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COMMUNITIES OF *DITYLENCHUS DESTRUCTOR* SATELLITE SPECIES OF NEMATODES IN INFECTED POTATO TUBERS: SPECIES COMPOSITION OF PHYTONEMATODE COMPLEX AND THE STRUCTURE OF THEIR INFRACOMMUNITIES

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Communities of *Ditylenchus destructor* Satellite Species of Nematodes in Infected Potato Tubers: Species Composition of Phytonematode Complex and the Structure of Their Infracommunities. Sigareva, D. D., Fedorenko, S. V., Bondar, T. I., Sokolova, O. O., Kornyushin, V. V. — Within September–April 2016–2017 potato tubers affected by the potato rot nematode *Ditylenchus destructor* were selected in the potato storage facilities of the Institute of Potato Production of NAAS. 18 species of phytonematodes were identified, including an *edificator species* of microparasitocenosis and 17 satellite species of this pathogenic phytohelminth, mycohelminths (6 species) and saprobiotic nematodes (11 species). These phytonematodes are representatives of two orders, Rhabditida (11 species from 4 families) and Tylenchida (7 species from 4 families). Nematodes that are part of the group formed by *D. destructor* and its satellite species are divided into five groups according to quantitative indicators (the prevalence of invasion P and intensity of invasion I). The dominant is one species — *D. destructor*. Satellite species of *D. destructor* are divided into four groups, these are subdominants (4 species) and common species (3 species), which together with the dominant species form the core of the group, as well as rare (6) and accidental (4). It was shown that in the course of the disease a regular succession occurs, the species composition of phytonematodes, the grouping structure, the relationship between the number of species of various trophic groups, and the number of certain species change. There are five stages in the course of the disease, from *D. destructor* colonisation of the tuber, the penetration of the mycohelminths and saprobionts into the focus of the disease, and the disappearance of *D. destructor*. The maximum number of *D. destructor* occurs at stage III, mycohelminths — at stage IV, and saprobiotic nematodes — at stage V. Infracommunities consist of from two to seven species. The distribution of all the studied tubers by the number of species in the infracommunity is close to normal, most often 3–4 species of nematodes were found, several species of nematodes of the same genus are extremely rare in a particular infracommunity.
K e y w o r d s: potato rot nematode, *Ditylenchus destructor*, phytohelminths, mycohelminths, saprobionts, phytonematode community structure, infra community, satellite nematodes.

Introduction

G. Thorne (1945) based on a detailed study of morphological features, distinguished the potato rot nematode as a separate species of the genus *Ditylenchus*, specific to potato and named it *Ditylenchus destructor* Thorne, 1945. It is similar to *D. dipsaci* (Kühn, 1854), but according to J. B. Goodey (1951), is different in two features: does not form a layer of nematodes on infected tubers or roots and never forms as many larvae as *D. dipsaci* and more often experiences adverse conditions in the egg stage. It is capable of hibernation at all stages of development. It takes 40 days to develop one generation at a temperature of 12–15 °C, and only 20 days at a temperature of 30 °C (Ustinov, 1955). The acidity of the medium does not matter much — it can exist at pH from 3.5 to 8.6. *D. destructor* populations in tubers usually consist of females, males, larvae and eggs. Males and females are morphologically similar, although females are somewhat longer and thicker.

Most often, the nematode affects the underground parts of the potato — tubers and stolons. Occasionally, it can also be found in the aerial parts of plants (Christie, 1959; Krylov, 1962). Penetration of the nematode into young tubers can occur in 2 ways: from the stems of the infected mother tuber or through the soil. At the penetration sites, the tuber peel is discolored, and then acquires a lead-gray color and is easily removed from the tuber pulp, often cracking of the peel is observed on the surface, the infected parts look depressed compared to healthy ones. Slowly the whole tuber collapses as dry rot. Reproduction of the nematode occurs at the border of healthy and infected tissue. Saprophytic fungi and bacteria penetrate into the infected part of the tuber and provide food for mycohelminths and saprobiotic nematodes (Kiryanova, Kral, 1969, 1971). Therefore, infected with *D. destructor* potato tubers, as a rule, are populated by a whole complex of phytonematodes. In tubers infected only with *D. destructor*, where dry and wet rot have not yet spread, only this species is found. If wet rot appeared in infected tubers, the number of tuber nematodes significantly decreases, but the number of saprophytes and mycohelminths that feed on saprophytic bacteria and fungi increases. Already during the first studies of affected by *D. destructor* potato, the authors noted that saprobiotic nematodes and mycohelminths (genera *Rhabditis*, *Diplogaster*, *Cephalobus*) subsequently penetrate infected potato tubers (Chukantseva, 1972; Skarbilovich, 1951). R. G. Ryss (1962) indicates the presence of *Aphelenchus avenae* Bastian, 1865, *Neotylenchus abulbosus* Steiner, as well as representatives of the genera *Hexatylus* Goodey, 1926, *Neotylenchus* Steiner, 1931 in infected tubers. A study of the nematode fauna of potato tubers in storage conditions (potato storages) revealed 18 phytonematodes species belonging to 4 orders (Chromadorida, Enoplida, Rhabditida, and Tylenchida), 7 families and 14 genera, among which representatives of the *Rhabditis* and *Mesodiplogaster* predominated. The majority of nematodes (58.2 %) were found in the tuber pulp. Later the author gives the species composition of nematodes that contains only saprobiotic species.

In Ukraine a complex of *D. destructor* satellite nematodes was investigated by L. Kostiuk (1999), K. Butenko (2004) and T. Zhilina (2004). The species composition of the phytonematode complex in affected potato tubers was most fully and thoroughly analyzed in the studies of T. Zhilina (2004). She identified 20 species of nematodes in infected tubers that belong to 17 genera, 8 families and 2 orders. The nematode complex consisted of phytohelminths — 1 species (5 %), mycohelminths — 5 species (25 %) and saprobionts — 14 species (70 %). By Cassagnau's coefficient of species constancy (1961), the author considers the dominant species that are found in > 50 % of samples, frequent 5–50 %, rare — < 5 %. *D. destructor* dominated among phytohelminths being found in 93.2 % of tubers, *Mesodiplogaster lheritieri* Goodey, 1963, was found in 56.8% of samples among saprobionts. Six species of saprobionts and three species of mycohelminths are common. The remaining 11 species were rare.

So, a complex of phytonematode species where several species coexist is gradually formed in potato tubers affected by *D. destructor*. They are mainly representatives of all three ecotrophic groups of nematodes — phytohelminths (*D. destructor*), as well as mycohelminths and saprobionts. The species composition of *D. destructor* satellite nematodes can vary significantly depending on the regions and storage conditions of tubers.

Material and methods

The bases for collecting material were the potato storage facilities of the Institute of Potato Production of NAAS. Samples of potato contaminated with tuber rot nematodes were collected during the planned sorting of tubers during the potato storage period. Selection of tubers for analysis was carried out within 8 months (September–April) 2016–2017. During the specified period, 47 tubers of potatoes were analyzed.

