to 2 mg/24 h compared with 25 mg/24 h per one millipede in the variant of the experiment without application of Omite preparation (Fig. 3c).
Fig. 1. Change in the food mass (a), body weight of (b), production of feces (c) by individuals of *Rossiulus kessleri* (Lohm.) during 20-day experiment in the gradient of concentration of Roundup preparation: on the abscissa axis – concentration of active agent of Roundup (isopropylamine salt of glyphosate, mg/g of litter), K – control (without Roundup); on the ordinate axis – mean daily change in the body weight, food or feces (mg/24 h), respectively; n = 10

Fig. 2. Changes in food mass (a), body weight of (b), production of feces (c) by the specimens of *Rossiulus kessleri* (Lohm.) during 20-h experiment in the gradient of the concentration of Urahan Forte preparation: on abscissa axis – concentration of active agent of Urahan Forte (potassium salt of glyphosate, mg/g of litter), K – control (without Urahan Forte); on the ordinate axis – mean daily changes in the body weight, mass of food and feces (mg/24h), respectively; n = 10
Under the impact of BI-58 preparation, the rates of breakdown of litter increased: the median of this characteristic equaled 15 mg/24 h in the conditions of absence of millipedes, and in the concentration of Dimethoate equaling $2.4 \times 10^{-1}$ 2.4 $\times 10^{-1}$ mg/g of litter it increased practically by two times 26-28 mg/24 h (Fig. 4a). Specimens of R. kessleri reliably did not increase the rates of breakdown of litter when exposed to the preparation BI-58 (Fig. 4b). Increase in the concentration of Dimethoate from $2.4 \times 10^{-2}$ to $2.4 \times 10^{-1}$ mg/g of litter caused reliable decrease in the body weight of R. kessleri by more than 4 mg/24 h (Fig. 4b). Similar decrease for the tempi of formation of excrement was seen at increase in the concentration of dimethoate from $2.4 \times 10^{-2}$ to $2.4 \times 10^{-1}$ mg/g of litter (Fig. 4c). Under the influence of low concentrations of the preparation Biotlin, the reliable differences between the variants of the experiment with and without R. kessleri millipedes were observed at the concentration higher than $1.2 \times 10^{-2}$ mg/g of litter (Fig. 5a). At maximum tested concentrations ($1.2 \times 10^{-1}$ and $1.2 \times 10^{-2}$ mg/g of litter), body weight of R. kessleri reliably did not differ from the control variants of the experiment (Fig. 5b). Intensity of production of excrement by millipedes reliably decreased from 24 to $10^{-12}$ mg/24 h (median) at all three studied concentrations of Imidacloprid (Fig. 5c).
The preparation Actellic led to complete death of the millipedes in two variants of the experiment with maximum concentration of Pirimiphos-methyl (3 \times 10^{-2} and 3 \times 10^{-1} mg/g of litter). At the same time, food was not consumed by R. kessleri exposed to the Actellic concentration of 3 \times 10^{-2} mg/g of litter (Fig. 6a); under influence of this concentration, body weight (Fig. 6b) and tempi of formation of excrement decreased in millipedes unobservably (Fig. 6c).

Under the influence of the preparation Nurelle D, we observed 100% death of R. kessleri while using the concentration of 3 \times 10^{-1} mg/g of litter. Even at the concentration of 3 \times 10^{-3} mg of Chlorpyrifos/g of litter the millipedes continued to feed, and the differences between the mass of litter in the variants with and without millipedes were not reliable (Fig. 7a). In the control (without influence of the pesticide) the body weight of R. kessleri increased by 0.7 mg/24 h, whereas at the concentration of 3 \times 10^{-2} mg of Chlorpyrifos of litter their body weight decreased on average by 0.5 mg/24 h (Fig. 7b). Tempi of formation of feces during the influence of this concentration also reliably decreased from 24 to 7 mg/24 h (Fig. 7c).

