

QUARANTINE SPECIES OF STEM NEMATODES ARE LIMITED IN UKRAINE

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Abstract. In the article, the authors analyzed and systematized the results of research obtained during the processing of domestic and foreign information sources regarding the prevalence, harmfulness, and bioecological features of stem nematode species of limited distribution in Ukraine, which are included in the A2 list by the State Production and Consumer Service. In Ukraine, there are two such species: potato stem nematode (*Ditylenchus destructor* Thorne) and stem nematode (*Ditylenchus dipsaci* Filipjev). In contrast to European countries, where the potato stem nematode is not of significant economic importance, in Ukraine its mass distribution in field conditions and the manifestation of the disease during the storage of agricultural products lead to significant crop losses. The stem nematode is considered one of the most harmful species of phytonematodes, especially in temperate climates. Without proper control, the stem nematode can lead to a complete loss of the crop (primarily onions, garlic, cereals, strawberries, decorative bulbous plants). *D. dipsaci* is locally distributed in temperate climates almost all over the world (Europe, North and South America, Africa, Asia, Australia and Oceania), but the species is unable to survive in tropical climates except in some mountainous areas.

Keywords: stem nematodes, plant quarantine, A2 list, prevalence, harmfulness, phytosanitary risk.

Potato stem nematode *Ditylenchus destructor* Thorne (KKB – DITYDE) belongs to the type Roundworms – Nematoda, order Tylenchida – Tylenchida, family Anguinidae (Anguinidae).

The main food plant of the nematode is the potato, but the species was sometimes detected on plants of the following genera: cocks (Iris) (on bulbs and rhizomes), carrots (Daucus), clover (Trifolium), peanuts (Arachis), as well as on seeded garlic. It is believed that *D. destructor* can parasitize 70 crops and weeds and about the same number of mushroom species.

In contrast to European countries, where the stem nematode has no significant economic importance, in Ukraine its massive spread in the field and the manifestation of the disease during the storage of agricultural products leads to significant crop losses.

In recent years, the potato stem nematode has been frequently detected in peanut plantations in South Africa. There is an assumption that this population may be a separate ecotype or pathotype. Until now, it has not been registered on local potato plantations.

As of 2022, the potato stem nematode is widespread in many European countries: Austria, Albania, Belgium, Belarus, Bulgaria, Great Britain, Greece, Estonia, Ireland, Italy, Latvia, Lithuania, Luxembourg, Moldova, the Netherlands, Germany, Norway, Poland, Russia, Romania, Slovakia, Hungary, Ukraine, Finland, France, the Czech Republic, Switzerland and Sweden; Asia: Azerbaijan, Iran, Kazakhstan, Kyrgyzstan, China, Korea (South), Pakistan, Saudi Arabia, Tajikistan, Turkey, Uzbekistan and Japan; Africa: South Africa; North America: Canada and the USA; Central America: Mexico; Oceania: New Zealand (Figure 1).



Fig. 1. World range of *Ditylenchus destructor* Thorne

The nematode cannot withstand prolonged drying, so the species has significant economic value only if it parasitizes in cool, moist soil. In the absence of a special rest phase in the development cycle, the species overwinters in the egg phase (in temperate climates) or any other phase (in warm climates). Under favorable environmental conditions, the larvae are reborn and immediately inhabit the plants. In a temperate climate, the optimal temperature for the revival of larvae is 15-20 °C, while in South Africa this indicator is 28 °C.

Invasive larvae enter tubers mainly through wounds on their surface, as well as through holes (although there is evidence that nematodes can enter newly formed

tubers from the soil at any point). Inside the plant, nematodes actively feed and reproduce. A mature female lays about 250 eggs, which begin to develop immediately. After 4-5 days, larvae hatch from the eggs, the development of which takes 67 days to the stage of a sexually mature individual. Thus, in the middle of the tubers, one female initiates the development of several generations, the life cycle of which is on average 15-45 days (depending on environmental conditions). Nematode development and population growth continues inside the tubers after harvest.

The potato stem nematode has a slender worm-like body, the morphometric characteristics of which can vary depending on the age of the pathogen and the species of the host plant. Males and females are very similar in appearance (Figure 2).

