



## PARASITIC NEMATODES IN POTATOES OF DIFFERENT VARIETIES AND THEIR INTERRELATIONS WITH SOME ARTHROPODS

Maria MELNIC<sup>1</sup>, Olesea GLIGA<sup>1</sup>, Dumitru ERHAN<sup>1</sup>, Stefan RUSU<sup>1</sup>, Elena IORDOSOPOL<sup>2</sup>

<sup>1</sup>Institute of Zoology, MECR, Chisinau, Republic of Moldova

<sup>2</sup>Institute of Genetics Physiology and Plants Protection, MECR, Chisinau, Republic of Moldova

Corresponding author: Olesea Gliga, e-mail: oleseagliga@gmail.com

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**Keywords:** potato, nematodes, arthropods, nematophagous, phytophagous.

**Introduction.** Potato (*Solanum tuberosum* L.) is susceptible to infestation with an associated complex of different species of parasitic and saprophytic nematodes, bacteria, fungi, and arthropods, which diminish the quality of the product.

**Material and methods.** 10 varieties of potatoes were investigated (*Agata*, *Albăstriu-mov*, *Bella rosa*, *Concorde*, *Desiree*, *Irga*, *Iagodca*, *Roko*, *Romano*, *Sprinter*), cultivated on the territory of the Republic of Moldova. The extraction of nematodes and mites was performed using the Baermann funnels, modified by Nesterov.

**Results.** Multiannual researches on the degree of potato infestation have shown that tubers are preferred by various arthropods (*Acaridae*), *Agriothes* spp. (*Elateridae*), *Gryllotalpa gryllotalpa*, (*Grillotalpidae*) and *Sciaridae* spp. (*Sciaridae*), which form different interactions with the parasitic nematodes of tubers (*Ditylenchus destructor*, *D. dipsaci*).

**Conclusions.** *Solanum tuberosum* infested by parasitic nematodes *D. destructor* in association with saprophytic nematodes and dry rot are colonized by nematophagous (mites – 80%) and phytophagous arthropods (wireworms – 40%; mole cricket – 50%; flies – 40%). Among the researched arthropods, *Rhizoglyphus echinopus* were more frequently found, which together with other species actively consume the primary and secondary parasitic nematodes, their mortality constituting up to 90%. In the traumatized by some arthropods potatoes, with the soil, secondary parasitic nematodes, also penetrate, carrying bacterial and fungal infections, subsequently stimulating the total rot of potato tubers.

**Cuvinte cheie:** cartof, nematode, artropode, nematofage, fitofage.

**NEMATODELE PARAZITE LA CARTOFII DE DIFERITE SOIURI ȘI INTERRELAȚIILE CU UNELE ARTROPODE**

**Introducere.** Cartoful (*Solanum tuberosum* L) este susceptibil la infestare cu un complex asociat de diferite specii de nematode parazite, saprofite, bacterii, ciuperci și artropode, ceea ce îi diminuează din calitatea lui ca produs.

**Material și metode.** Au fost cercetate 10 soiuri de cartofi (*Agata*, *Albăstriu-mov*, *Bella rosa*, *Concorde*, *Desiree*, *Irga*, *Iagodka*, *Rocko*, *Romano*, *Sprinter*) cultivate în Republica Moldova. Extragerea nematodelor și a acarienilor s-a efectuat cu utilizarea pâlniilor Baermann, modificate de Nesterov.

**Rezultate.** Cercetările multianuale, privind gradul de infestare a cartofilor, au demonstrat că tuberculii sunt preferați de diferite artropode: *Agriotes* spp. (*Elateridae*), *Gryllotalpa gryllotalpa*, (*Grillotalpidae*) și *Sciaridae* spp. (*Sciaridae*), care stabilesc interrelații diverse cu nematodele parazite (*Ditylenchus destructor*, *D. dipsaci*) ale culturii.

**Concluzii.** *Solanum tuberosum*, infestat de nematodul parazit *Ditylenchus destructor*, în asociere cu nematodele saprofite și cu putregaiul uscat, este colonizat de artropodele nematofage (acarieni – 80%) și fitofage (viermii-sârmă – 40%; coropișnițe – 50%; musculițe – 40%). Printre artropodele cercetate, mai frecvent a fost atestat *Rhizoglyphus echinopus* (Fumouze & Robin, 1868), care, împreună cu alte specii, consumă activ nematodele parazite primare și pe cele secundare, mortalitatea acestora constituind până la 90%. În cartofii traumați de unele artropode, odată cu solul, pătrund și nematodele parazite secundare, purtătoare ale infecțiilor bacteriene și fungice, stimulând ulterior putrefacția totală a tuberculilor.

