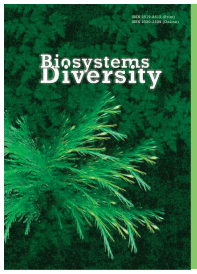

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Nematocidial activity of aqueous solutions of plants of the families Cupressaceae, Rosaceae, Asteraceae, Fabaceae, Cannabaceae and Apiaceae

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In natural ecosystems of animals, introduction of larvae of parasitic nematodes into the litter layer from excrement facilitates their migration and search of new host vertebrate animals. In such conditions they are constantly affected by biologically active substances of the abundant species of plants which grow in pastures. Currently, the influence of substances present in the above-ground part of plants on the vitality of larvae of helminths in the environment remains unstudied. In this article, we present the results of our research on the nematocidial activity *in vitro* in the aqueous solutions of 21 species of plants distributed in the territory of Steppe Ukraine: *Sanguisorba officinalis* L., *Rosa canina* L., *Crataegus sanguinea* Pall., *Crataegus pentagyna* Waldst. & Kit. ex Willd., *Armeniaca vulgaris* Lam., *Taraxacum officinale* F. H. Wigg., *Iva xanthiifolia* Nutt., *Artemisia campestris* L., *Arctium minus* (Hill) Bernh., *Ambrosia artemisiifolia* L., *Cannabis sativa* L., *Humulus lupulus* L., *Melilotus officinalis* (L.) Pall., *Vicia cracca* L., *Lotus ucrainicus* Klok., *Onobrychis arenaria* (Kit.) DC., *Foeniculum vulgare* Mill., *Eryngium planum* L., *Conium maculatum* L., *Juniperus communis* L., *Thuja occidentalis* L. The level of vitality of nematode larvae of the Strongylida (*Haemonchus contortus* (Rudolphi, 1803)) and Rhabditida (*Strongyloides papillosus* (Wedl, 1856)) orders varied depending on the species of plant, and also five experimental concentrations in aqueous solutions tested in seven replications. The most notable nematocidial effect was exerted by *Taraxacum officinale* F. H. Wigg. – we observed death of larvae of third stage development L_3 *H. contortus* and larvae of first-third stages L_{1-3} *S. papillosus* at 24 h exposure to 3% aqueous solution. Three percent aqueous solutions of *S. officinalis* and *A. artemisiifolia* displayed nematocidial properties only against *S. papillosus*: death of L_{1-3} *S. papillosus* was observed. Aqueous solutions of *R. canina*, *A. vulgaris*, *A. minus*, *H. lupulus*, *V. cracca*, *L. ucrainicus*, *O. arenaria*, *E. planum*, *C. maculatum*, *J. communis*, *Th. occidentalis* had lethal effect only on non-invasive larvae (larvae of the first and second stage L_{1-2}) of *S. papillosus*. They displayed no nematocidial properties towards invasive larvae of *H. contortus* and *S. papillosus*. At exposure to aqueous solutions of the rest of the studied species of plants, over 50% of L_3 *H. contortus* and L_{1-3} *S. papillosus* larvae remained alive. The determined patterns allow us to state that while living in the litter and soil in the root zone of plants nematode larvae undergoa negative influence caused by some plant species.

Keywords: *Strongyloides papillosus*; *Haemonchus contortus*; aqueous solution of plants; nematocidial properties; parasitocenosis.

Introduction

Species of nematodes of the Strongylida and Rhabditida orders parasitize many domestic and wild mammals (Boyko et al., 2016; Boyko & Brygadyrenko, 2019). One of the commonest representatives of the Strongylida order is *H. contortus* (Boyko et al., 2019). Rhabditida pathogens of nematodiases in mammals are most often represented by *Strongyloides papillosus* (Wedl, 1856). Helminthiases cause economic losses to livestock farms every year (Peter et al., 2015; Stachurska-Hagen et al., 2016; Thamsborg et al., 2017; Zazharska et al., 2018). *S. papillosus* is a nematode which affects the gastrointestinal tract and breathing organs (at the larva stage). During the experimental infestation of rabbits, Nakamura et al. (1994) observed an up to 44% decrease in the body weight with following exhaustion of the studied laboratory animals. Kváč et al. (2007) recorded lethal cases of 25% of the calves one a farm in a mountain area in the Czech Republic as a result of parasitization by *S. papillosus*. The autopsy revealed that all the dead animals had pathological changes in the lungs due to migration of the larvae. The age peculiarities of susceptibility to this species of nematode were also indicated by Wymann et al. (2008). Parasitization by helminths equalled 39% in cattle aged up to 1 month, 59% for cattle aged up to 2–3 months – and 42% for cattle aged up to 5–6 months. The presence of *Strongyloides* spp. in the organism of mammals is followed by decrease in erythrocytes. At the same time, the lowest parameters were observed in the animals with high infestation intensity, and the number of neutrophils and eosinophils increased to 44.2 ± 2.5% and 13.3 ± 0.6% respectively with high intensity of strongyloidiasis of animals (Dimitrijević et al., 2016).