Berman's funnel method was used to isolate phytonematodes from potato tubers (Sigareva, 1986). For this purpose, each studied potato was thoroughly washed under a stream of running water; the infected part in the area of a stolon was cut and finely ground. The 10 g hinge was placed on paper filters, which were set in advance on brass support nets. The tuber sample on the grid was dipped into the funnel so that the sample was covered with a thin layer of water. Exposure of extraction of phytonematodes from the samples lasted 72 hours. Tubes with nematodes were fixed with TAF. Temporary water–glycerol preparations were prepared from fixed nematodes according to the method of E. S. Kiryanova (Kiryanova, Kral, 1971). Nematodes were examined and identified using an MBI-15 microscope. Ecological-trophic grouping of nematodes was performed according to Zubin (1972), Van Gundhi, Freckman (1977).

To identify tuber nematodes by morphological features, the most commonly used in nematology indices were applied. They were calculated according to the diameters of different parts of the body and certain morphological formations using De Man formula (Dekker, 1972). According to this formula, the most important for identification of phytonematodes are the following measurements of body parts and their ratio:

L — total body length, a = total length/esophagus length, b = total length/greatest body diameter, c = total length/tail length, V, % (in females) — the distance from the head end of the body to the vulva as a percentage of the total body length.

According to De Man formulae, the sizes of females are as follows: L = 0.72–1.44 mm; a = 33–35; b = 8–10; c = 15–20; V = 78–83 %; males: L = 0.75–1.3 mm; a = 34–40; b = 7–8; c = 12–16. In males and females, the chitinous stylet of the head is well developed. Spear with rounded heads typical of the genus, its average length is 10 μm. Basal dilatation of the esophagus with 3 large and 2 small nuclei, located just beyond the junction of the esophagus with the intestine (Thorne, 1961). The cuticle of the body is annular near the head, its rings about 1 μm wide and relatively indistinct. The lateral margin of the cuticle with 6 notches (lines). Females: the ovary extends almost to the junction of the esophagus with the intestine, oocytes are arranged in 2 rows. The length of the eggs in the uterus is slightly greater than the width of the body, the ratio of their length to width is 1.5 to 1. The vestigial posterior uterus does not function as spermatheca. The distance from the vulva to the anus is 1.75–2.5 times the length of the tail. Males: with spicules that have elongated ribs, one of which is shorter than the other. The bursa begins at the level of the heads of the spicules and continues backward at a distance equal to 2/3 of the length of the tail.

Potato rot nematodes were obtained by cutting a piece of parenchyma from the affected potato on the verge of the affected and healthy tissue. The sample was transferred into a drop of water on a glass slide and viewed under a microscope. The males and females of *D. destructor* have a filiform shape, their movements are serpentine, slow and awkward, the stylet is clearly visible in the buccal cavity. As tissue breaks up, the potato rot nematode migrates to adjacent healthy tissue. Saprophytic nematodes, bacteria and fungi develop in the rotting parts, thereby increasing the lesion (Chukantseva, 1972).

Saprophytic nematodes have more coarse body, characterized by fast and irregular snake-like movements, as well as the absence of a stylet (Ryss, 1962). In order not to confuse them with *D. destructor*, it is useful to apply the method of coloring with a 1% solution of methylene blue proposed by Professor A. A. Paramonov (1956). Within 5–10 minutes, the saprophytic nematodes turn blue, *D. destructor* are not stained (Muge, 1959).

To determine the status of dominance the indicators of prevalence (P) and intensity of invasion (I) were used:

$$P= N_1/N_2\times100,$$

where: N_1 — number of samples where definite species was recorded; N_2 — number of investigated samples.

And $I = N_1/N_2$,

where: N_1 — general number of nematodes of a definite species; N_2 — number of tubers affected by this nematode species.

The species with a detection rate of P > 50 % of the samples were considered as dominant, the subdominant — 20–50 %, the common 20–10 %, rare < 10 %, those of the latter where only a few specimens were found, were considered as random (Kornyushin, 2011).

Results

Taxonomic and ecotrophic structure of *D. destructor* satellite phytonematode communities in infected tubers.

In total, we found 18 species of phytonematodes with species edificator *D. destructor* and 17 satellite species (fig. 1). Infracommunities of nematodes in the affected potato tubers belong to 14 genera, 9 families and 2 orders in potato tubers. The order Rhabditida, to which 5 families, 10 genera and 11 species belong, is most fully represented by number of species. The order Tylenchida includes 4 families, 5 genera, and 7 species of phytonematodes.