## INTRODUCTION

Potato (*Solanum tuberosum* L.) is the third most important food crop plant of the world, producing high yields of nutritionally valuable food in the form of tubers, which has captured the attention of many researchers. Because the tubers are susceptible to infestation with an associated complex of different species of parasitic and saprophytic nematodes, bacteria, fungi, and arthropods, it is important to know their impact on the host organism. Primary parasitic nematode *Ditylenchus destructor* (Thorne, 1945) creates pathways in tubers for secondary parasitic invertebrates such as: saprophytic nematodes, bacterial and fungal infections, these being vectors of infections for susceptible potato varieties. As a result, the resistance of potato crop is diminished and their complete putrefaction occurs (1).

The nematode *D. destructor* is widespread in all countries where potatoes are growing. For example, in Romania, in the process of studying the level of infestation with the nematode *D. destructor* and other pathogens, which cause diseases of the seed potato of 4 varieties – Desiree, Ostara, Dahlia, Alka, it has been established that the most susceptible to both the *D. destructor* and the pathogenic species of microorganisms are the varieties Desiree and Ostara (2). The frequency of the attack with *D. destructor* was 44.8% and 27.38%, respectively, and the Dahlia and Alka varieties were more resistant, the frequency of the attack constituted 5.26% and 4.10%, respectively. In the Republic of Belarus, in all agro-climatic zones, there have been cases of crop loss of up to 43%, due to infestation with the *D. destructor* nematode of food and seed potatoes (3).

In phases 1, 2 of ditylenchosis the primary monotypic population of tubers is infested only with the population of a single species of nematode – *D. destructor* (females, males, larvae of different ages) (4). In such tubers, parasitic nematodes do not cause obvious external symptoms of ditylenchosis and only by removing the shell, small stains on the surface of the tuber pulp appear low visible. In phases 4-5 of ditylenchosis, the tissue invaded by different secondary invertebrate organisms becomes evidently rotten (5). Ditylenchosis disease of tubers initially manifests by cracking the shell, similar to dry rot, caused by *Fusarium* spp.

The purpose of the research was the study of the

species of nematophagous and phytophagous arthropods, as well as the complex of infections associated with potato tubers of 10 varieties, during the autumn-winter-spring periods.

## MATERIAL AND METHODS

The researches were carried out during 2014-2019 years. The research was conducted on 10 varieties of potatoes (Agata, Albastru-mov, Bella rosa, Concorde, Desiree, Irga, Iagodca, Roko, Romano, Sprinter), which were cultivated in the north, south and central areas of the Republic of Moldova. For each variety, 15-20 potatoes tubers per sample were researched. The primary and secondary parasites of tubers have been determined in advanced phases of ditylenchosis. The extraction of nematodes and mites was performed using the Baermann funnels, modified by Nesterov (6). The density of nematodes extracted from the tubers affected by ditylenchosis was calculated by applying the De Grisse chamber. The fixed preparations were carried out according to the method proposed by Seinhorst (7). The stereoscopic microscope was used to establish morpho-physiological changes in the infested potatoes.

Statistical analysis was performed using MS Excel program.

## RESULTS

### *Nematodes associated with rot*

It was determined that all the potatoes varieties collected during the autumn-winter-spring periods were infested with parasitic nematodes *Ditylenchus destructor* (advanced phases of ditylenchosis, 4-5) in combination with *D. dipsaci*, saprophytic nematodes, bacteria and fungi (fig. 1).

The results of the research showed a wide spread (100%) of the obligate primary parasite *D. destructor* in all investigated 10 potato varieties. The varieties of Albastru-mov, Desiree and Irga differed in frequency and density of infestation. The most frequent (100%) *D. destructor* is associated with dry rot caused by *Fusarium* spp. and less frequent (60%) with wet rot caused by *Erwinia* spp.