One of the main factors of *S. papillosus* vitality in the external environment is the level of moisture (Nath, 1978; Taira et al., 1991; Jacquiet et al., 1992; Fritsche et al., 1993; Ndao et al., 1995; Gruner et al., 1998; Chompoochan et al., 1998). The study of the impact of this parameter remains relevant nowadays (Sissay et al., 2007; Das et al., 2017). According to Nath (1978), the survival of eggs and larvae of *S. papillosus* in the external environment depends on moisture. The eggs do not develop below 87% relative moisture and at the temperature of 25–30 °C. The larvae of this nematode species are not resistant to drying. The data of Taira et al. (1991) also indicate the distribution of helminths of this species in livestock on Guadeloupe in the humid tropical zone. At the same time, these authors determined that *S. papillosus* dominated in the age group of cattle up to two months with subsequent decrease in the animals' growth. The distribution of *S. papillosus* and other nematodes of the gastro-intestinal tract of sheep and goats in South-West Mauritania (Trarza) equaled less than 20%. Perhaps, such low parameters also indicate the climatic peculiarities of desert areas. The researchers presume that the Ruminantia become infested during the wet season (Jac-

quiet et al., 1992). That a high level of moisture in the substrate provided favourable environmental conditions for the development of *S. papillosus* nematodes was established in the research by Fritsche et al. (1993), and also by Ndao et al. (1995), conducted in Gambia. Fritsche et al. (1993) recorded 55% of sheep and goats as hosting *S. papillosus*. Their number peaked between July and October (also during the wet season). Over the dry seasons, their number in Ruminantia decreased. The studies by Pandey et al. (1994) in Zimbabwe indicate similar results for the relatively favourable impact of heightened moisture during the wet season on the development of *S. papillosus* in sheep and goats. A high number of *Strongyloides* eggs in the feces of goats was also recorded during the wet season with their consequent decrease over the period with a low level of moisture. For analysing the severity of strongyloidiasis many researchers conduct experimental infestation of animals. Taira et al. (1991) during a laboratory experiment infecting rabbits with a culture of *S. papillosus* from calves observed the dynamic of evacuation of the eggs of this species in the feces. Kobayashi et al. (2009) successfully conducted a surgical implantation of mature *S. papillosus* into the duodenum of Mongolian gerbils (*Meriones unguiculatus* Milne-Edwards, 1867). This experiment became a good model for the following laboratory studies on the disorders in the functioning of gastrointestinal tract of mammals parasitized by *S. papillosus* (Kobayashi et al., 2009). Kobayashi & Horii (2008) conducted an experimental *S. papillosus* infection of rabbits. The objective of their research was to study the lethal mechanism of the impact of helminths on the gastrointestinal tract. During the experiment, they observed significant decrease in the body weight, consumption of food and mass of feces. At the same time, they recorded disorders in the gastrointestinal tract along with an increase in the number of eggs in the rabbits' feces (Kobayashi & Horii, 2008). Therefore, disorders in the functioning of the gastrointestinal tract cause anorexia, loss of weight and consequent death of rabbits infected with mature *S. papillosus* (Taira, 1974; Nakamura & Motokawa, 2000; Kobayashi & Horii, 2008).

To deal with nematodiases, farmers use various preparations of synthetic origin (Boyko & Brygadyrenko, 2019). Quite often in the literature one can find data on use of plant-based preparations, as well as aqueous and alcohol extracts from medical plants (Rahmann & Seip, 2006; Burke et al., 2009; Lu et al., 2010; Lateef et al., 2013; Boyko et al., 2016). Species composition of pasture plants and nematode parasites of mammals in different climatic zones is different (Boyko et al., 2009). The objective of this article is to evaluate the impact of representatives of Rosaceae, Asteraceae, Fabaceae, Cannabaceae, Apiaceae families, common in the territory of Steppe Ukraine, on the vitality of *S. papillosus* and *H. contortus* nematode larvae in experiment *in vitro*.