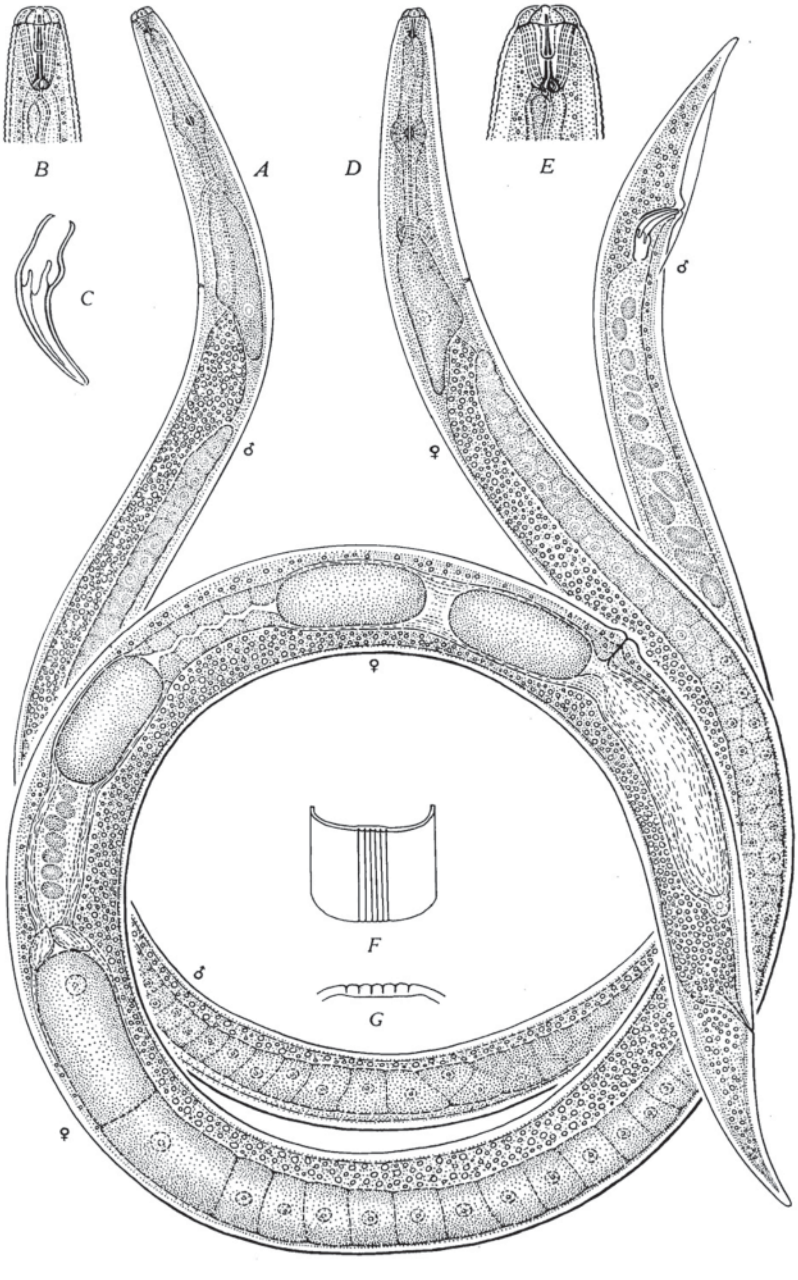

Fig. 1. *Ditylenchus destructor* (Thorne, 1945) potato nematode: *A* — female; *B* — head end of the female; *C* — spicules; *D* — male; *E* — head end of the male; *F, G* — lateral field by the Thorne, 1945.

Systematic list of phytonematode species found in potato tubers:

Nematoda (Rudolphi, 1808)

Order Rhabditida Chitwood, 1933

Family Diplogasteroididae Paramonov, 1952
Genus *Diploscapter* Cobb, 1913
Diploscapter rhizophilus Rahm, 1928/1929
Family Cephalobidae (Filipjev, 1934)

Genus *Cephalobus* Bastian, 1865
Cephalobus persegnis Bastian, 1865
Genus Eucephalobus Steiner, 1936
Eucephalobus oxiuroides (de Man, 1876) Steiner, 1936
Eucephalobus mucronatus (Kozlowska, Roguska-Wasilewska, 1963) Andrassy, 1967
Genus *Acrobeloides* (Cobb, 1924)
Acrobeloides buetschlii (de Man, 1884) Steiner and Buhrer, 1933
Genus *Chiloplacus* Thorne, 1937
Chiloplacus symmetricus (Thorne, 1925) Thorne, 1937
Family Rhabditidae Oerley, 1880
Genus *Caenorhabditis* Osche, 1952
Caenorhabditis elegans (Maupas, 1900) Dougherty, 1953
Genus *Mesorhabditis* (Osche, 1952)
Mesorhabditis monhystera (Butschli, 1873) Dougherty, 1955
Genus *Rhabditis* Dujardin, 1845
Rhabditis sp.
Genus *Pelodera* Schneider, 1866
Pelodera teres Schneider , 1866
Family Panagrolaimidae (Thorne, 1937)
Genus *Panagrolaimus* Fuchs, 1930
P*anagrolaimus rigidus* (Schneider, 1866) Thorne, 1937

Order Tylenchidae (Filipjev, 1934)
Family Anguinidae Nicoll, 1935
Genus *Ditylenchus* Filipjev, 1934
Ditylenchus destructor Thorne, 1945
Family Tylenchidae Oerley, 1880
Genus *Aglenchus* (Andrassy, 1954)
Aglenchus agricola (de Man, 1834) Andrassy, 1954
Family Neotylenchidae Thorne, 1941
Genus *Nothotylenchus* Thorne, 1941
Nothotylenchus sp.
Family Aphelenchidae (Fuchs, 1937)
Genus *Aphelenchus* Bastian, 1965
Aphelenchus avenae Bastian, 1965
Family Aphelenchoididae (Skarbilovich, 1947)
Genus *Aphelenchoides* Fischer, 1894
Aphelenchoides asterocaudatus Das, 1960
Aphelenchoides bicaudatus (Immamura, 1931) Filipjev et Sch. Stekhoven, 1941
Aphelenchoides limberi Steiner, 1936

Within the order Tylenchida four family are distinguished: Anguinidae, Tylenchidae and Aphelenchidae, Aphelenchoididae of which Anguinidae is represented by phytohelminths and three other mycohelmints.

The order Rhabditida consists of four families: Diplogasteroididae, Cephalobidae, Rhabditidae and Panogrolaimidae, represented exclusively by saprobiotic species. Family Diplogasteroididae contains only one genus *Diplogaster*. The Cephalobidae family contains four genera : the *Cephalobus, Eucephalobus, Acrobeloides* and *Chiloplacus*. The Rabditidae contains four genera too: *Cenorhabditis, Mesorhabditis, Rhabditis* and *Pelodera.* The Panogrolaimoides family contains only one genus Panogrolaimus. Most (13 of 15) genera are also represented by one species (9), the genus *Eucephalobus* — by two species, and only the genus *Aphelenchoides* includes three species (fig. 2, A).

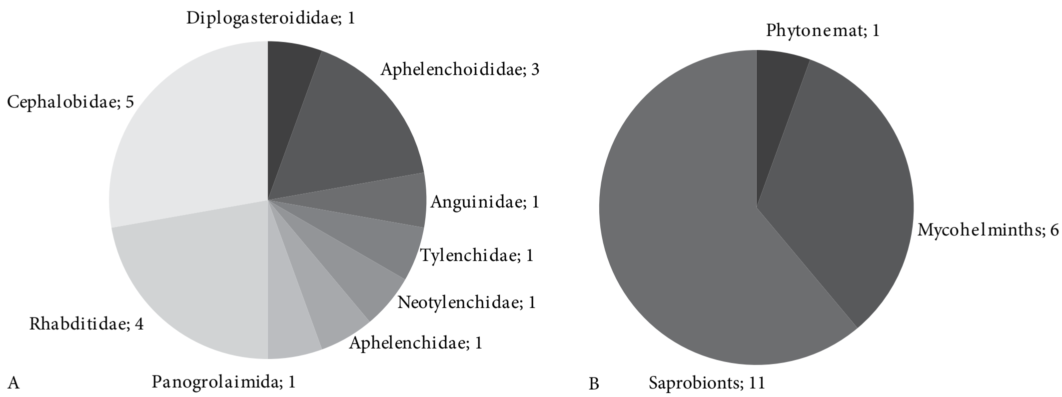

Fig. 2. Structure of phytonematode communities inhabiting tubers damaged by *D. destructor* (A — taxonomic structure, B — ecotrophic structure).

Along with *D. destructor*, which is a pathogenic phytohelminth and an edificator species, two more ecotrophic groups of nematode satellites of *Ditylenhus* are included in the community of phytonematodes, inhabiting the lesion site caused by ditylenchosis that occur in potato tubers during storage in winter storage facilities (fig. 2, B). Representatives of mycohelminths settle in potatoes affected by *D. destructor* because this species destroys the living tissues of tubers in the course of life, creating favorable conditions for the settlement of certain species of fungi. It is the colony of these fungi consumed by mycohelminths nematodes**.** Such species are six in four genera. *Aphelenchoides asterocaudatus* and *Aph. limberi* predominate in frequency and number of infrapopulations (table 1).

