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The effects of soil organic matter content and soil texture on the population number of *Pratylenchus loosi*, in tea plantation of Iran

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Abstract

Tea, *Camellia sinensis* (L.) Kuntze, is an edible and evergreen plant which has numerous medicinal and calmative characteristic. Tea root lesion nematodes, *Pratylenchus loosi*, is one of the most important pests in Iran, which causes loss in quantity and quality of tea. 183 soil and root samples were taken randomly from tea plantations in Iran. The highest mean population of *P. loosi* in soils and roots was observed in soils with organic matter less than 2 percent. The results of regression analyses showed that there is no significant correlation between soil organic matter content with neither the population number of nematodes in one g of feeder roots nor the population density in 100 g of soil. Three types of soil texture classes were observed such as loam, sandy loam and sandy clay loam as if the highest percentage of infestation was observed in sandy loam soil. The results of regression analysis showed that there was no significant correlation between sand, silt and clay percent with mean population of nematode in sandy loam, sandy clay loam and loam soils. Total results showed that tea root lesion nematode in light texture soils are more active and had greater population numbers.

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Introduction

Tea, *Camellia sinensis* (L.) O. Kuntze, is an edible and evergreen plant which has numerous medicinal benefits due to its antibacterial and antioxidant properties (James, 1983). Tea is considered as a strategic economic crop in Iran. According to Food and Agriculture Organization statistics in 2010, the total world production of tea was 3,950,000 tonnes. The area harvested in Iran already is about 32,000 ha. Plant parasitic fungi and nematodes are the most important pathogens that cause damage on tea plants. Among the plant parasitic nematodes, *Meloidogyne* spp., *Pratylenchus* spp., *Paratrichodorus porosus* and *Rotylenchus reniformis* are recognized as major biotic constraints in many tea production systems throughout the world with root lesion nematodes being the most important (Willson, 1999). At present, tea root lesion nematodes, *P. loosi*, is known as the most important damaging pests of tea in Iran and many tea growing countries. This nematode causes severe damage to tea plants reducing growth and yield of infested plants (Seraji, 2007; Seraji *et al.*, 2010). Despite their plant parasitic status, most plant–parasitic nematodes also spend part of their life cycle in the soil.

Soil texture is one of the important factors influencing the distribution of *Pratylenchus* species (Castillo & Volvas, 2007). One of the alternative strategies for management of plant pararsitic nematodes is the application of organic amendments in the soil (Agyarko & Asante, 2005, Oka *et al.*, 2000, Thoden *et al.*, 2012).

Oka *et al.* (2000) pointed that organic amendments have consistently been shown to have beneficial effects on soil nutrients, soil physical conditions, soil biological activity and thereby improving the health of plants and reducing populations of plant parasitic nematodes.

On the other hand numbers of free living microbivorous nematodes increase rapidly in the soil following the addition of organic and inorganic fertilizers while there can be a corresponding decrease in the numbers of plant parasitic nematodes (Akhtar & Malik, 2000, Thoden *et al.*, 2012).

Organic amendments can both improve tolerance of the plant to nematodes and also reduce nematode populations (Crow & Dunn, 1994). Kimenju *et al.* (2004) reported that application of organic amendments stimulated the activity of natural antagonists of plant parasitic nematodes. Akhtar & Malik (2000) suggested that free-living nematodes reproduce rapidly when presented with organic substrate and play an important role in recycling of plant nutrients making them available to plants. Koenning *et al.* (2003) have revealed the nematicidal potential of organic products used as soil amendments. When incorporated into the soil, organic substrates undergo biologically mediated decomposition to release NH4+, formaldehyde, phenols and volatile fatty acids, among other compounds (Wang *et al.*, 2004). However, in some studies the addition of organic amendments, besides increasing yields, also increased number of plant parasitic nematodes, highlighting the positive effects caused by microbivorous nematodes (Thoden *et al.*, 2012). So in this study we tried to define the effects of soil organic matter and texture on the population densities of *P. loosi* in tea plantations in Iran.