Materials and methods

In the experiment we used feces of sheep which were spontaneously infested with strongyloidiasis and haemonchosis. Conditions of their maintenance were satisfactory. The animals had free access to fodder and water. In the summer the sheep grazed and spent some time time in a pen. During cold periods, they lived in a pen outdoors. Feces of sheep were examined for presence of eggs of nematodes in the Dnipro National Agro-Economic University. Analysis of feces was undertaken using the McMaster and Baermann tests (Baermann test) (Zajac et al., 2011).

Cultures of *S. papillosus* and *H. contortus* were cultivated out of fresh (1–5 hour) feces of sheep, collected in May 2018 in the territory of Dnipropetrovsk Oblast (Ukraine). Exposure of cultivation of *S. papillosus* and *H. contortus* larvae was 8 days. Larvae were obtained using the Baermann method: 15 g of feces with larvae were immersed with 20 mL of distilled warm (36 °C) water. Emergence of larvae from feces was observed after 2 hours. Then the distilled water with larvae (by 4 mL) was poured into test tubes and centrifuged for 5 minutes at 1,500 rpm. The supernatant liquid (3 mL) was removed with a pipette, and 1 mL of the sediment with larvae was evenly stirred and put in the amount 0.1 mL into 1.5 mL plastic test tubes. In the experiment, we used five concentrations of aqueous solution of each of the 21 species of plants (*Sanguisorba officinalis* L., *Rosa canina* L., *Crataegus sanguinea* Pall., *Crataegus pentagyna* Waldst. & Kit. ex Willd., *Armeniaca vulgaris* Lam., *Taraxacum officinale* F. H. Wigg., *Iva xanthiifolia* Nutt., *Artemisia campestris* L., *Arctium minus* (Hill) Bernh., *Ambrosia artemisiifolia* L., *Cannabis sativa* L., *Humulus lupulus* L., *Melilotus officinalis* (L.) Pall., *Vicia cracca* L., *Lotus ucrainicus* Klok., *Onobrychis arenaria* (Kit.) DC., *Foeniculum vulgare* Mill., *Eryngium planum* L., *Conium maculatum* L., *Juniperus communis* L., *Thuja occidentalis* L.) (Table 1).

Into the test tubes with larvae, 1 mL aqueous solution of plant was added (n = 7) in each. The exposure of larvae in aqueous solutions of plants and without solution (control) lasted 24 h. Then in the contents of each test tube, living and dead larvae were counted. In calculation of LD_{50}, average value and standard deviation (SD) is shown.

Results

Mortality of non-invasive larvae of *S. papillosus* $L_{1–2}$ for most species of the plants compared with invasive stages of *S. papillosus* L_3 and *H. contortus* L_3 was reliably higher (Table 1). The most resistant to the effect of the studied plants were third stage larvae of *H. contortus*, they had the lowest mortality of any of the experimental larvae when exposed to aqueous solutions of medicinal herbs. A total of 100% of dead larvae was found only with solution of *T. officinale*. Third stage larvae of *S. papillosus* were more sensitive to the influence of aqueous solutions of plants. All 100% of *S. papillosus* larvae of this stage died after exposure to aqueous solution of *T. officinale*. Around 85% of larvae remained vital after exposure to aqueous solution of *A. artemisiifolia*. Only about 62% of third stage larvae of *S. papillosus* died under exposure to aqueous solution of *S. officinalis*.

The least resistant to the aqueous solutions of the tested plant species were *S. papillosus* larvae of the first and the second stage of development. They were affected by 14 out of 21 species of the tested medical plants: 100% killing power was exerted by aqueous solution of *V. cracca*, over 90% of the larvae died in aqueous solutions of *T. officinale*, *A. minus*, *S. officinalis* and *L. ucrainicus*, around 80% of larvae died in aqueous solutions of *T. occidentalis*, *A. artemisiifolia*, *J. communis* and *R. canina*. The lowest mortality (52–74%) among larvae was observed after exposure to aqueous solutions of *H. lupulus*, *A. vulgaris*, *O. arenaria*, *C. maculatum*, *E. planum*.