Representatives of ecotrophic groups of nematodes appear in the affected tubers already in the first stage of the course of the disease (fig. 3).

Representatives of the second ecotrophic group of saprobiont nematodes are found in the areas of ditylenchosis lesion of potato tubers a little later, when the part of the cells of the potato parenchyma is destroyed and the fungi are joined by saprobiotic microorganisms, and *saprobiont nematodes* feed on them. This ecotrophic group of nematodes is more diverse, with 11 species from 10 genera. Three species *Mesorhabditis monhystera*, *Acrobeloides buetschlii*, and *Chiloplacus simmetricus* predominate in frequency of occurrence and population size (table 1).

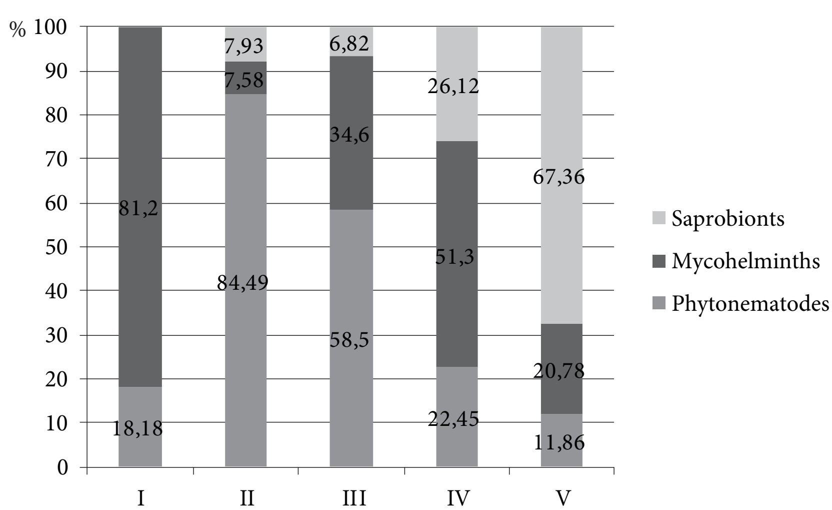

Fig. 3. The dynamics of the ratio of the total number of various trophoecological group nematodes during the disease of potato tubers caused by *D. destructor*.

Table 1. Distribution of recorded species of phytonematodes by ecotrophic groups in tubers infected with *D. destructor* (2016–2017)

No	Species, ecotrophic groups	The total number of nematodes in all analyzed tubers, specimens				
	Stage of the disease development	I	II	III	IV	V
	Number of tubers	1	10	14	14	8
	Phytonematodes					
1	*Ditylenchus destructor*	10	4350	7553	729	615
	Total	10	4350	7553	729	615
	The proportion of phytonematodes in the total number of identified nematodes, %					
	Mycohelminths					
2	*Aphelenchoides asterocaudatus*	35	253	1064	279	
3	*Aphelenchoides bicaudatus*		1	211		1
4	*Aphelenchoides limbery*		50	3163	1309	1041
5	*Aphelenchus avenae*		56	7	3	1
6	*Aglenchus agricola*	10	30			35
7	*Nothotylenchus* sp.			33	79	
	Total	45	390	4478	1670	1078
	The proportion of mycohelminths in the total number of detected nematodes, %					
	Saprobionts					
8	*Caenorhabditis elegans*			1		2886
9	*Eucephalobus oxiuroides*			57		11
10	*Eucephalobus mucronatus*			3		
11	*Chiloplacus simmetricus*		219	82	31	238
12	*Acrobeloides buetschlii*		176	721	7	249
13	*Diploscapter rizophilus*			5	11	3
14	*Cephalobus persegnis*					2
15	*Mesorhabditis monhystera*		13	10	120	4
16	Rhabditis sp.				679	93
17	*Pelodera teres*					8
18	*Panagrolaimus rigidus*			2		
	Total	0	408	881	848	3494
	The proportion of saprobionts in the total number of detected nematodes,%					
	Total	55	5148	12912	3247	5187

In general, the taxonomic structure of the phytonematode complex, which is formed in the affected by *D. destructor* potato tubers under conditions of preservation in the storage of the Institute of Potato Production of NAAS of Ukraine, consists of five groups, which are determined by quantitative indicators (fig. 4).

According to the method of tuber selection for the study, potatoes infected only with *D. destructor* were included into the common sample. Therefore, the intensity of infestation by this species of nematodes, which is an edificator for the phytonematode satellite community, is a priori absolute (100 %). *D. destructor* is a dominant phytonematode in this community. However, the infestation rate of this species (number of infrapopulations) in one tuber varies widely from 1 specimen to 2,128–5,235 specimens, with an average infection (I) of 282 specimens, which is quite high. In addition, in 4 cases of the studied tubers only *D. destructor* was detected in the absence of any nematode satellite species.

The group of subdominant species is quite large, with four species (P 30–50 %). Two species of mycohelminths *Aphelenchoides asterocaudatus* and *Aphelenchoides limbery*

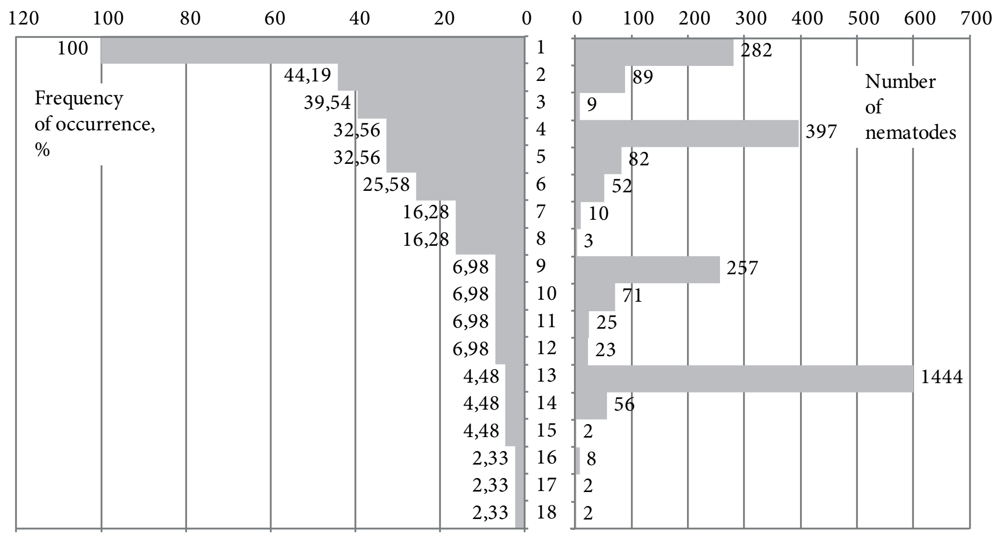

Fig. 4. Structure of phytonematode community in the foci of *D. destructor* infected tubers. Dominant: 1— *D. destructor*; subdominants: 2 — *Aphelenchoides asterocaudatus*, 3 — *Mesorhabditis monhystera*, 4 — *Aphelenchoides limbery*, 5 — *Acrobeloides buetschlii*, 6 — *Chiloplacus simmetricus*; common species: 7— *Aphelenchus avenae*, 8 — *Diploscapter rizophilus*; rare species: 9 — *Rhabditis* sp., 10 — *Aphelenchoides bicaudatus*, 11— *Aglenchus agricola*, 12 — *Eucephalobus oxiuroides*, 13 — *Caenorhabditis elegans*, 14 — *Nothotylenchus* sp.; accidental species: 15 — *Eucephalobus mucronatus*, 16 — *Pelodera teres*, 17 — *Panagrolaimus rigidus*, 18 — *Cephalobus persegnis.*