### *Nematophagous arthropods – mites*

It was established that the studied potatoes tubers (Agata, Albastru-mov, Bella rosa, Concorde, Desiree, Irga, Iagodca, Roko, Romano, Sprinter) infested with parasitic nematodes *D. destructor*,

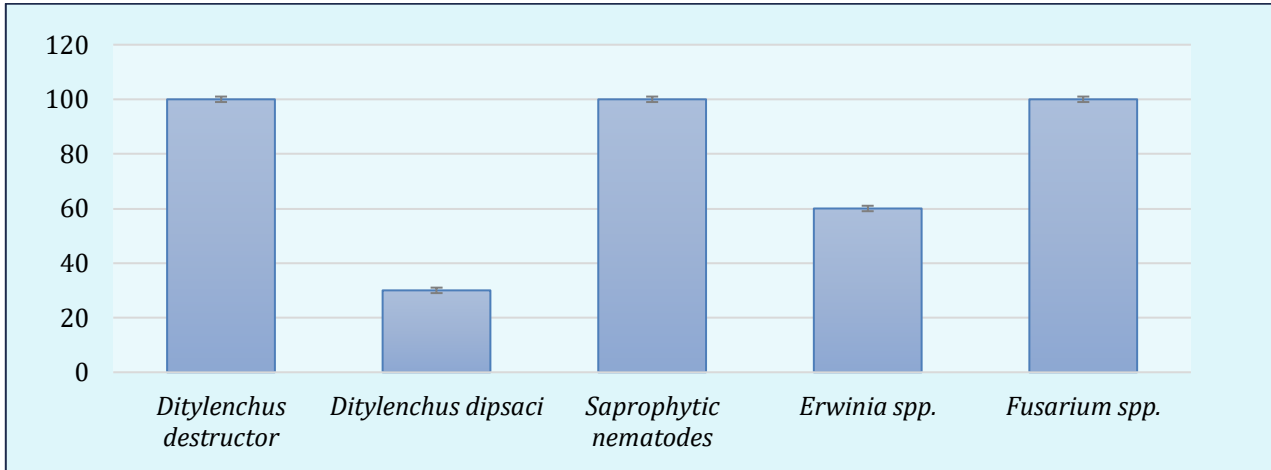


Figure 1. Frequency of parasitic and saprophytic nematodes, of dry and wet rot (%) in the investigated potato varieties.

*D. dipsaci*, saprophytic nematodes, bacteria and fungi, were also colonized by different species of mites (fam. Acaridae), which have been commonly found in all the researched varieties (fig. 1, 2).

It should be mentioned the fact that the potato tu

bers infested by *D. destructor* in association with saprophytic nematodes and dry rot were colonized by nematophagous (frequency 80%, different species of mites) and phytophagous arthropods (wireworms – 40%; mole cricket – 50%; flies – 40%) (fig. 2).

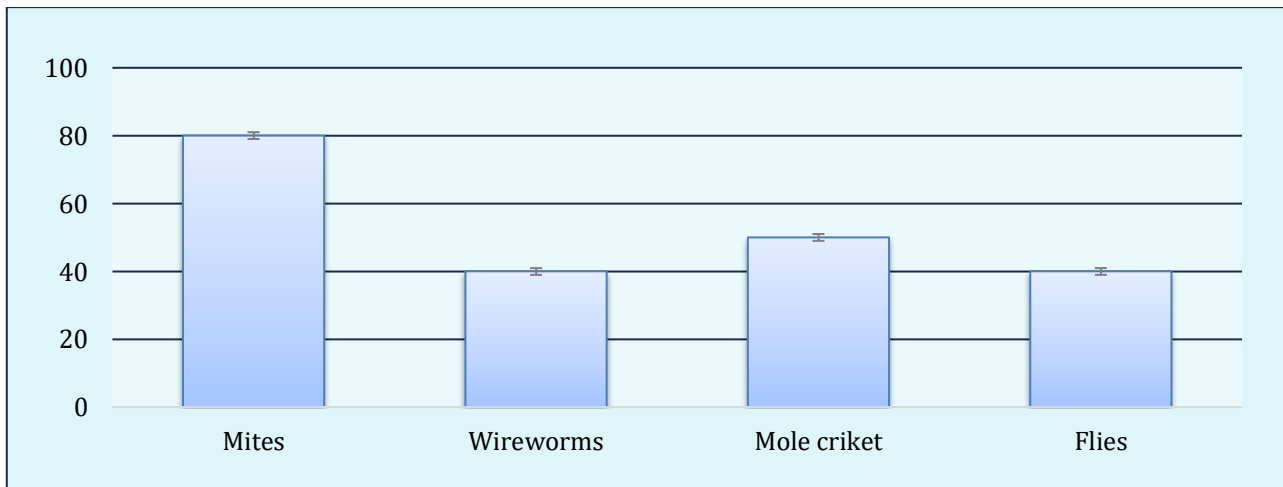


Figure 2. Frequency of nematophagous and phytophagous arthropods (%) in the investigated potato varieties.