Discussion

Plants are a large source of substances with various mrdical properties. They are also used as anthelminthic, antibacterial preparations and insecticides. Anthelminthic activity of onion (*Allium cepa* L.) and coconut (*Cocos nucifera* L.) is described in the study by Mehlhorn et al. (2011). Lem et al. (2014) point to anthelminthic activity *in vivo* of *Terminalia glaucescens* (Combretaceae). Marley et al. (2003) found that in Great Britain, lambs which grazed in a pasture with *Cichorium intybus* L. and *Lotus corniculatus* L. were less infested than lambs which were grazing on perennial ryegrass and white clover (*Lolium perenne* L., *Trifolium repens* L.). In our studies on the impact of aqueous solution of *Lotus ucrainicus*, we recorded nematocidial action only against non-invasive stages of nematodes of *S. papillosus*. Plants rich in tannin affect nematodes of ruminants (Hoste et al., 2006). Waghorn (2008) also indicates the anthelminthic effect of tannin containing plants. Studies by Hassan et al. (2014) *in vivo* and *in vitro* revealed high nematocidial activity of extracs from *Cichorium intybus* and *Artemisia absinthium* L. (Asteraceae) towards nematodes of the gastrointestinal tract of sheep.

Many researchers have studied the impact of medical plants on the representatives of one of the largest orders of nematodes – Strongylida. Urban et al. (2008) proved the anthelminthic effect of *Allium sativum* L. (Alliaceae), *Artemisia absinthium* L. (Asteraceae), *Carum carvi* L. (Apiaceae), *Consolida regalis* Gray (Ranunculaceae), *Inula helenium* L. (Asteraceae), *Juglans regia* L. (Juglandaceae), *Satureja hortensis* L. (Lamiaceae) and *Valeriana officinalis* L. (Valerianaceae) *in vitro* against third stage larvae of *Trichostrongylus colubriformis* (Strongylida).

The influence of some species of *Artemisia* on nematodes of the genus *Trichostrongylus* and representatives of the orders Strongylida (*Haemonchus*, *Nematodirus* genera) and Rhabditata (*Strongyloides*) is reported by Sharma (1993).