and two species of saprobionts *Mesorhabditis monhystera* and *Acrobeloides buetschlii* are among them. The intensity of infection is usually average, although it can reach very high values — 397 (up to 1260–3144) specimens of *Aphelenchoides limbery*. At the same time, *Mesorhabditis monhystera* is relatively small in number — 9 (1–67) specimens.

Slightly smaller is the group of common species (P 10–30 %), which includes only 3 species, one species of the mycohelminthic *Aphelenchus avenae*, which is small in number, 10 (1–50). Of two species of saprobiont nematodes, one *Chiloplacus simmetricus* has an average abundance I of 52 (1–219) specimens, while the abundance of the other species, *Diploscapter rizophilus*, is very low — I only 3 (1–7) specimens.

All other *D. destructor* satellite phytonematodes were not common. These ten species were found 1–3 times (P 2.33–6.98 %) and, accordingly, they are classified as rare. However, the various species of this group differ significantly from each other in terms of the number of infrapopulations. A significant part of rare species has an average level of abundance (23–71 specimens). In particular, these are all three species of mycohelminths assigned to this group (*Aphelenchoides bicaudatus*, *Aglenchus agricola* and *Nothotylenchus* sp.), as well as *Eucephalobus oxiuroides* from saprobionts. Two more species of saprobionts are distinguished by high (*Rhabditis* sp. — 257 specimens) and ultrahigh (*Caenorhabditis elegans* — 1444 specimens) numbers in the affected tubers. Probably, their rare occurrence in our samples is due to some external factors for the system. The remaining four species of saprobionts (*Pelodera teres*, *Eucephalobus mucronatus*, *Panagrolaimus rigidus* and *Cephalobus persegnis*) were found 1–2 times in an amount of less than 10 specimens. They can be considered random components of the *D. destructor* satellite nematode community.

In general, subdominant species make up 23.53 % of all 17 *D. destructor* satellite species. Common species make up another 17.65 %, while rare species make up 58.83 %, of which 23.53 % are accidental species. The specific frequency of occurrence is repeated for two pairs (14 and 7 times), 4 species were found 4 times and 3 and 2 species were recorded 3 times (fig. 4).

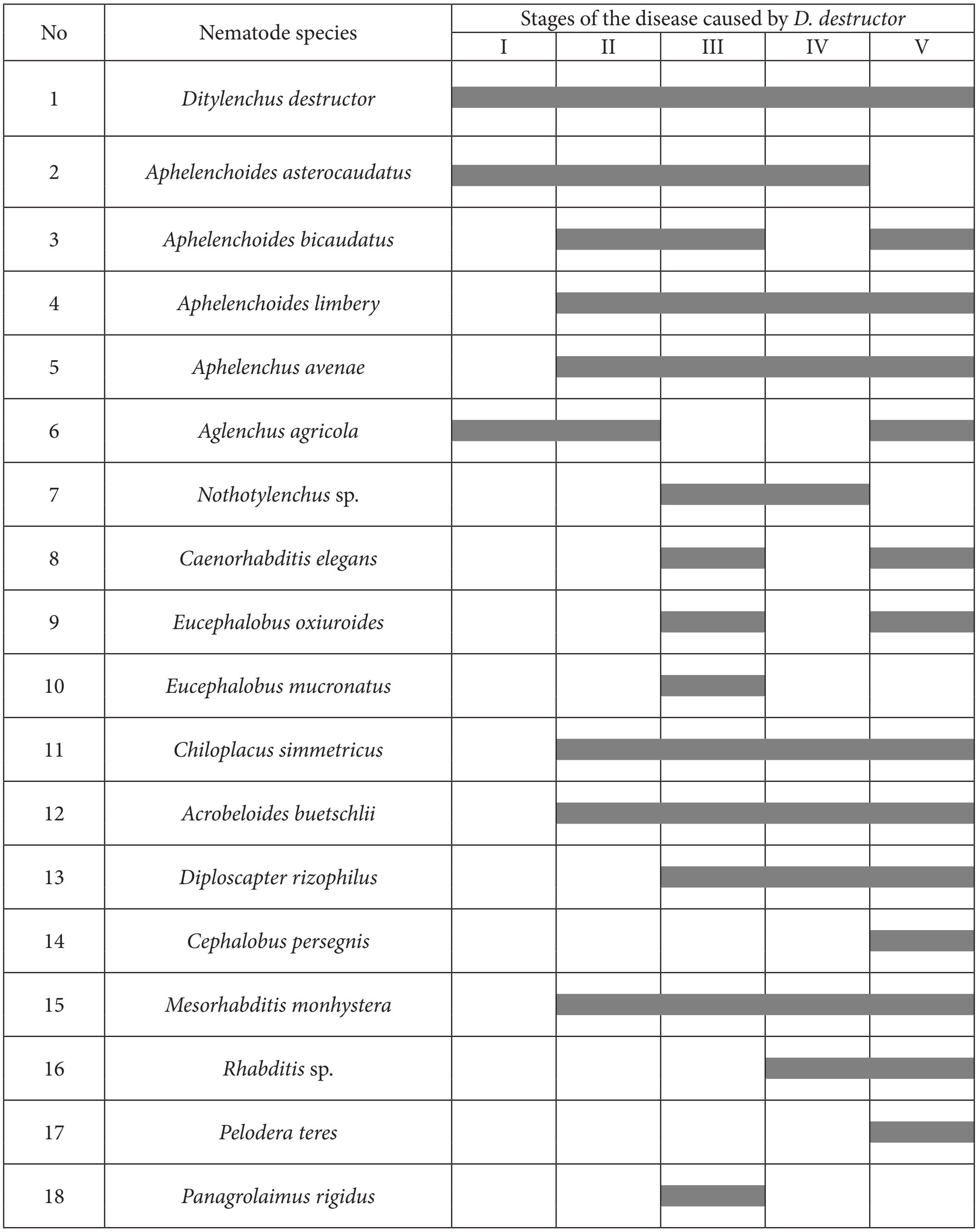

Fig. 5. Occurrence of various species of phytonematodes during successive stages of the pathological process.