Impact of fungicides Ridomil Gold, Thiovit Jet, Penncozeb 80 WP, Falcon, Tilt, Horus on R. kessleri. When exposed to Ridomil Gold in the concentration of 4 \times 10^{-3} mg/g, the individuals of R. kessleri continued to consume food in the same tempi as in the variant of the experiment without exposure to pesticides (Fig. 8a). Increase in the concentration of this pesticide to 4 \times 10^{-2} mg/g caused the millipedes to stop feeding. The body weight of the specimens reliably did not change (Fig. 8b), and the intensity of defecation reliably decreased (Fig. 8c) already at minimum of the tested concentrations of Ridomil Gold preparation. Thus, already in lowest tested concentration (4 \times 10^{-2} mg/g of litter), Ridomil Gold preparation had a negative impact on the metabolism of R. kessleri.

Introduction of the preparation Thiovite Jet to the litter, even in the highest concentrations, did not decrease food consumption by R. kessleri (Fig. 9a). Interestingly, the addition of sulfate to the litter reliably increased the body weight of millipedes: median of gain in body weight increased from 0.7 mg/24 h in the control to 1.5−1.7 mg/24 h for the sulfate in the concentration of 4.8 \times 10^{-2} − 4.8 \times 10^{-1} mg/g of litter (Fig. 9b). No reliable changes in the intensity of production of excrement were seen in the gradient of Thiovite Jet concentration (Fig. 9c). According to the results of the laboratory experiment, R. kessleri stopped consuming food affected already by average concentration of Penncozeb 80 WP pesticide – 4.8 \times 10^{-2} mg of Mancozeb/g of litter (Fig. 10a). Under the influence of Mancozeb, no reliable changes in body weight of millipedes were observed compared with the control (Fig. 10b), though the tempi of formation of excrement decreased by approximately two times already while using the concentration of 4.8 \times 10^{-2} mg/g of litter (Fig. 10c).

As with Falcon pesticide, reliable differences in the mass of litter between the variants of the experiment with and without R. kessleri remained during influence of the concentration of 1.1 \times 10^{-2} mg of Tebuconazole/g of litter (Fig. 11a); during the influence of highest concentration of Falcon, the food consumption of millipedes stopped. Body weight of R. kessleri in all the variants with this pesticide reliably did not differ from the control group of millipedes (Fig. 11b). Already the minimum tested concentration of this pesticide caused a reliable more than two-fold decrease in the tempi of formation of feces (Fig. 11c).

Exposure to Tilt preparation caused cessation of consumption of litter by millipedes already in minimum tested concentration of propiconazole – 1.5 \times 10^{-2} mg/g of litter (Fig. 12a). Body weight of R. kessleri reliably did not change under the effect of Tilt compared with the control (Fig. 12b), though we observed a tendency towards decrease in body weight while using lowest tested concentration of the pesticide: perhaps, millipedes exposed to this concentration continued to consume food. Tempi of formation of feces also reliably did not change at the increase in the concentration of Tilt (Fig. 12c).
The preparation Horus had no reliable effect on food consumption of *R. kessleri* even in the highest tested concentration – 4.5 • 10⁻¹ mg of cyprodinil/g of litter (Fig. 13a).

It is interesting that the lowest of the tested concentrations (4.5 • 10⁻³ mg of cyprodinil/g of litter) caused maximum increase in the body weight of the millipedes: from 0.7 to 1.9 mg/24 h (Fig. 13b). The tempo of formation of excrement reliably did not change at increase in the concentration of cyprodinil in the food of the millipedes (Fig. 13c). Finally, we should note (Table 2) that the food treated with the large amount of the pesticides we tested did not absorb in the intestine during the experiment, remaining there and therefore poisoning the millipedes.