Table 1
Description of plants used in the experiment

Family	Species	Part of plant	Active substance
Rosaceae	*Sanguisorba officinalis* L.	leaves, thin stems, inflorescences, seeds	Leaves contain ascorbic acid (up to 0.92%)
	Rosa canina L.	leaves, thin stems, inflorescences, seeds	Vitamins C, A, B_1, B_2, K, P, mineral substances, organic acids, flavonoids, tannins and sugar; and vanillin in seeds
	Crataegus pentagyna Waldst. & Kit. ex Willd.	leaves, thin stems, inflorescences, seeds	Flavonoids: hyperoside, quercitrin, quercetin, vitexin, acetylvitexin, and also hydroxycinnamic acids – caffeic and chlorogenic acids
	Crataegus sanguinea Pall.	leaves, thin stems, inflorescences, seeds	Flavonoids: hyperoside, quercitrin, quercetin, vitexin, acetylvitexin, and also hydroxycinnamic acids – caffeic and chlorogenic acids
	Armeniaca vulgaris Lam.	leaves, thin stems, inflorescences, seeds	Tannins, flavonoids, carbohydrates, vitamin C, phenol-carbon acids, carotene, nitrogen-containing compounds (amygdalin, hydrogen cyanide), essential and fatty (oleic, linolenic, arachidic and other acids) oils
Asteraceae	*Ambrosia artemisiifolia* L.	leaves, thin stems, inflorescences, seeds	Phenol-carbon acids (ferulic, isoferulic, caffeic, chlorogenic, glycoside-caffeic acids), coumarins (scopoletin, scopoline, esculetin, umbelliferone, skimmin) and flavonoids (iaceidin, quercetin, isorhamnetin, isorhamnetin-3-rutinoside, quercemeritrip, isoquercitrin, glycosides of xanthomycrol and 4',5-dihydroxy-3,6,7,8-tetramethoxyflavone)
	Artemisia campestris L.	leaves, thin stems, inflorescences	Natural aminoacids, and also salts of potassium, tannins, and essential oils enriched with vitamins of groups A, B, ascorbic acid
	Iva xanthiifolia Nutt.	leaves, thin stems, inflorescences	Sesquiterpenoids, including sesquiterpenoid coronopilin, flavonoids, essential oil, saponins, low percentage of alkaloids
	Taraxacum officinale F. H. Wigg.	leaves, inflorescences	Taraxacin and taraxacerin, 2–3% natural rubber substances, taraxanthin, flavoxanthin, vitamins C, A, B_2, E, PP, lutein, triterpene alcohols, arnidiol, faradiol, choline, saponins, resins, salts of manganese, iron, calcium, phosphorus, up to 5% of protein
	Arctium minus (Hill) Bernh.	leaves, inflorescences, seeds	Seeds contain 25–30% of fatty oil. The roots contain inulin, essential oils, fatty oils, tannins, stigmasterol, amarines, protein, fatty acids: stearic and palmitic acids
Cannabaceae	*Cannabis sativa* L.	leaves, thin stems, inflorescences, seeds	Seeds contain 30–38% of fatty oils mainly composed of glycerides which are not saturated with fatty acids (linoleic, linolenic and butyric), proteins, aminoacids (glycol, alanine, valine, leucine, isoleucine, phenylalanine, threonine, tyrosine, aspartic acid, trigonelline, oxyproline and others), carbohydrates, quebrachitol, phenol compounds (cannabinol and cannabidiol) and traces of alkaloids
	Humulus lupulus L.	leaves, thin stems, inflorescences	Essential oil (myrcene (30–50%) and myrcenol, linalool, geraniol, farnesene, caryophyllene, luparol, luparenol, ethers of formic, acetic, butyric and other acids), resin extracts of hop, wax, natural gum, bitter acids, valeric, n-aminobenzoic and hop acids, glycoside lupulin, carotene, ascorbic acid, choline, thiamine, nicotinic acid, yellow colouring substance, tannins (3%), flavonoids, 0.095–0.190% of ascorbic acid
Fabaceae	*Vicia cracca* L.	leaves, thin stems, inflorescences, seeds	Protein (30%), ascorbic acid, tocopherol, calcium, carotene, flavonoids, phosphorus; seeds of pea contain glycoside vicianin
	Melilotus officinalis (L.) Pall.	leaves, thin stems, inflorences, seeds	Coumarins and their derivatives (0.4–0.9%: coumarin, dicoumarol, dihydrocoumarin, glycoside melilotoside), flavonoids (robinin, flovin, kaempferol and its derivatives), melilotin, essential oil (0.01%), saponins, derivatives of purine (allantoin), phenol-carbon acids (hydroxycinnamic, coumaric, melilotic), phenol triterpene compounds, nitrogen compounds, aminoacids, tannins, fatty-like substances (up to 4.3%), macro- and microelements (accumulate molybdenum, selenium); seeds contain fatty acids (palmitic, stearic, oleic, linoleic, linolenic, arachidic, behenic, lignoceric)
	Lotus ucrainicus Klok.	leaves, thin stems, inflorescences, seeds	Contains aminoacid canavanine, long chain fatty acids, lipids, phenol-carbon acids (n-coumaric, ferulic), flavonoids (quercetin, kaempferol, cormiculatin, gossypetin, guayaverin, quercetin, quercitrin), carotenoid (alpha and beta), carotenes (xanthophyll, violaxanthin)
	Onobrychis arenaria (Kit.) DC.	leaves, thin stems, inflorescences	Significant amount of carbohydrates, aminoacids, proteins, fats and cellulose, some enzymes; in herb – carotene and ascorbic acid; seeds contain sucrose, raffinose, fatty oils with solid fatty acids (up to 7–8% of fatty oils)
Apiaceae	*Foeniculum vulgare* Mill.	leaves, thin stems, inflorescences, seeds	Essential oil (up to 6%) which contains: anethole (up to 60%), α-pinene, α-phellandrene, dipentene, limonene, camphene, timolol, foeniculin, estragole, ethylphenhan, fenchone (20%), methyl chavicol (10%); protein substances, fatty oil (up to 18%) which contains petroselinic, oleic, linoleic, palmitic acids, coumarins
	Eryngium planum L.	leaves, thin stems, inflorescences	Phenol-carbon compounds, glycolic, malic, oxalic, citric, malonic acids, essential oil (up to 0.14%), tannins, flavonoids (quercetin, kaempferol); 0.5% triterpene saponins, carotene, ascorbic acid
	Conium maculatum L.	leaves, thin stems, inflorescences, seeds	Poisonous alkaloid coniin, methylconiine, conhydrine, pseudoconhydrine, coniceine, and also fatty oil which contains glycerides, petroselinic and petroselidinic acids; in leaves – up to 0.1% of alkaloids (coniin), essential oil (up to 0.08%), caffeic acid; in flowers – up to 0.24% of alkaloids, quercetin, kaempferol; in seeds – up to 2% of alkaloids and 0.08% of essential oil and caffeic acid
Cupressaceae	*Juniperus communis* L.	leaves, thin stems	Sugar (up to 42%), colouring substances, organic acids (formic, acetic, malic), resins (9.5%), essential oil (up to 2%) which contains terpenes camphene, cadinene, terpineol, pinene, borneol
	Thuja occidentalis L.	leaves, thin stems	Essential oil, tannins, resin, aromadendrin, taxifolin, pinipicrin, pinene, thujin, sesquiterpene, flavonoids and other biologically active substances were also found in it