Materials and methods

183 soil and root samples from different location in Northern Iran (Rasht, Fuman, Shaft, Lahijan, Siahkal, Langroud, Roudsar, Ramsar) were collected during September, October and November 2010. Sampling was done once in each region. Tea plantations in Iran were located in an area of about 350 square kilometres near the Southern Caspian Sea (located in Northern Iran, Guilan and Mazandaran provinces) with geographic coordinates 37° 21′ 43″ N latitude, 49° 29′ 83″ E longitude in west and 36° 51′ 26″ N latitude, 50° 41′ 20″ E longitude in East. Therefore, to fully cover the tea plantations for every two square kilometres, a random sample was prepared (at least six to 10 points). Before going to the desired location, we selected locations on satellite maps (Google Earth) noting the geographic coordinates of the sites. Six to

ten soil samples were taken from 5 to 40 cm depth in the root zone at each site. Tea fine roots (about 10 g) were also collected in the same site. Soil samples mixed then one kg soil and about 10 g roots were taken to the laboratory in the Iranian Tea Research Institute (Lahijan, Guilan). Samples for nematode analysis were stored in a refrigerator (at 4^{oc}) until nematode extraction. For soil analysis, each soil sample was air dried and passed through a 0.02 mm sieve. Classification of tea plantations soils based on soil taxonomy were available and were located in two orders; alfisols and inceptisoles.

Nematodes were extracted from soil by centrifugal flotation using a modified way of the method introduced by Jenkins (1964). The extraction from roots followed the method described by Coolen & d'Hered techniques (1972). From each soil sample approximately 250 g soil and 3 g of tea feeder roots were used. The obtained nematode suspensions were adjusted to 10 ml a subsample of 1 ml was transferred to a counting slide and nematodes identified and enumerated under a microscope (With a magnification of 20). Then the samples were returned to the suspension and the process was repeated five times. Finally the average number of nematodes per 1 g of tea fine roots and 100 g of soil was calculated.

The determination of soil organic carbon content was done by using the Walkley-Black chromic acid wet oxidation method (Walkley & Black, 1934). The soil texture was determined by the using hydrometer method.

Statistical analysis

All data were subjected to ANOVA using statistical software package SPSS 16. Regression analyses were performed to determine the effect of organic matter content and sil texture on the population density of tea root lesion nematode using the Curve Expert 1.5 (Hyams, 2010) software at $P \leq 0.001$.

Results

About 82% of the samples were infested with *Pratylenchus loosi* with population densities ranging from 0.66 to 884 per gram of roots and 1 to 186 in 100 g of soil.

Effects of soil organic matter content on population level of Pratylenchus loosi

Organic matter contents varied from 1.38 to 9.17%. Most samples (72.65%) had organic matter contents between 2 to 4% (Table 1). Most of them showed organic matter contents of 4%. The highest mean soil as well as root population numbers of *P. loosi* were observed in soils with organic matter contents of less than 2% (Table 1).

Table 1. Percentage of infestation and mean population of *P. loosi* in different soil organic matter ranges.

Organic matter classes	N. samples	N. infested sample	Percentage of infestation[a]	Mean soil population[b]	Mean root population[c]
OM<2	18	16	88.88	88.2 (r=0.03[ns])	4.8 (r=0.04[ns])
2<OM<4	133	111	83.46	42.1 (r=0.36[ns])	96.4 (r=0.12[ns])
4<OM<6	28	20	71.43	24.5 (r=0.12[ns])	46 (r=0.22[ns])
OM>6	4	3	75	9.3 (r=0.04[ns])	14.22 (r=0.07[ns])
Totall	183	150	82	46.2 (r=0.26[ns])	91(r=0.37[ns])

[a] Proportion of infested samples per organic matter class;

[b] Mean population of P. loosi per 100 g of soil;

[c] Mean population of P. loosi per 1 g of root;

[r] = Regression coefficient; ns= no significant.

Fig. 1 shows the nematode population numbers at different soil organic matter contents. According to the Fig. 1A, population numbers of *P. loosi* in roots only reached 200 individuals/g root when the organic matter content was less than 4%. A similar distribution can be seen for the soil population numbers (Fig 1B).

Table 2. Modelling of relationship between soil organic matter with pathogenicity characters of *P. loosi* in Iran.