Each of the preparations in the tested concentrations either inhibited food consumption and formation of excrement or underlay decrease in body weight of the tested animals.
Fig. 11. Changes in mass of food (a), body weight of (b), production of feces (c) by individuals of Rossiulus kessleri (Lohm.) over 20-day experiment in the gradient of concentration of the preparation Falcon: on abscissa axis – concentration of active substance of Falcon (mixture of Tebuconazole, 167 g/L, triadimenol, 43 g/L, and spiroxamine, 250 g/L, concentration as indicated according to Tebuconazole, mg/g of litter), K – control (without exposure to Falcon); on ordinate axis – mean daily changes in body weight, mass of food or feces (mg/24 h), respectively; n = 10

Fig. 12. Changes in mass of food (a), body weight of (b), production of feces (c) by individuals of Rossiulus kessleri (Lohm.) over 20-day experiment in the gradient of concentration of the preparation Tilt: on abscissa axis – concentration of active substance of Tilt (propiconazole, mg/g of litter), K – control (without exposure to Tilt); on ordinate axis – mean daily changes in body weight, mass of food or feces (mg/24 h), respectively; n = 10

Fig. 13. Changes in mass of food (a), body weight of (b), production of feces (c) by individuals of Rossiulus kessleri (Lohm.) over 20-day experiment in the gradient of concentration of the preparation Horus: on abscissa axis – concentration of active substance of Horus (cyprodinil, mg/g of litter), K – control (without exposure to Horus); on ordinate axis – mean daily changes in body weight, mass of food or feces (mg/24 h), respectively; n = 10

Discussion

Agricultural production in the current conditions is impossible without use of pesticides (Relyea, 2004; Pardon et al., 2019). Agroecosystems – the biocenoses created and managed by man, – may develop certain productivity only in the conditions of constant maintenance of the composition of plants and invertebrates. Only a small part of pesticides introduced into agroecosystems reaches their target (eradicates species of organisms which are harmful to humans), while the rest destroy non-target organisms, remaining in the areas adjacent to the agroecosystems. Negative consequences related to introduction of pesticides involve not only direct killing of living organisms, but first of all change in the soil-forming processes: disturbance of the balance between humification (ingress of partially dissolved organic substances into the higher layers of soil which increases its fertility) and mineralization (destruction of organic compounds to carbon dioxide, atmospheric nitrogen and soluble salts which ingress into the atmosphere and groundwater). Balance between mineralization and humification is maintained first of all by the bacteria and fungi, and by soil
animals to a lesser extent. Ingress of pesticides to the litter changes the proportion between bacteria, fungi and animals in the food chains of the detritus ecosystem. The abundance of separate species of living organisms in the soil and litter is closely associated with the interactions of these species (Brygadyrenko, 2015, 2016). Absence of one or presence of other bacteria and fungi can slow the digestion of food by the organisms of species (Brygadyrenko, 2015, 2016). Absence of one or presence of other pesticides accumulate in trophic chains, part of these excess amounts of pesticides spread beyond the borders of the treated territory (Jabin et al., 2004; Golten et al., 2006). However, the potential danger of many pesticides to soil biota is studied insufficiently (Hopkin, 1990).

Mass death of predatory and parasitic animals caused by chemicals which regulate the number of phytophages leads to growth of plant-feeding organisms which in the conditions of absence of natural enemies quickly increase their populations – the so-called “pesticide syndrome” develops. Sub-lethal doses of insecticides may cause changes in the behaviour of individuals, causing changes in the spatial structure of their populations (vertical distribution of individuals in litter and on the surface of soil). Changes in the composition of fauna beneficial to humans after the treatment with pesticides may be reflected in species composition of pests in agroecosystems. At the same time, old and well-known pests become replaced by new pests – species which are rare and economically insignificant at the beginning of the process (Pardon et al., 2019). Threat increases also due to the fact that some pesticides can remain in the soil for many years. Even the lowest concentrations of them could be dangerous for soil saprophages (Hopkin, 1990). Small concentrations of pesticides in many cases inhibit the immune system, often exerting allergic, carcinogenic and mutagenic actions towards invertebrates and humans.