Tubers occupancy by phytonematodes at different stages of the course of the disease.

According to the nature of the development of the potato tubers disease, most authors (Paramonov, Bryushkova, 1956; Zhilina, 2004, and others) divide it into 5 main stages, taking into account the appearance of diseased tubers and the cells occupancy by various groups of phytonematodes: phytohelminths, mycohelminths, and saprobionts (table 2). We took the same approach. At stage I of the disease, the tubers are almost no different from healthy ones; in the affected tissues, *D. destructor* is in small amounts (10 specimens per 10 g of tissue) as well as mycohelminthics, namely *Aphelenchoides asterocaudatus* (35 specimens) and *Aglenchus agricola* (10 specimens in 10 g of tissue). At stages II and III of the disease, the number of potato rot nematodes increases sharply (up to 435 and 540 specimens per 10 g of tissue). At the same time tubers are being populated by mycohelminths (up to 40 and 320 specimens in 10 g of tissue) and saprobionts (41 and 67 specimens in 10 g of tissue). At these stages, *Aphelenchoides asterocaudatus* and *Aphelenchoides limberi* (76 and 226 specimens per 10 g of tissue) reached the highest numbers among mycohelminths, *Acrobeloides buetschlii* (52 specimens) and *Chiloplacus symmetricus* (22 specimens per 10 g of tissue) among saprobionts.

The abundance of *D. destructor* peaks at stage III of the the disease, as well as the number of mycohelminths (320 and 1540 per potato on average, 4 species), but the maximum number of saprobionts (439, 9 species) occurs at stage V of the disease. At the same time, the number of *D. destructor* is only 77 in one tuber.

Thus, the IV–V stages of ditylenchosis are accompanied by changing the number of phytonematode species and their abundance in favor of saprobionts and mycohelminths. The number of *D. destructor*, respectively, is 52–77 specimens per 10 g of tuber, and the number of mycohelminthes and saprobionts remains high and even increases (table 2). At these stages of the disease, the most prevalent species among mycohelminths are *Aphelenchoides limberi* — (14 affected tubers), *Aphelenchoides asterocaudatus* — 19 affected tubers out of 43, and saprobionts — *Caenorhabditis elegans*, *Chiloplacus symmetricus* (11 affected tubers), *Acrobeloides buetschlii* (14 tubers), *Rhabditis* sp., *Mesorhabditis monhystera* (17 tubers).

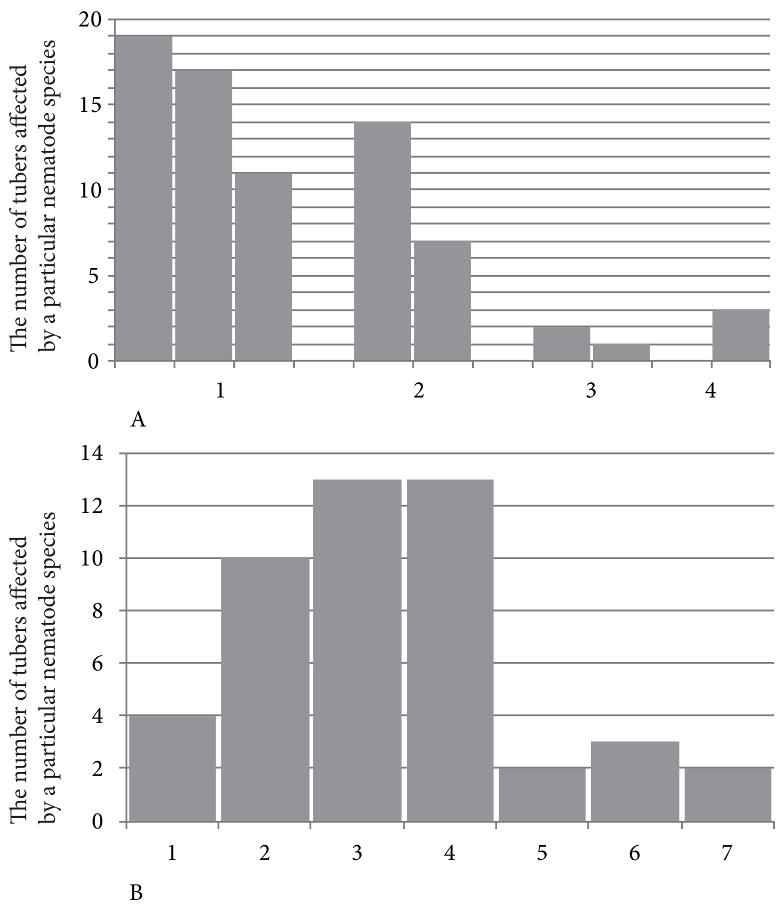

Fig. 6. Distribution of phytonematode infracommunities inhabiting potato tubers affected by *Ditylenchus destructor*: *A* — the occurrence of certain *Ditylenchus destructor* satellite species of phytonematodes in damaged tubers; *B* — the number of satellite species of phytonematodes in one damaged tuber.

As we can see, potato ditylenchosis is a complex microbiocenotic process, in which whole complexes of phytonematodes accompanying the species edificator *D. destructor* are involved. In affected tubers, the conditions for the existence of phytonematodes gradually create conditions for the existence of other species. Accordingly, the succession of the species composition of the nematode infracommunities occurs. At first, as a result of the vital activity of *D. destructor*, the healthy tissue of tubers is destroyed, subsequently it is colonized by fungi and bacteria and groups of nematodes that feed on them — mycohelminths and saprobionts. Later, conditions become unfavorable for the existence of *D. destructor*.

The material we analyzed clearly shows how the ratio between nematodes of various environmental groups changes in the degree of development of the disease. If at the first three stages of the disease parasitic nematodes (*Ditylenchus destructor*) comprised from 18.2 to 58.2 % of the nematode population, with the development of the disease they did not exceed 22.1–11.8 % of the nematode complex. As for mycohelminths and saprobionts,

Table 2. Dynamics of phytonematode abundance in tubers infected with *D. destructor* (2016–2017)