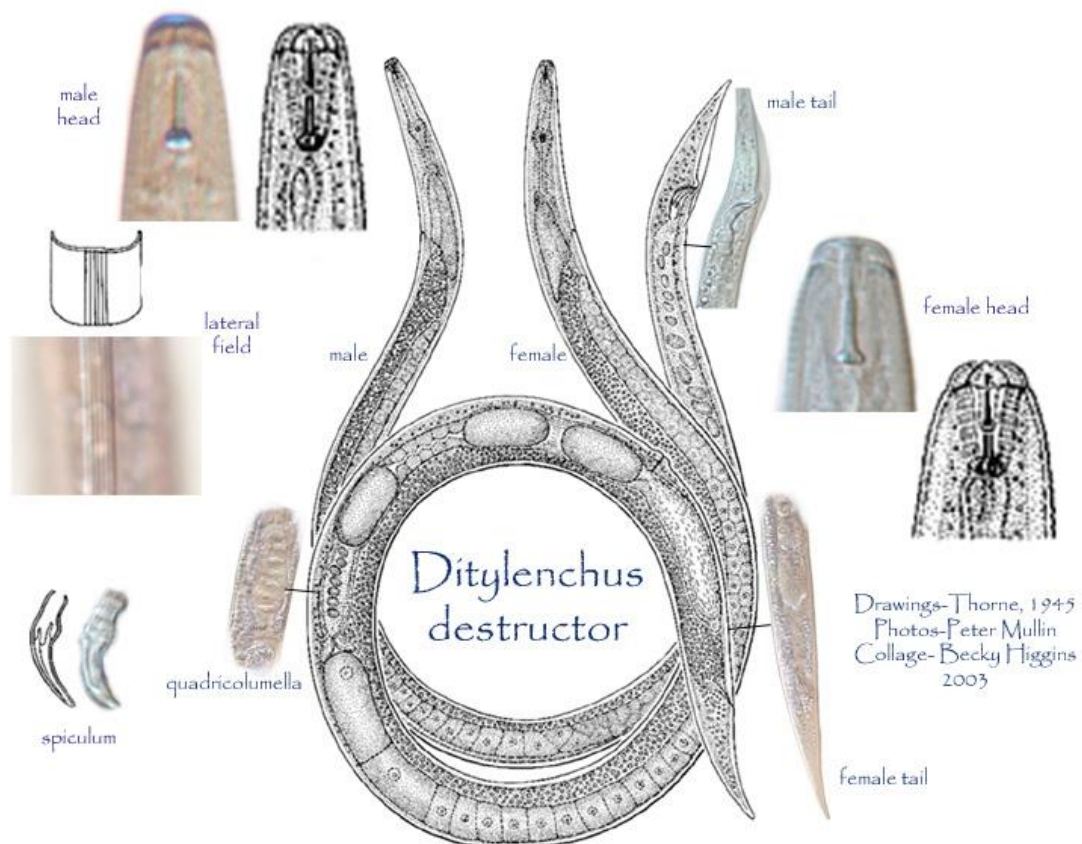


Fig. 2. Potato stem nematode

Female: 0.72-1.44 mm long and 20-30 μm wide, stylet 10-12 μm , vulva – 78-83%, ovary unpaired, anterior (Figure 2).

Male: 0.75-1.35 mm long, 20-25 μm wide, stylet 9-11 μm , spicules paired, well-developed bursa, which begins at the level of the heads of the base of the spicules and extends for 2/3–3/4, covering the tail (Figure 2).

The larva: in all 4 stages of development (the first stage takes place in the egg) is very similar to the adult, but smaller in size and does not have developed genitals. It differs from the closely related species *D. dipsaci* by the longer length of the posterior uterus and the smaller size of the eggs (Figure 2).

There are no specific terrestrial symptoms of potato disease, only with a high degree of infection, the plants have a depressed appearance and prematurely wither.

Invasion at the initial stage can be detected by cutting the skin of the tuber and seeing small white dots that stand out against the background of healthy tissue. Later, these dots increase in size, darken, and their texture changes. Over time, the disease can be detected by dark, as if depressed, spots on the surface of the tubers, in some places the skin of the tubers in these areas is separated from the pulp and shriveled. The tissues under it acquire a gray or dark brown color (the latter mainly occurs due to the colonization of tubers by secondary pathogens – fungi, bacteria and saprobiotic nematodes). However, damage to plants by other stem nematodes, *D. dipsaci*, does not lead to shriveling of the tuber skin, and the interlayer of the affected (darker) tissue penetrates the inside of the tuber in strands. Symptoms of the disease in this case are more noticeable on the above-ground organs of plants, in particular, diseased plants are distinguished by small, deformed leaves (Figure 3).