An increased density of mites (hundreds of individuals along with their eggs/potato tubers) was detected in 5 of the investigated varieties - Agata, Bella rosa, Desiree, Irga, Roko. In the study process, for the first time, cases of active mite's attack on primary (*D. destructor*, *D. dipsaci*) and secondary (facultative, most of them of the order Rhabditida) parasitic nematodes have been observed. The latter (females, males, larvae, eggs) are devoured and consumed by mites (fig. 3). Under such conditions, the mites lay eggs, from which

the larval forms develop, which then leads to the post-embryonic development cycle.

All mite species present in potatoes tubers infested with nematodes and microorganisms proved to be active predators. It has been established that mites attack nematodes (from families: Anguinidae, Cephalobidae, Neodiplogasteridae, Rhabditidae etc.) usually, in the region of median intestine, along the reproductive tract (ovary, oviduct in this area the holes are obser-

ved, through which the content of the organs is absorbed, first of all the gonad with the nematode eggs (fig. 4 (3a, 3b)). High mortality up to 90%

both of primary and secondary parasitic nematodes has often been observed in such suspension (fig. 4 (1b)).

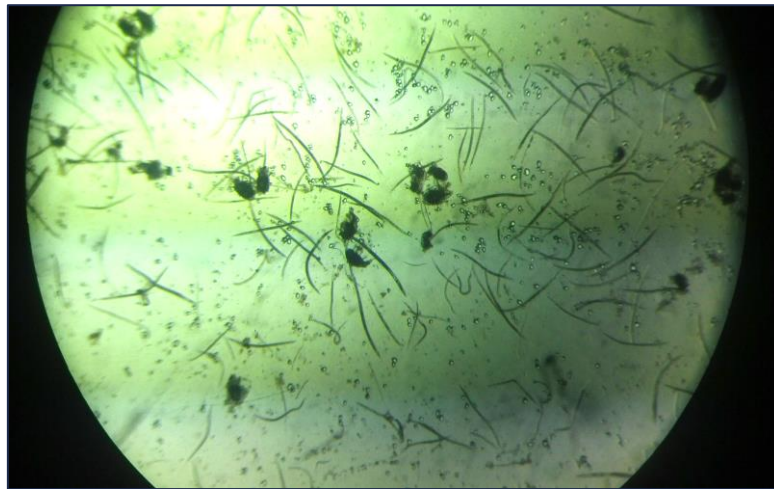


Figure 3. The nutritional process of mites with parasitic and saprophytic nematodes, which were extracted from infested potatoes by *D. destructor*, *D. dipsaci*, in advanced phases of ditylenchosis.