Note: Table contains generalized information from many literature sources on substances present in plants, some of which can exert a killing effect on nematodes.

The impact of *Artemisia brevifolia* Wall. on vitality of *H. contortus in vitro* is described by Iqbal et al. (2004). Aqueous extract of this plant had no significant effect on nematodes in contrast to methanol extract. Our studies demonstrate similar results on the impact of aqueous solution of *Artemisia campestris* L. on vitality of *H. contortus* and *S. papillosus in vitro*.

Table 1

Mortality (%) of nematode larvae of *S. papillosus* and *H. contortus* in aqueous solution of different plant species of Rosaceae, Asteraceae, Fabaceae, Cannabaceae, Apiaceae and Cupressaceae families during 24 h laboratory experiment (x ± SD, n = 7)

Family	Species	Stage of the development of nematode	Concentration of aqueous solution of plant, %						LD_{50}
			3.000	0.750	0.188	0.047	0.012	0.000	
Rosaceae	*Sanguisorba officinalis* L.	*S. papillosus*, L_3	61.7 ± 41.0	47.2 ± 41.3	33.3 ± 37.3	6.1 ± 8.7	5.6 ± 12.4	0.0 ± 0.0	0.814 ± 0.263
		S. papillosus, L_{1-2}	90.0 ± 8.5	96.0 ± 5.2	95.9 ± 4.4	21.5 ± 17.0	16.5 ± 6.9	11.1 ± 5.8	0.074 ± 0.019
		H. contortus, L_3	0.0 ± 0.0	5.6 ± 12.4	6.1 ± 8.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Rosa canina L.	*S. papillosus*, L_3	7.3 ± 5.5	3.9 ± 5.6	2.8 ± 6.2	2.8 ± 6.2	2.8 ± 6.2	1.4 ± 3.1	–
		S. papillosus, L_{1-2}	85.3 ± 5.1	86.8 ± 3.5	80.2 ± 5.3	37.3 ± 9.7	28.0 ± 6.5	23.7 ± 2.2	0.067 ± 0.025
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.4 ± 5.3	–
	Crataegus sanguinea Pall.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	28.4 ± 6.1	3.2 ± 4.6	1.3 ± 2.1	1.7 ± 2.6	0.5 ± 1.0	1.7 ± 1.8	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Crataegus pentagyna Waldst. & Kit. ex Willd.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	10.4 ± 4.3	9.0 ± 4.3	11.0 ± 4.0	7.3 ± 1.9	9.3 ± 4.3	10.0 ± 3.0	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Armeniaca vulgaris Lam.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	52.4 ± 11.6	59.6 ± 9.5	26.7 ± 15.7	15.9 ± 12.1	8.2 ± 8.5	7.8 ± 8.1	0.631 ± 0.217
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
Asteraceae	*Taraxacum officinale* F. H. Wigg.	*S. papillosus*, L_3	100.0 ± 0.0	49.7 ± 24.2	27.8 ± 20.8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.760 ± 0.420
		S. papillosus, L_{1-2}	99.1 ± 1.1	94.0 ± 5.8	41.3 ± 12.6	21.5 ± 17.0	13.4 ± 3.6	6.8 ± 4.6	0.386 ± 0.227
		H. contortus, L_3	100.0 ± 0.0	0.1 ± 0.2	1.9 ± 4.1	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	1.775 ± 0.100
	Iva xanthiifolia Nutt.	*S. papillosus*, L_3	27.3 ± 18.1	12.1 ± 13.3	15.3 ± 15.5	6.9 ± 10.1	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	37.9 ± 8.0	33.8 ± 3.7	48.7 ± 7.9	29.6 ± 9.0	14.6 ± 6.7	9.6 ± 3.9	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Artemisia campestris L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	1.5 ± 3.4	1.9 ± 4.1	1.0 ± 2.2	0.0 ± 0.0	0.0 ± 0.0	0.7 ± 1.6	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Arctium minus (Hill) Bernh.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	94.2 ± 1.7	76.8 ± 9.7	72.6 ± 4.6	61.4 ± 4.6	10.9 ± 4.5	7.5 ± 3.0	0.032 ± 0.014