Fig. 3. Consequences of damage to potatoes by the potato stem nematode

Affection of bulbs of roosters and tulips usually starts from the bottom, spreading later to newly formed scales that cover gray and black necrotic spots. The roots of the plants also darken, the leaves do not develop well, and in some places they have yellow tips. Black spots appear on the shell of affected peanut plants, which stretch along the veins. The fruit acquires a flabby brown or black color, the embryo has brown chlorotic spots.

Nematodes spread along with infected seed and planting material (potato tubers, bulbs, rooted plants), soil and agricultural tools. Possible spread by birds, irrigation water, etc.

The best results in controlling stem nematodes are achieved using clean planting material. Stem nematodes in bulbs, tubers, roots of asparagus and strawberries are destroyed with the help of disinfection (Hydrogen cyanide). Infected bulbs of roosters are disinfected by immersing them in water containing 0.5% formaldehyde at a temperature of 43.5 °C for 2-3 hours. (contradicted for some varieties that cannot withstand such processing).

Stem nematode – *Ditylenchus dipsaci* Filipjev (KKB – DITYDI). Synonyms: *Anguillula devastatrix* Kühn, *A. dipsaci* Kühn, *A. secalis* Nitschke, *Anguillulina dipsaci* (Kühn) Gervais & Van Beneden, *A. dipsaci* var. *communis* Steiner & Scott, *Ditylenchus allocotus* (Steiner) Filip'ev & Sch. Stek., *D. amsinckiae* (Steiner & Scott) Filip'ev & Sch. *D. dipsaci* var. *tobaensis* Schneider, *D. fragariae* Kir'yanova, *D. sonchophila* Kir'yanova, *D. trifolii* Skarbilivich, *Tylenchus allii* Beijerinck, *Tylenchus devastatrix* (Kühn) Oerley, *T. dipsaci* (Kühn) Bastian, *T. havensteinii* Kühn, *T. hyacinthi* Prillieux, *T. putrefaciens* Kühn., belongs to the type Roundworms – Nematoda, order Tylenchida – Tylenchida, family Anguinidae (Anguinidae).

The stem nematode is capable of parasitizing more than 450 plant species, including weeds. Among the main fodder plants are onions, garlic (*Allium*), peas (*Pisum*), beans (*Phaseolus*), alfalfa (*Medicago*), corn (*Zea mays* L.), rye (*Secale*), potatoes (*Solanum tuberosum* L.), strawberries (*Fragaria*), beet (*Beta*), tobacco (*Nicotiana*), hyacinth (*Hyacinthus*), narcissus (*Narcissus*), tulip (*Tilira*), phlox (*Phlox*).

At the same time, *D. dipsaci* has more than 10 physiological races, for which the range of host plants is very limited. For example, a race that can reproduce on rice, rye, and onions can be considered polyphagous, since it can inhabit many other plants. It is most likely specific on alfalfa, clover, and strawberry, because there is an extremely limited range of alternative hosts for it. The tulip race can be a parasite on the daffodil, and the one found on the daffodil cannot survive on the tulip. Among the most famous races of the stem nematode, we can distinguish strawberry, red clover, alfalfa, rye, oat, beet, phlox, hyacinth, tulip, but the most harmful is the onion–garlic race. Parasitism of the latter on onions and garlic in some places leads to losses of 40–60% of the crop in field conditions, and, in addition, a significant part of it is lost during storage.

D. dipsaci is considered one of the most harmful species of phytonematodes, especially in temperate climates. Without proper control, the stem nematode can lead to a complete loss of the crop (primarily onions, garlic, cereals, strawberries, decorative bulbous plants).

D. dipsaci is locally distributed in temperate climates almost all over the world (Europe, North and South America, Africa, Asia, Australia, and Oceania), but the species is unable to survive in tropical climates except in some mountainous areas.