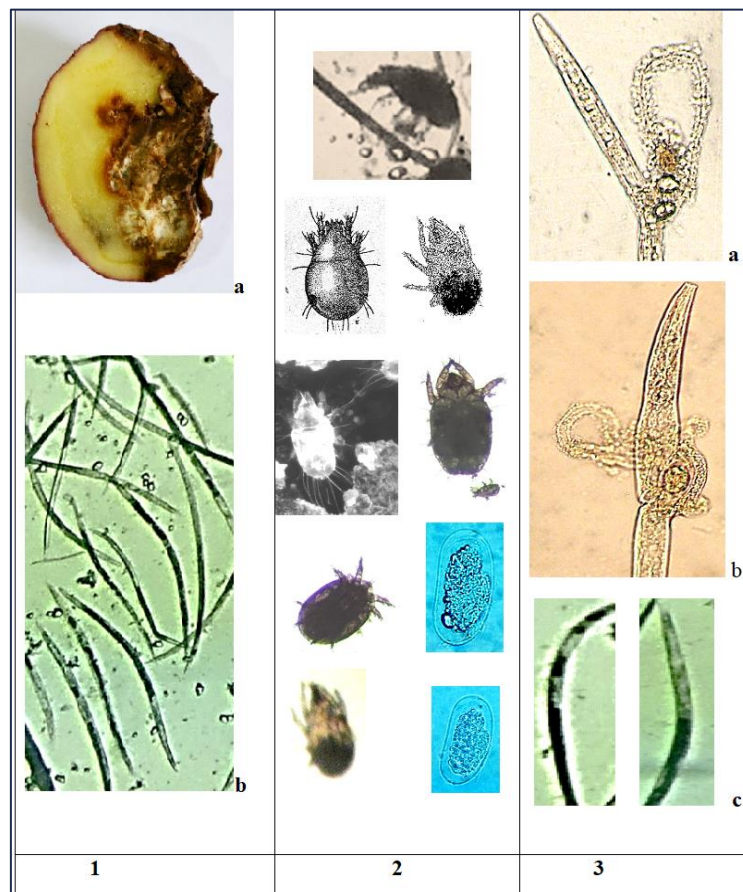


Figure 4. 1a - Potato tuber (section) infested by *D. destructor* in association with secondary parasites (saprophytic nematodes, microorganisms, nematophagous mites); 1b - nematodes *D. destructor*, *D. dipsaci*, devoured by mites; 2 - species of nematophagous mites and their eggs; 3 - different species of dead nematodes after being in contact with mites (a, b - saprophytes; c - devoured portions of *D. destructor*).



There have been observed cases, when in the infested potatoes, in which the density of mites accounted for hundreds of mature individuals/ tuber, all species of nematodes disappeared, as well as the bacterial and fungal infections. As a result, portions of the tuber with completely macerated substrate remained, containing only empty cells. Also, the starch granules disappeared in these phases of ditylenchosis. In such tubers, the passing from the infested to the non-infested portion and the extension of nematological, bacterial and fungal infections was not observed. Only a clear differentiation of not infested and infested portion of potatoes was observed, which demonstrates the cessation of parasitic, fungal and bacterial infection.

Mites were detected in the investigated potato tuber varieties (a significant number), which belong to the species of *Rhizoglyphus echinopus* F. & R., family Tyroglyphidae (fig. 4.2). It is a widespread phytophagous species, with a 0.4-0.5 mm body length.

Loads of eggs deposited by mites in the potatoes tubers was also observed. A female lays about 800 eggs on average. A generation might develop over

30 days. Their development occurs at a humidity of more than 60%, being very sensitive to this factor. It should be mentioned, that the tubers contain an increased amount of water, depending on the potato variety (between 77% - variety Albastriu-mov and 83% - Agata variety), but in the infested potatoes by primary parasites the amount of water increased with 5-6%, creating favourable conditions for existence not only for mites, but also for bacteria, fungi and saprophytic nematodes.

**Phytophagous arthropods**

Wireworms of click beetles (family Elateridae). The research results showed that potato harvest at some of the analysed varieties (Agata, Bella rosa, Desiree, Roko, Sprinter, Concorde), originating from field culture, has been quite affected by some harmful phytophagous arthropods, such as wireworms of the click beetles (fig. 5b, 5c). Wireworms depreciate qualitatively the potato tubers due to the galleries, which are dug into the pulp. During the laboratory studies of the affected portions, it was observed that soil penetrated into those galleries (fig. 5a) made by the wireworms, thus favouring the colonization with bacterivore saprophytic nematode species.



Figure 5. Potatoes attacked by wireworms of click beetles in association with saprophytic nematodes and mites (a; b) (c - wireworms, exterior aspect).

The laboratory investigation of the soil extracted from pulp galleries of the affected tubers demonstrated the presence of nematodes that eliminate bacterial and fungal infections of the order Rhabditida, families: Rhabditidae, Cephalobidae and order Dorylaimida, family Dorylaimidae (sp. *Mesodorylaimus bastiani*). In addition, crowds of mites were detected in the tested soil.

**Mole cricket (*Gryllotalpa gryllotalpa* L.)**

Among the phytophagous arthropods in some of the investigated varieties (Agata, Desiree, Irga, Roko, Iagodca), the presence of the common mole cricket (*G. gryllotalpa*, family Gryllotalpidae) has been established (fig. 6c). The observations made on individual lots in *Roko* potatoes variety determined 100% of mole cricket attack (fig. 6a, 6b). Soil penetrates the mechanically traumatized

tubers by mole cricket, in which the bacterial saprophytic nematodes were frequent found, which brought microorganisms on the body or intestine

and cause both dry rot (*Fusarium* spp.) and wet rot (*Erwinia* spp.).