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Ambrosia artemisiifolia L.	*S. papillosus*, L_3	85.4 ± 16.2	72.4 ± 13.3	28.2 ± 9.2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.448 ± 0.203
		S. papillosus, L_{1-2}	86.5 ± 13.7	84.1 ± 6.0	78.4 ± 3.6	4.2 ± 0.7	4.4 ± 1.6	5.5 ± 2.5	0.110 ± 0.086
		H. contortus, L_3	6.1 ± 8.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
Cannabaceae	*Cannabis sativa* L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	39.2 ± 8.8	31.7 ± 10.2	27.3 ± 3.9	5.8 ± 2.0	3.1 ± 0.4	4.4 ± 2.0	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Humulus lupulus L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	52.1 ± 13.1	55.6 ± 4.1	28.5 ± 5.5	22.8 ± 5.7	24.6 ± 4.8	17.9 ± 3.8	0.701 ± 0.329
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
Fabaceae	*Melilotus officinalis* (L.) Pall.	*S. papillosus*, L_3	4.2 ± 9.3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	24.3 ± 6.7	9.6 ± 7.4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Vicia cracca L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	100.0 ± 0.0	58.3 ± 34.4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.691 ± 0.283
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Lotus ucrainicus Klok.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	98.2 ± 1.3	84.9 ± 6.2	71.9 ± 2.7	59.3 ± 11.2	40.8 ± 5.6	6.0 ± 2.9	0.028 ± 0.019
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Onobrychis arenaria (Kit.) DC.	*S. papillosus*, L_3	2.8 ± 6.2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	60.2 ± 8.9	42.5 ± 14.4	31.1 ± 5.9	5.0 ± 1.4	3.4 ± 0.9	3.6 ± 1.2	1.830 ± 0.980
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
Apiaceae	*Foeniculum vulgare* Mill.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	24.7 ± 12.7	11.1 ± 5.4	6.4 ± 6.6	9.9 ± 7.5	12.7 ± 9.5	6.2 ± 3.9	–
		H. contortus, L_3	2.1 ± 4.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Eryngium planum L.	*S. papillosus*, L_3	8.9 ± 13.1	2.8 ± 6.2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.1 ± 4.7	–
		S. papillosus, L_{1-2}	73.6 ± 4.7	78.6 ± 7.0	68.1 ± 15.5	45.7 ± 18.9	25.1 ± 19.2	28.8 ± 11.6	0.062 ± 0.034
		H. contortus, L_3	3.3 ± 7.5	2.8 ± 6.2	2.1 ± 4.7	0.0 ± 0.0	0.0 ± 0.0	3.3 ± 7.5	–
	Conium maculatum L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	61.9 ± 19.2	35.6 ± 2.7	20.6 ± 6.8	15.9 ± 3.5	20.0 ± 6.0	17.5 ± 4.2	2.043 ± 1.091
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
Cupressaceae	*Juniperus communis* L.	*S. papillosus*, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	89.7 ± 4.4	69.5 ± 3.3	59.6 ± 3.7	23.9 ± 2.3	16.1 ± 3.9	17.5 ± 4.2	0.139 ± 0.080
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–
	Thuja occidentalis L.	*S. papillosus*, L_3	21.8 ± 18.6	14.4 ± 17.9	18.6 ± 14.0	5.2 ± 7.3	1.9 ± 4.1	0.0 ± 0.0	–
		S. papillosus, L_{1-2}	89.7 ± 3.1	89.3 ± 6.3	94.9 ± 1.6	31.6 ± 6.5	30.9 ± 7.9	25.5 ± 4.3	0.026 ± 0.019
		H. contortus, L_3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	–

Note: "–" – mortality of LD_{50} nematode larvae was calculated only for groups in which mortality exceeded 50% in concentrations of aqueous solution of plant of up to 3.0%.