No	Species, ecotrophic groups	The number of nematodes in one tuber, specimens					The average number in one tuber, specimens
	Stage of the disease development	I	II	III	IV	V	
	Number of tubers	1	10	14	14	8	
	Phytonematodes						
1	*Ditylenchus destructor*	10	435	540	52	77	282
	Total	10	435	540	52	77	
	The proportion of phytonematodes in the total number of identified nematodes, %	18,2	84,3	58,2	22,1	11,8	
	Mycohelminths						
2	*Aphelenchoides asterocaudatus*	35	25	76	20		89
3	*Aphelenchoides bicaudatus*		1	15		1	71
4	*Aphelenchoides limbery*		5	226	94	130	397
5	*Aphelenchus avenae*		6	1	1	1	10
6	*Aglenchus agricola*	10	3			3	25
7	*Nothotylenchus* sp.			2	6		56
	Total	45	40	320	121	135	
	The proportion of mycohelminths in the total number of detected nematodes, %	81,8	7,6	34,5	51,5	20,7	
	Saprobionts						
8	*Caenorhabditis elegans*			1		361	1444
9	*Eucephalobus oxiuroides*			4		1	23
10	*Eucephalobus mucronatus*			1			2
11	*Chiloplacus simmetricus*		22	6	2	30	52
12	*Acrobeloides buetschlii*		18	52	1	31	82
13	*Diploscapter rizophilus*			1	1	1	3
14	*Cephalobus persegnis*					1	2
15	*Mesorhabditis monhystera*		1	1	9	1	9
16	*Rhabditis* sp.				49	12	257
17	*Pelodera teres*					1	8
18	*Panagrolaimus rigidus*			1			2
	Total	0	41	67	62	439	
	The proportion of saprobionts in the total number of detected nematodes, %	0	7,8	7,2	26,4	67,4	
	Total	55	516	927	235	651	

the process goes in the opposite direction. Their specific weight in the total mass of nematodes sharply increases starting from stage II and reaches a maximum at stages IV and V, when the number of mycohelminths is 51, 5, and 20.7 % of the total number of phytonematodes, respectively, and saprobionts, respectively, 26.4 and 67.4 %.

Among the mycohelminths, *A. asterocaudatus* (I–IV stage) and *A. limbery* and *A. avenae* (II –V stage) were more frequently recorded. *A. agricola* (stage III and V) and *A. bicaudatus* (stage II and III) were not so common. *Neotylenchus* sp. was found at the III-IV stage of the ditylenchosis. Among the saprobionts *Ch. symmetricus* (stage II–V) was usually recorded. *A. buetschlii* (stage II–IV), *M. monhystera* (stage II–IV), and *D. rizophilus* (stage III–V) were found quite frequently. All others were rare, they were only found in one or two stages of the process (fig. 5).

Infracommunity: species composition of phytonematodes in one affected potato, taxonomic and ecotrophic structure of the species complex.

In total, 47 potato tubers affected by *D. destructor* were selected (fig. 6, A). The number of nematode species and their abundance in each tuber were counted (table 2). The analysis of the obtained data showed that the distribution of infracommunities by the number of species that form a complex of nematodes in one tuber is close to normal.

Only *D. destructor* was found in only four tubers (8.51 %). It is likely that in these cases the penetration of nematodes into the potatoes has occurred recently. All other 43 tubers were inhabited by a phytonematode complex consisting of different species (from two to seven) (fig. 6, B).

In the vast majority of cases (36 tubers, 76.6 %), the average number of nematode species (2–4) was found. In particular, in ten (21.28 %) tubers, two species of *D. destructor* phytohelminth nematodes and one of the mycohelminth species (*Aphelenchoides asterocaudatus* or *Aphelenchoides limbery*) were found in 5 cases, or 5 cases of saprobionts from five different species, three or four species of phytonematodes were recorded in one tuber 13 times, together 26 cases 53.32 %. In three species infracommunities more often there was one species of mycohelminths and saprobionts (8 cases) or, respectively, two species of mycohelminths (3 cases) or saprobionts (2 cases). Saprobionts prevailed in four species infracommunities. The ratio of 1 species of mycohelminths to 2 species of saprobionts was found 7 times. Four more times only 3 species of saprobionts were found. Mycohelminths prevailed in only two cases (2 species to 1 species of saprobionts). Probably, obtained ratios of these two ecotrophic groups of nematodes are due to the stage of the ditylenchosis.

The largest in number of species infracommunities (5–7 species) were recorded less frequently in tubers (14.89 %). In five-species infracommunities, which were recorded twice, mycohelminths and saprobionts were represented equally, 2 species of each group. Six-species groups were found three times, saprobionts prevailed in them. The ratio of mycohelminths and saprobionts, respectively, was 1 to 4, 2 to 3, and 3 to 2 species. Also in the seven-species groups that were found twice, the ratio of mycohelminths and saprobionts, respectively, was 2 to 4 and 3 to 3 species. Two species of the genus *Aphelenchoides* were found together only 4 times, all three species — once. Both species of *Eucephalobus* were recorded together also once.

The number of each of the *D. destructor* phytonematode satellite species varies widely, as shown above, and changes substantially during the course of ditylenchosis. In general, the average number of mycohelminths in one tuber varies from 40–45 to 320 ind., while the number of saprobionts increases from 41 to 439 ind. (table 2). So, we can assume that the relative "mass" of these two *trophoecological* satellite groups of *D. destructor* is generally comparable. At the same time, the number of the phytohelminth edificator itself varies from 10 to 540 specimens at different stages of the disease.

Discussion

Potato rot nematode (*D. destructor*, Thorne, 1945) is close to the stem nematode (*D. dipsaci* Kühn, 1854), but its main host plant for is potato; it is settled mainly in tubers, as well as in stolons. The penetration of nematodes into young tubers can occur in two ways: from the stems of an infected parent tuber or through soil. Infected tubers are destroyed by the type of dry rot. In the diseased part of the tuber saprophytic fungi and bacteria penetrate, which are the foods for mycohelminths and saprobiotic nematodes (Kiryanova, Kral, 1971). Therefore, potato tubers are usually populated by a whole complex of nematodes. According to various authors, *D. destructor* satellite species belonging to four orders (Chromadorida, Enoplida, Rhabditida, and Tylenchida) and can be found in the potato tuber infected with this nematode.

The species composition of the nematode complex populated the lesions of the disease in potato tubers during winter storage was also studied in Ukraine. According to L. A. Kotyuk (1999) *Ditylenchus destructor* Thorne, 1945 and *Pratylenchus pratensis* (de Man, 1880) Filipjev, 1936 were found among the phytohelminths in potato tubers in Zhytomyr Region; *Aphelenchus avenae* Bastian, 1865 mainly occured among mycohelminths; *Panagrolaimus rigidus* (Schneider, 1886) Thorne, 1937, *Cephalobus persegnis* Bastian, 1865, *Rhabditis brevispina* (Claus, 1862) Buetschli, 1873 were among the saprobionts.

E. A. Butenko, (2004) notes that infection of potato tubers with ring rot and rhizoctonia leads to a significant decrease (by 88.7 %) in the number of the potato tuber nematodes. Among the identified species of nematodes *D. destructor* dominated among phytohelminths; *Plectus parietinus*, *Rhabditis brevispina*, *Pelodera teres*, *Pristionchus lheritieri*, *Panagrolaimus rigidus*, *Cephalobus persegnis*, *Eucephalobus oxiuroides*, *Chiloplacus soosi* and *Acrobeloides butschlii* dominated among saprobionts.