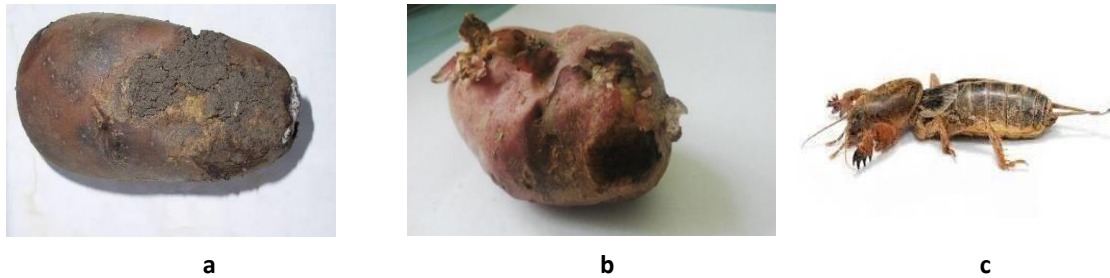


Figure 6. a, b – Potatoes tubers infested by *D. destructor* (phases 3, 4 of ditylenchosis) in association with secondary parasites and pests (with cavities formed by mole cricket); c – *G. grillotalpa* L. (exterior aspect).

Under laboratory conditions, most of the harvested tubers, affected by mole cricket bites, over a storage interval of 1.5-2.0 months at room temperature, turned into waste caused by the rot (fig.

7c). The insects produced huge cavities (fig. 7b) in the potato tubers, causing qualitative and quantitative deterioration of the potato crops, thus being transformed into waste.

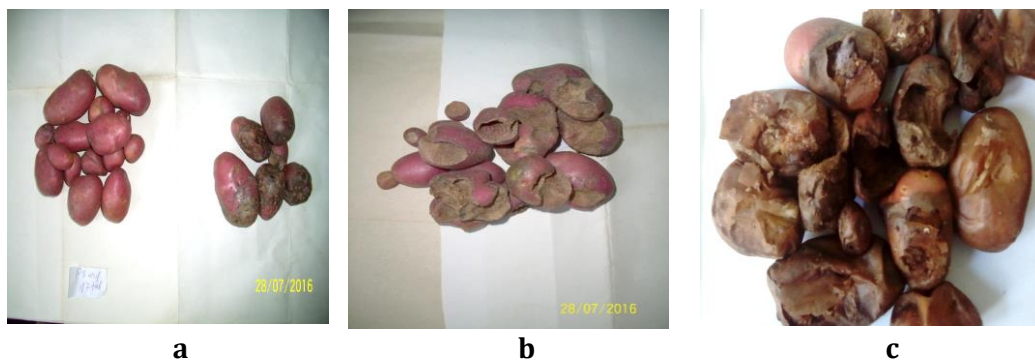


Figure 7. Roko variety potatoes (from field experiments, obtained from a mother tuber): a – healthy potatoes; b – traumatized by mole cricket; c – traumatized by mole cricket after being kept in laboratory conditions for over 2-3 months at room temperature – 100% infestation of wet rot.

### Flies (Sciaridae spp.)

In the researches carried out on 10 varieties of potatoes, flies were detected only in some of them such as Agata, Deziree, Iagodca, Roko, which were selected from the field crops, with primary and secondary parasites and some pests. The affected tubers are characterized by perforated surfaces, whereas the flies' larvae were frequently detected in the damaged tissue (fig. 8c). It has been determined that these belong to the Sciaridae family.

It was determined that the tubers associated with Sciaridae spp. occur in the advanced stages of ditylenchosis, phases 4-5, when these are in the process of decomposition. A series of parasites and pests might colonize such tubers, such as phytoparasitic nematodes, saprophytes, microorganisms and mites. Sciaridae spp. usually lives in

wet soil, attacks plant roots and feeds with their living tissue.

### DISCUSSIONS

In the conditions of the Republic of Moldova, a wide spread (100% frequency) of tubers obligatory primary parasite – *D. destructor* in all 10 researched varieties was observed. Researches has shown an association of parasitic nematodes *D. destructor* and *D. dipsaci* (frequency 30%) only in the varieties Albastrui-mov, Bella rosa, Desiree. In our opinion, this association displays a local character. Nematode *D. destructor* is frequently (100%) accompanied by secondary parasitic nematodes (saprophytes), most being of the