As of 2022, the stem nematode is common in many European countries: Austria, Albania, Belgium, Belarus, Bulgaria, Bosnia and Herzegovina, Great Britain, Greece, Denmark, Estonia, Ireland, Iceland, Spain, Italy, Cyprus, Latvia, Lithuania, Malta, Moldova, the Netherlands, Germany, Norway, North Macedonia, Poland, Portugal (including the Azores), Russia, Romania, Serbia, Slovakia, Slovenia, Hungary, Ukraine, Finland, France, Croatia, the Czech Republic, Montenegro, Switzerland and Sweden; Asia: Azerbaijan, Armenia, Georgia, Yemen, Israel, Iraq, Iran, Jordan, Kazakhstan, Kyrgyzstan, China, Cyprus, Korea (South), Oman, Pakistan, Syria, Turkey, Uzbekistan, Japan (Honshu Island); Africa: Algeria, Kenya, Morocco, South Africa, Reunion Island and Tunisia; North America: Canada and the USA; Central America and the Caribbean: Haiti, the Dominican Republic, Costa Rica and Mexico; South America: Argentina, Bolivia, Brazil, Venezuela, Ecuador, Colombia, Paraguay, Peru, Uruguay and Chile; Australia and Oceania: Australia and New Zealand (Figure 4).



Fig. 4. World range of *Ditylenchus dipsaci* Filipjev

D. dipsaci is a migratory endoparasite that inhabits parenchymal tissues of stems, bulbs, and tubers of plants, causing the destruction of the inner layer of cell walls.

After diving into plants, stem nematode larvae molt several times, turning into adult males and females. After fertilization, the female lays eggs (from 200 to 500),

from which the next generation of nematodes develops immediately, without a resting stage, and the cycle repeats again.

With an increase in the number of nematodes inside the plant, the disease progresses rapidly. Later, the dead plant cells are colonized by other microorganisms and rot, while the nematode larvae crawl onto healthy parts of the same plant or migrate into the soil, where they infect new plants. During one growing season, several generations of stem nematode develop, the duration of development of each of which, depending on the fodder plant and environmental conditions, can be from 20 to 73 days.

The stem nematode can maintain viability for many years both in air-dry conditions and in clay soils. It has been established that the harmfulness of nematodes is much higher on such soils than on sandy soils.

To isolate nematodes, it is enough to grind the suspicious plant organ and put it in water: the nematodes will leave plant remains and actively move in the water. An 800-fold increase is sufficient for morphological and morphometric studies. Nematodes at all stages of development have a slender worm-like body, narrowed on both sides (Figure 5).

Female 1.2 mm long ("giant race" on beans – 2 mm); stylet – 10-12 μm with clearly developed basal tubercles. Ovary unpaired, vulva – 80-82%; lateral fields with four incisions; the terminus of the tail is sharp (Figure 5).

Male 1.0-1.3 mm long, 27-34 μm wide; spicules paired; a bursa is present, which starts from the base of the spicules and ends before reaching the end of the tail (Figure 5).

Nematode parasitism often causes deformation of shoots, leaves and flowers of plants, necrotization and subsequent rotting of the neck of the stem, roots, bulbs and tubers are observed.

During the growing season, onion plants affected by ditylenchosis have chlorotic, deformed leaves that wilt prematurely. Deformation of leaves has not been established on garlic plants, but they turn yellow prematurely and die; in some places the stalk of garlic thickens, cracks appear on it. The bulbs become plump, and their bottoms become rotten (Figure 6).

Garlic heads are mostly loose and loose. By cutting a damaged bulb crosswise, you can easily notice unevenly thickened scales, which later turn brown or gray because of their colonization by various putrefactive microorganisms. A clear sign of damage to plants by ditylenchosis is round or crescent-shaped cracks at the bottom of the bulbs. Sometimes the thickened outer and inner scales of the bulb gradually slide up, forming "rags" in the area of the bottom.