The species composition of the phytonematode complex in potato tubers affected by *D. destructor* was most fully and thoroughly analyzed in T. Zhilina's research work (2004). She identified 20 species of nematodes, which belong to 17 genera, 8 families and 2 orders. The nematode complex consisted of phytohelminths — 1 species (5 %), mycohelminths — 5 species (25 %) and saprobionts — 14 species (70 %). By Cassagnau's coefficient of species constancy (1961), the author considers the dominant species that are found in > 50 % of samples, frequent 5–50 %, rare < 5 %. *D. destructor* dominated among phytohelminths being found in 93.2% of tubers, *Mesodiplogaster lheritieri* Goodey, 1963, was found in 56.8 % of samples among saprobionts. The author includes 6 species of saprobionts (*Caenorhabditis elegans* (Maupas, 1900) Dougherty, 1953; *D. spengeliide* Man, 1912; *Acrobeloides buetschlii* (deMan, 1884) Steinerand Buhrer, 1933; *Pelodera teres* Schneider, 1866; Rurgensita, Rhabensitis 1914) and 3 species of mycohelminths (*Seinura oxura* (Paesler, 1957) Goodey, 1960; *Seinura demani* T. Goodey, 1928, JB Goodey, 1960; *Aphelenchoides bicaudatus* (Immamura, 1931) Filipjevet Sch. Stekhoven, 1941). The remaining 11 species were rare.

Under different potato storage conditions the species composition of phytonematodes and their numbers, as well as the proportion of tubers affected by *D. destructor* can differ significantly. In Ukraine, thirty-one species of phytonematodes were recorded in potato tubers, among them two species of phytohelminths, eight species of mycohelminths and twenty species of saprobiont nematodes. In addition to the previously found 18 species, 13 more species were recorded. Among them there are mycohelminths *Seinura oxura* (Paesler, 1957) and *S. demani* (Goodey, 1928), and saprobionts *Plectus parietinus* (Bastian, 1865), *Rhabditis brevispina* (Claus, 1862), *Diplogasteroides spengelii* (de Man, 1884), *Tridontus longicaudatus* 1965), *Demaniella ciburgensis* (Steiner, 1914), *Mesodiplogaster lheritieri* (Maures 1919), *Protorhabditis* sp., *Chiloplacus soosi* (Andrassy, 1953), *Pristionchus* sp. and *Cephalobus persegnis*

(Bastian, 1865). In potato tubers *Pratylenchus pratensis* (de Man, 1880) was also recorded along with phytonematode *D. destructor*.

Conclusion

In potato tubers affected by *D. destructor*, 18 nematode species belonging to 15 genera, 9 families, and 2 orders were identified. By number of species, the order Rhabditida, to which 4 families, 10 genera and 11 species belong, are most fully represented. The order Tylenchida includes 5 families, 5 genera, and 7 species of phytonematodes.

In general, the taxonomic structure of the phytonematode complex, which is formed in tubers affected by *D. destructor*, consists of five groups that differ in quantitative terms — the frequency of occurrence and the average number of infracommunities in one tuber. One species dominates — *D. destructor*. Species of phytonematodes, mycohelminths and saprobionts that are satellites of *D. destructor*, an edificator of microparasitocenosis of affected tubers, is divided into four groups. These are four subdominant species, accounting for 23.53 % of all associated species, three common species — 17.65 %, six rare species and four *accidental species*, total — 58.83 %. There are five stages in the course of ditylenchosis, depending on the appearance of the affected tubers and the nature of their population by different ecotrophic groups of phytonematodes: phytohelminths, mycohelminths and saprobionts. In the first three stages of the course of ditylenchosis, the number of *D. destructor* parasitic nematodes increases from 18.2 to 58.2 % of the total number of all phytonematodes, and subsequently their share at IV–V stages decreases to 22.1–11.2 %. Thus, it can be assumed that with the further development of the pathological process, the potato rot nematodes disappear and for a certain period of time only mycohelminths and saprobiont nematodes remain (conditionally the sixth stage); subsequently, the conditions in the affected tuber become unsuitable for the existence of fungi and, accordingly, mycohelminths. Saprobiont nematodes probably remain until complete destruction of the tuber (the seventh stage of the pathological process caused by *D. destructor*). The number of mycohelminthic peaks in the fourth stage, and further decreases significantly. The maximum number of saprobionts is at the fifth stage. Accordingly, the ratio of species number and the total number of nematodes of different trophic ecological groups decreases.

Some of *D. destructor* satellite nematode species are present in infracommunities of infected tuber for four of five stages of the process. They are all subdominants or common species, which are also characterized by a fairly high number of infrapopulations. There is reason to believe that these species are characterized by high ecological plasticity and can exist in a wide range of environmental conditions, soil and internal conditions of the affected potato.

In the course of the disease in a separate tuber, a regular succession occurs, the species composition of nematodes, the structure of the infracommunity, the ratio between the number of nematode species, and number of certain species change. Representatives of mycohelminths settle in potato affected by *D. destructor* already at the first stage of the disease, when the destruction of tuber tissue by *D. destructor* creates favorable conditions for the settlement of certain species of fungi. Saprobionts appear in the foci of affected tissue a little later, when some of the tuber parenchyma cells are destroyed and saprobiotic microorganisms join the fungi — the main food of saprobiont nematodes.

A potato can be considered as a separate self-sufficient organism, which represents a juvenile stage of development, which, when planted in the soil, will turn into a bush of potatoes, what corresponds to the sexual stage of plant development, that reproduces sexually, forming seeds. Thus, the tuber fully corresponds to the larval stage of development of invertebrates (caterpillars of butterflies, larvae of beetles, etc.), as well as vertebrates (tadpoles of amphibians, fish fry, etc.). So, the aggregate of specimens of a certain species of phytonematodes in one tuber can be considered as infrapopulation, and the complex of species of nematodes inhabiting one potato can be considered as infracommunity.

Among the 47 tubers selected for the study of *D. destructor* affected ones, only four were inhabited by one species of phytonematodes, namely this one. In the vast majority of tubers (43) phytonematode complex consisting of two or seven species was revealed. The distribution of all studied tubers by the number of nematode species in the infracommunity is close to normal. Most often, 3–4 species of nematodes were registered in the affected tubers. Tubers with two species of nematodes were less frequently recorded. The more diverse composition of the phytonematode complex (5–7 species in one tuber) was infrequently found.

In the tubers affected by *D. destructor*, the trophoecological structure of phytonematode infracommunity is variable; the ratio of the number of mycohelminths and saprobiotic species in one tuber may be different. Representatives of only one of these two trophoecological groups met infrequently. In the affected potato there can be several species (from two to four) of the same group, however, several phytonematodes from the same genus are rarely recorded together. The total number of specimens of all nematode species of one or another ecotrophic group in one potato also varies widely. However, the average aggregate "mass" of mycogelminths and saprobionts is comparable across all infracommunities.

Under different conditions of potato storage the species composition of phytonematodes and their number, as well as the proportion of tubers affected by the disease, may differ significantly.

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