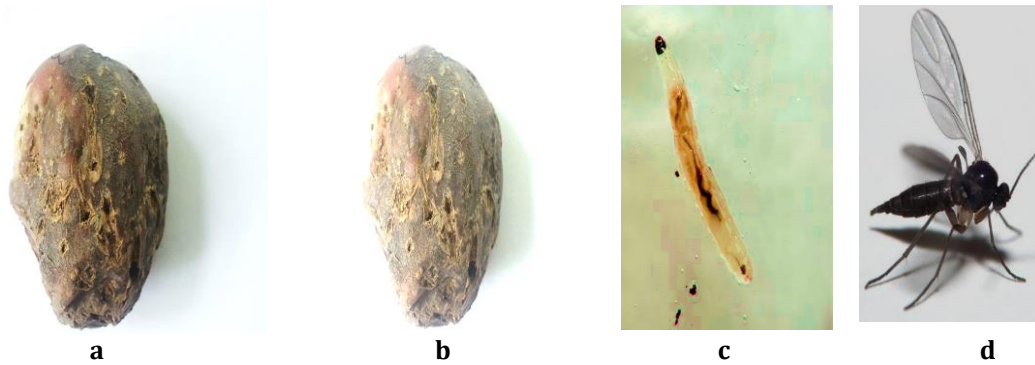


Figure 8. a – Roko variety potatoes infested by *D. destructor* in association with saprophytic nematodes, microorganisms, mites, flies; b – holes in the pulp of the tuber through which *Sciaridae* spp. penetrated; c – larva; d – imago.

order Rabdithida. A frequent association (100%) of the nematode *D. destructor* with dry rot caused by *Fusarium* spp. was determined. The wet rot *Erwinia* spp was found less frequently (60%) in the investigated varieties. It was observed that both primary parasitic nematodes and secondary saprophytic nematodes are not associated with wet rot and commonly leave the environment.

Our research reported an 80% frequency of mites, namely in the varieties of Agata, Albastriumov, Bella rosa, Concorde, Desiree, Irga, Iagodca, Roko. It has been determined that mites are common in tubercles infested with *D. destructor* in association with saprophytic nematodes and dry rot. Also, it was observed cases of active attack of mites on parasitic and saprophytic nematodes. Similar researches on nematode-mite interrelationships have been followed by other authors (8), who observed some processes regarding the perforation and nutrition of mites of the species *Tyrophagus putrescentiae* with the nematodes of the species *Rhabditis* spp., *Cephalobus* spp., *Heterodera orizae*. According to some researches (9) special importance is given to the mites of the genus *Caloglyphus* (Acarina: Acaridae), which shows a high efficacy in decreasing the density of parasitic nematodes. All stages of these mites attack the egg mass, laid by females, juvenile forms and females of the species *Meloidogyne*, *Globodera rostochiensis* and *Heterodera schachtii*, as well as endo- and ectoparasitic migratory species: *Paratylenchus* spp., *Pratylenchus* spp., *Tylenchorhynchus* spp. It has been established that the species *Hypoaspis sculifer* (family Gamasidae) attack the phytophagous nematode *Tylenchorhynchus dubius*, reducing their populations up to 68% (10). The extensive researches of some authors (11, 12,

13) determined that the mites of the species *T. putrescentiae* and *Hypoaspis calcuttaensis* are both nematophagous, consuming invasive larvae of the species *Meloidogyne javanica* and consumers of harmful fungi of plants: *Fusarium*, *Alternaria*, *Mucor* etc. Researchers from Florida (Orlando) (14) observed that the mite *Coleoscurus simplex* (Ewing) (Cunaxidae: Coleoscurinae) colonizes greenhouse pot cultures of root knot nematodes (*Meloidogyne* spp.), where it preys on vermiform nematodes and soil arthropods. This is the first report of nematophagy in a cunaxid mite. Previous researches (15) have shown that potatoes tubers infested with *D. destructor* (phases 3-4-5 of ditylenchosis) are always colonized by various species of nematophagous mites from the family Acaridae, more common being individuals of the species *Rhizoglyphus echinopus*. The phytophagous mite *R. echinopus*, also called bulb mite, infests both field plants and those stored in the warehouses (10, 14). They attack nematodes in the region of the gonad and oviduct with eggs that they consume actively, causing the mortality of both parasitic and saprophytic forms. Some species of mites (Gamasidae sp.) devour and use for food the parasitic species *D. dipsaci* and *D. destructor*, the daily norm being up to four small nematodes. At the same time, the presence of mites is important in regulating the density of nematodes under natural conditions (14). In our researches, all mite species present in potatoes tubers infested with nematodes and microorganisms proved to be active predators of phytophagous and saprophytic nematodes.