Ditylenchus dipsaci
(After Thorne) Thorne, 1961

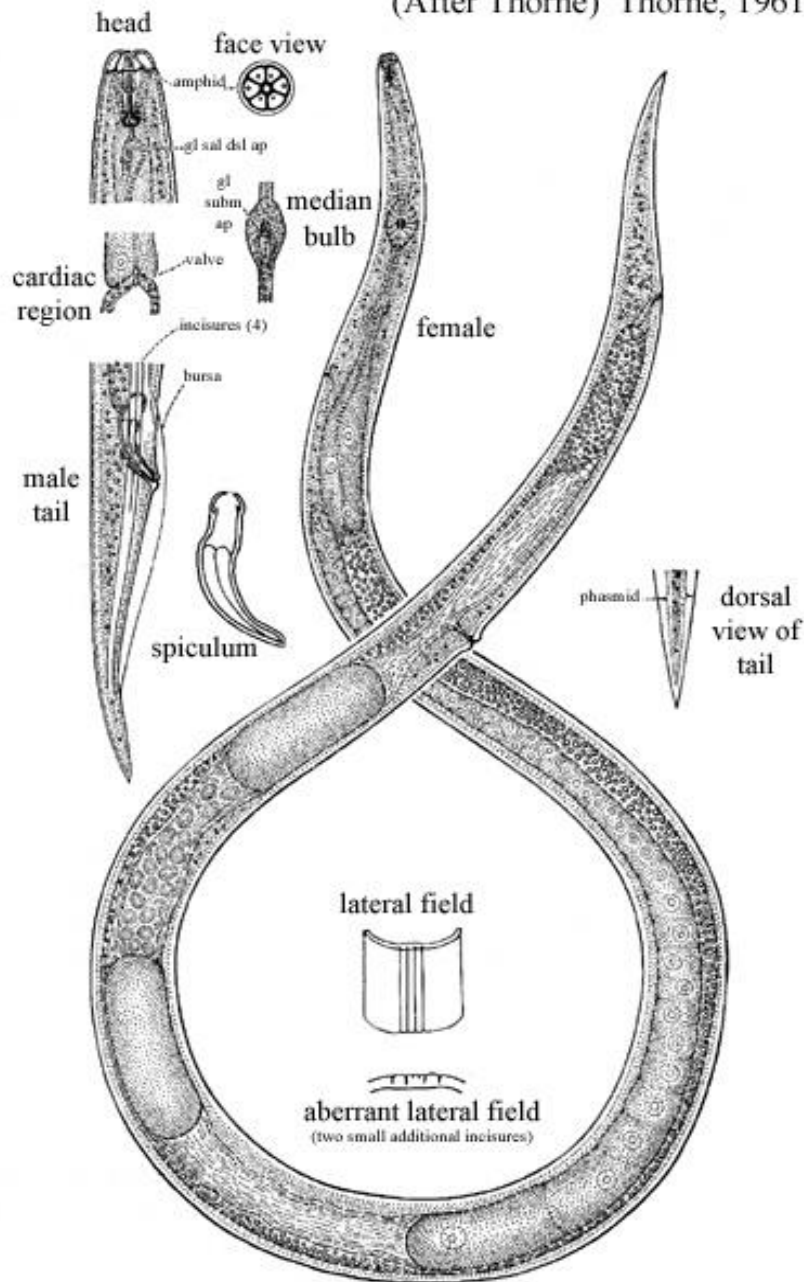


Fig. 5. Stem nematode

Another visual sign of ditylenchosis is the formation of the so-called nematode "felt" on the surface of affected bulbs during their storage in storage (Figure 7).

It has a grayish color, very similar to mold. These are tens of thousands of nematode individuals that have crawled to the surface of the affected dry bulb and are there in a resting stage until more favorable conditions for further growth and development occur. During storage, the specific strong smell of bulbs and garlic affected by ditylenchosis is especially clearly manifested, which also helps to identify the nematode disease.



Fig. 6. Bulbs affected by stem nematode

It is difficult to diagnose a weak infection, because in this case the outer scales of the bulbs have a completely healthy appearance, while the inner ones are destroyed, sometimes completely. Such "empty" bulbs are characteristic of an infected seedling that is stored at relatively high temperatures. On alfalfa crops, the disease manifests itself in clusters, more strongly in a humid climate. Infected plants are delayed in growth and development, the base of the stem thickens (swells), it becomes noticeably shorter. With a strong degree of damage, plants die. Parasitism of the stem nematode on tobacco plants also causes deformation of the base of the stem, which subsequently breaks ("stem break").

In addition to the above symptoms (swelling, deformation of the stem), necrosis is visible on the beans, which later turn red-brown, later – black (depending on the variety and environmental conditions). Necrosis covers the stem over time and increases in size. Newly formed fruits have a dark brown color. Infected seeds are darker, smaller, and sometimes speckled. A giant race causes more symptoms on culture. In natural conditions, in the absence of a host plant or in the case of arid conditions, *D. dipsaci* can survive for many years. The nematode spreads mainly together with seed and planting material (in particular, with bulbs), being both in the middle of tissues and on the surface – in the form of "nematode felt". The distribution of nematodes is also possible together with agricultural tools, sewage and rainwater, plant remains, weeds, birds, etc.