Table 2
evaluation of effects of pesticides on food consumption, changes in body weight of and rates of formation of feces by *R. kessleri*

| Products | Concentration causing reliable change in body weight, mg/g | Concentration which impacts tempo of formation of feces, mg/g |
|----------|----------------------------------------------------------|----------------------------------------------------------|
| Roundup  | 2.8 × 10^-3, 2.8 × 10^-3, 1.2 × 10^-3 | 2.8 × 10^-3, 2.8 × 10^-3 |
| Unihan Forte | 3 × 10^-1, 3 × 10^-1, 3 × 10^-1 | 3 × 10^-1 |
| Ormite | unreliably | unreliably |
| Ridoral Gold | 4 × 10^-3 | 4 × 10^-3 |
| BI-58 EC | unreliably | 2.4 × 10^-3, 2.4 × 10^-3 |
| Biolan | 1.2 × 10^-1, 1.2 × 10^-2 | 1.2 × 10^-1, 1.2 × 10^-2 |
| Thiovit Jet | unreliably | unreliably |
| Penncozeb 80 WP | 4.8 × 10^-3 | 4.8 × 10^-3 |
| Actellic | 3 × 10^-3 | 3 × 10^-3 |
| Nurelle D | 3 × 10^-1 | 3 × 10^-1 |
| Falcon | 1.1 × 10^-1 | 1.1 × 10^-1 |
| Tilt | 1.5 × 10^-3 | 4.5 × 10^-3 |
| Horus | unreliably | unreliably |

Metabolic processes in insects are usually more intense compared with diploponds, whereas decrease in the body weight in the similar experiment is not seen often (Shulman et al., 2017; Martynov & Brygadyrenko, 2017, 2018a, 2018b). Usually death of insects is observed already after cessation of consumption of food with a certain toxic substance. Diploponds are able to fall into summer diapause in the conditions of dry climate or digest the consumed food very slowly in certain conditions (Kanelo, 1999; Kozak & Brygadyrenko, 2018). This allows one to determine a quite long phase of slowing in the consumption of food or phase of thinning of animals during laboratory experiments.

In our experiment we surveyed the effects of insecticides, fungicides and herbicides on *R. kessleri* – species of saprophage that usually develops quickly in the litter. Sub-lethal doses of pesticides may cause changes in the behaviour of individuals, causing changes in the spatial structure of their populations (vertical distribution of individuals in litter and on the surface of soil). Changes in the composition of fauna beneficial to humans after the treatment with pesticides may be reflected in species composition of pests in agroecosystems. At the same time, old and well-known pests become replaced by new pests – species which are rare and economically insignificant at the beginning of the process (Pardon et al., 2019). Threat increases also due to the fact that some pesticides can remain in the soil for many years. Even the lowest concentrations of them could be dangerous for soil saprophages (Hopkin, 1990). Small concentrations of pesticides in many cases inhibit the immune system, often exerting allergic, carcinogenic and mutagenic actions towards invertebrates and humans.

**Conclusion**

Individuals of *R. kessleri* exposed to herbicides Roundup and Unihan were observed to have no reliable change in their body weight, but the intensity of food consumption and formation of feces increased. Food processed with these herbicides was not absorbed in the intestines of millipedes, but in the period of 20 days they did not die.

In most cases, during the influence of Ormite, BI-58, Biolan, Actellic and Nurelle D insecticides on litter, changes in the amount of consumed food were unreliable. Under the influence of many of the insecticides we used, the amount of excrements of millipedes reliably decreased, and the body weight of the animals reliably did not increase. This indicates that the food treated with the insecticides in the tested concentrations was not absorbed in the intestine, remaining there and thus gradually poisoning the animals. Less often, the applied pesticides caused death of millipedes (Actellic and Nurelle D).

During the use of Ridomil Gold, Thiovit Jet, Penncozeb 80 WP, Falcon, Tilt and Horus, the amount of excrements reliably decreased or remained with no changes; the body weight of millipedes increased or did not change. The surveyed fungicides affect the animals differently depending on the concentration in the litter.

The determined patterns suggest that *R. kessleri* in agroecosystems and forest ecosystems bordering them (windbreaks, ravine or floodplain forests) can be significantly affected by the manufacture-recommended doses of pesticides and over ten-fold lower doses. The organism of *R. kessleri* is quite an inert system which during 20 days can limit the food consumption without significant effect on vitality. Therefore similar experiments should be carried out for a longer period of time.

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