Khan et al. (2015) also studied the impact of methanol extracts of two species of *Artemisia*: *A. indica* and *A. roxburghiana* on the development of eggs, larvae and mature individuals of *H. contortus in vitro*. They found that methanol extracts such as *A. roxburghiana* exerted maximum anthelminthic action in concentration of 50 mg/mL on the development of eggs (mortality was 85.0 ± 21.2% and 80.0 ± 28.3%, respectively), larvae (18.0 ± 2.8% и 17.0 ± 4.2%) and mature helminths (8.5 ± 2.1% and 8.0 ± 2.8%). Under exposure to *A. indica*, a stronger effect was observed in all concentrations compared with *A. roburghiana*. Sangwan & Sangwan (1998) reported negative effect *in vitro* of the extract from fresh leaves of *Melia azedarach* L. (Meliaceae) on larvae of *H. contortus*. Kamaraj et al. (2010) also evaluated *in vitro* ovicidal and larvicidal effects of *M. azedarach* on *H. contortus*. Both of the extracts were evaluated in five concentrations: 12.5, 6.2, 3.1, 1.6, and 0.8 mg/mL. Aqueous and alcoholic extracts from leaves prevented development of 99.4% of eggs and 100.0% of larvae at 12.5 mg/mL. The impact of plants on this species of nematode was also studied by Al-Qarawi et al. (2001): they indicate the anthelmintic effect of some species of plants of the *Calotropis* genus. Kabore et al. (2009) also studied *in vitro* the impact of aqueous extracts of two species of medicinal plants (*Anogeissus*

leiocarpa (DC.) Guill. & Perr. (Combretaceae) and *Daniellia oliveri* (Rolfe) Hutsh. & Dalziel (Fabaceae) on the development of eggs, first stage larvae and mature nematodes of *H. contortus*. In the experiment they used leaves of *A. leiocarpa* and bark of *D. oliveri* in six concentrations. Effective dose (ED_{50}) for trunk bark of *D. oliveri* equaled 245.9 μg/mL. This indicator for leaves of *A. leiocarpa* was 409.5 μg/mL. Thus, highest ovicidal impact was caused by trunk bark extract of *D. oliveri*. ED_{50} for extracts from *A. leiocarpa* and *D. oliveri* against L_1 *H. contortus* equaled 411.4 and 362.3 μg/mL, respectively.

The study we conducted on 21 species of plants of six families on vitality of nematode larvae of *H. contortus* and *S. papillosus* determined different extents of their toxic impact on parasites. *Taraxacum officinale* had nematocidial effect on L_3 larvae of *H. contortus* and $L_{1–3}$ *S. papillosus* in 3% concentrations. *S. officinalis* and *A. artemisiifolia* displayed lower impact towards free-living stages of nematodes. Nematocidial properties of these species were recorded against larvae of *S. papillosus*. The rest of the studied species of plants had no nematocidial properties when used as aqueous solutions. 11 out of 21 solutions in 3% concentrations exerted killing effect only on non-invasive larvae of *S. papillosus*.

Conclusion

Thus, pasture plants can exert nematocidial activity towards eggs, larvae and mature nematodes. However, the possibility of consumption of these plants by different species of ruminants can be rather limited due to the toxic effect of these substances on mammals. Accumulation on the soil surface of litter which contains high concentrations of tannins, alkaloids and other groups of physiologically active compounds, potentially can contribute to improvement of the parasitological condition in pastures. The natural composition of the pasture flora most often includes 10 to 100 species of plants (higher in mesophilous, lower in xerophilous conditions of humidity). At the same time, plants are distributed across territories not uniformly, but rather mosaically, forming different geobotanical associations. Perhaps, nematode larvae also have a scattered distribution across territories, which for infesting their mammal hosts, need to travel several meters in the soil and litter and climb up a plant (which could also be toxic for nematode larvae) to a height of 10–30 cm for most efficient infestation of the host. Peculiarities of interaction between plants and larvae of such harmful species for livestock as *H. contortus* and *S. papillosus* are only beginnig to be studied. In his sphere, interesting discoveries could be made both in finding species with notable anthelminthic activity, and general patterns of functioning of pasture parasitocenosis in spatial and seasonal dynamics.

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