Fig. 7. Nematode "felt"

One of the effective ways to prevent the spread of *D. dipsaci* can be the timely culling of diseased plants throughout the entire cycle, starting with the seed material before planting, then – the detection of ditylenchosis foci directly in the field, then – picking the harvested crop before putting it in storage, and finally – periodic removal affected plants (bulbs, garlic, etc.) during storage. Preventive measures should also include the removal of weeds and post-harvest residues from the infected areas, deep early plowing helps the best decomposition of post-harvest residues in the soil. The use of crop rotations cannot effectively control the spread of infection because the stem nematode has many host plants. Due attention must be paid to the cleanliness of agricultural tools, containers, and storage facilities. They can be disinfected using a 4% formalin solution.

Crops, planting material or seeds collected from infected areas should not be stored for a long time, especially next to uninfected lots; it should be used only for commercial purposes. If it is necessary to use seed or planting material from these batches, it is recommended to pre-treat with hot water, the temperature regime of which operation depends on the type of plant material and its condition.

It is recommended to use nematicides on some ornamental plants. Certain efficiency is provided using nematode-resistant plant varieties.

Conclusions. 1. According to the results of the analysis of domestic and foreign professional information sources, it was established that in Ukraine there are two types of stem nematodes limited to common quarantine species: potato stem nematode (*Ditylenchus destructor* Thorne) and stem nematode (*Ditylenchus dipsaci* Filipjev).

2. In contrast to European countries, where the potato stem nematode does not have significant economic importance, in Ukraine its massive spread in the field and the manifestation of the disease during the storage of agricultural products leads to significant crop losses.

3. The stem nematode is considered one of the most harmful species of phytonematodes, especially in temperate climates. Without proper control, the stem nematode can lead to a complete loss of the crop (primarily onions, garlic, cereals, strawberries, decorative bulbous plants). *D. dipsaci* is locally distributed in temperate climates almost all over the world (Europe, North and South America, Africa, Asia, Australia and Oceania), but the species is unable to survive in tropical climates except in some mountainous areas.

References:

1. Bashinska O.V. (2009). Ilyustrovaniy dovidnik regulovanih shkidlivih organizmiv v Ukrayini. Kyiv: Urozhaj, 249.

2. European and Mediterranean Organization for Quarantine and Plant Protection. Official site. URL: [https://www.eppo.int/european and Mediterranean Plant protection organization](https://www.eppo.int/european-and-Mediterranean-Plant-protection-organization).

3. Fedorenko V.P. et al. (2012). Strategiya i taktika zahistu roslin. T. 1. Strategiya. Kyiv: Alfa-steviya, 500.

4. Nasonova L.V. (2008). Nematody, mollyuski, kleshi, gryzuny, vredyashie selskohozyajstvennym rasteniyam: ucheb.-metod. posobie N. Novgorod: Nizhegorodskaya gos. s.-h. akademiya, 163.

5. Stankevych S.V. (2017). Metodi oglyadu ta ekspertizi pidkarantinnih materialiv: navch. posib. Kharkiv: FOP Brovin O. V., 255.

6. Stankevych S.V. et al. (2020). Monitoring shkidnikiv i hvorob silskogospodarskih kultur. Kharkiv: FOP Brovin O. V., 624.

7. Stankevych S.V. et al. (2021). Karantinni organizmi (z osnovami ekspertizi pidkarantinnih materialiv). Kharkiv: FOP Brovin O.V., 459.

8. Stankevych S.V. et al. (2022). Karantinni fitonematodi. Zhitomir: Vidavnictvo «Ruta», 94.

9. Stankevych S.V., Lezhenina I.P., Zabrodina I.V. (2022). Karantinni organizmi, obmezheni poshireni v Ukrayini. Kharkiv: Vidavnictvo Ivanchenka I.S., 140.

10. Stankevych S.V., Lezhenina, I.P., Zabrodina I.V. (2022). Regulovani nekarantinni shkidlivi organizmi. Kharkiv: Vidavnictvo Ivanchenka I.S., 75.