The harvested tubers are attacked by some phytophagous arthropods, such as wireworms of click beetles (fam. Elateridae), mole cricket



(family Grilloalpidae) and flies (fam. Sciaridae).

The research results showed that 40% of the analysed varieties (Agata, Concorde, Roko, Sprinter) were attacked by the wireworms of click beetles. According to the experimental data of some authors (16), among the click beetles, the species of the genus *Agriotes* (family Elateridae) are widely spread in the Republic of Moldova. The presence of 6-8 wireworms per 1m<sup>2</sup> causes tuber damages up to 60%, and the installation of various pathogens in the galleries left, favours the rot of tubers, leading to crop losses of up to 50% (17). In the process of laboratory investigations of the affected parts, it was observed that soil penetrates into the galleries through the pulp, which favours the colonization with bacterivore saprophytic nematode species of the order Rhabditida, these being active decimators of microbial infections. Similar results were obtained by other researchers (18), where potato tubers affected by wireworms are often subsequently infested by pathogenic bacteria and fungi, which cause dry and wet rot.

In monoculture conditions, the mole cricket (family Grilloalpidae) attack (frequency 50%) was also observed namely on the varieties Albastruimov, Dessiree, Irga, Roko, Sprinter. It is a

widespread polyphagous species in Europe, as well as in the Republic of Moldova, especially in conditions of increased humidity or on irrigated lots. In the superficial horizon of the soil (0-20cm) a single female lays up to 500 eggs (19). Like the wireworm, the mole cricket cause tuber damage through mechanical damage, with the possibility of penetrating the soil with bacterivorous saprophytic nematodes, accompanied by various microorganisms, which stimulate the appearance of rot (*Fusarium* spp., *Erwinia* spp.) that over a period of storage turn the tubers into waste.

In the potato tubers, cultivated in monoculture condition, the presence of Sciaridae spp. (frequency 40%) was detected in the varieties Agata, Deziree, Iagodca, Roko infested by the nematode *D. destructor* in combination with the saprophytic nematodes and microorganisms, in the 4-5 phases of ditylenchosis. These could be transmitters of both *D. destructor* nematodes (larvae, eggs) and various microbial infections. According to some researches (20), eggs and larvae of the nematode *D. dipsaci* were detected in the intestines of flies from the Drosophilidae family. According to the authors, the transmission of parasitic nematodes from one deposit to another might occur due to the flies.

## CONCLUSIONS

1. *Solanum tuberosum* infested by *Ditylenchus destructor* in association with saprophytic nematodes and dry rot are colonized by nematophagous (frequency 80%, different species of mites) and phytophagous arthropods (wireworms - 40%; mole cricket - 50%; flies - 40%).
2. Potatoes tubers are preferred by different arthropods (Acaridae), *Agriotes* spp. (Elateridae), *G. gryllotalpa* (Grilloalpidae), Sciaridae spp. (Sciaridae), which form different interactions with the parasitic nematodes of tubers. Among the studied arthropods, *Rhizoglyphus echinopus* (Fumouze & Robin, 1868) were more frequently found, which together with other species (*Agriotes* spp. (Elateridae), *G. gryllotalpa* (Grilloalpidae), Sciaridae spp. (Sciaridae)), actively consume the primary (*D. destructor*, *D. dipsaci*) and secondary parasitic nematodes (saprophytes, most being of the order Rhabditida), their mortality accounting for up to 90%.
3. The Sciaridae spp. might carry the eggs of primary parasitic nematode species in potatoes from warehouses. Some phytophagous arthropods might damage potatoes, thus the secondary parasitic nematodes from the soil might penetrate and bring bacterial and fungal infections, subsequently stimulating the total rot of potatoes tubers.

## CONFLICT OF INTERESTS

The author does not declare any conflict of interest.

## ACKNOWLEDGMENT

The research was carried out within the project: 20.80009.7007.12 Diversity of hematophagous arthropods, zoo and phytohelminths, vulnerability, climate tolerance strategies and development of innovative procedures for integrated control of species of socio-economic interest.



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Maria MELNIC, ORCID ID: 0000-0002-5263-8845  
 Olesea GLIGA, ORCID ID: 0000-0002-4917-5106  
 Dumitru ERHAN, ORCID ID: 0000-0001-9722-4382  
 Stefan RUSU, ORCID ID: 0000-0002-3322-9173