Y	X	Best fitting model	r	STD
Pf R[a]	Organic matter	Modified Exponential, y= 33.53*e^ (2.87/x)	0.27	132.6
		Root Fit, y= 33.53*17.63^(1/ x)	0.27	132.6
		Reciprocal Model, y=1/0.004*x-0.002	0.27	132.6
Pf S[b]	Organic matter	Reciprocal Model, y=1/0.007*x+ 0.001	0.32	38.92
		Power Fit, y= 111*x^(-0.94)	0.32	38.92
		Logarithmic fit, y=85.86-40.41*lnx	0.31	38.96

[a] Mean population of *Pratylenchus loosi* per one g of root;

[b] Mean population of *P. loosi* per 100 g of soil.

Due to the non-uniformity of the samples in the different ranges of organic matter classes it was impossible to provide appropriate models (Table. 2). The results of regression analysis showed that there is no significant correlation between neither the amount of soil organic matter and the mean population number of *P. loosi* gram of root nor the population number of *P. loosi* per 100 g of soil. Nevertheless, mean population of *P. loosi* in soils with high percent of organic matter was lower than soils with low percent of organic matter (Fig. 1).

Effects of soil texture on population level of *Pratylenchus loosi*

Table 3 shows the influence of different soil types on the root and soil population numbers of *P. loosi*. Between the different soil types (sand, sandy loam and sandy clay loam) the proportion of infested samples ranged from 76% to 84%, respectively, with sandy loam soils showing the highest proportion of infested samples (Table 3). Mean numbers of soil and root population numbers were highest in sandy loam soils and lowest in sandy clay loam.

Table 3. Percentage of infestation and mean population of *P. loosi* in different soil texture.

Soil texture	N. samples	N.infested sample	Percentage of infestationa	Mean soil Populationb	Mean root populationc
Sandy loam	89	73	82	51.3	121.2
Sandy clay loam	64	54	84.4	37.1	74.5
loam	30	23	76.7	39.4	85.2

[a]Number of infested samples in each soil texture into all of samples in each range;

[b] Mean population of P. loosi per 100 g of soil;

[c] Mean population of P. loosi in 1 g of root.

The results of regression analysis showed that there is no significant correlation between sand, silt and clay percent with mean population of nematode in sandy loam, sandy clay loam and loam soils (Table 4). We found that the mean population of nematodes decreased as soil silt content increased (Fig. 2). Similarly, mean population numbers of nematodes increased as the soil sand content increased (Fig. 3A) and decreased when soil clay content increased (Fig. 3B).

Discussion

The present study was undertaken to investigate the effects of soil organic matter content and soil texture on the population level of the tea root lesion nematode, *Pratylenchus loosi* in tea plantations of Iran. Results showed that there is no significant correlation between nematode population number and soil organic matter content. However, the mean population number of nematode was higher in soils with low value of organic carbon. The highest numbers of *P. loosi* were observed in soils with organic matter content lower than 4%. These findings are similar to previously made observations that the population number of tea root lesion nematodes was higher in soils of 3% organic matter content than in soils with 10% of organic matter (Gnanapragasam & Sivapalan, 1991).

Other studies found an increase of plant parasitic nematodes following the addition of soil organic matter (Thoden *et al.*, 2012). It has been accepted that the application of organic substrates leads to a build-up of micro-organisms which serve as a food substrates for free-living nematodes hence fostering their build-up. Populations of free-living nematodes such as bacteriovorous, fungivorous and predacious have been shown to rapidly increase following the addition of organic amendments (Akhtar & Malik, 2000; Thoden *et al.*, 2012). In addition, free-living nematodes may accelerate the decomposition of soil organic matter and increase mineralization of nitrogen and phosphorous thus triggering a chain reaction that favours their build-up (Kimenju *et al.*, 2004). Many studies have shown that addition of organic matter in the form of compost or green manures can reduce populations of plant parasitic nematodes (Walker, 2004).

Table 4. Correllation coefficient between soil particle percent in different soil texture with population number of *Pratylenchus loosi* in soils and roots.

Soil texture	Soil particle percent	Population of nematodes in roots	Population of nematodes in soils
Loam	Sand	0.08 ns	0.14 ns
	Clay	-0.05 ns	-0.04 ns
	silt	-0.31 ns	-0.27 ns
Sandy loam	Sand	0.18 ns	0.26*
	Clay	0.23 ns	-0.05 ns
	silt	-0.24*	-0.31**
Sandy clay loam	Sand	0.01 ns	0.04 ns
	Clay	-0.07 ns	-0.12 ns
	silt	-0.18 ns	-0.06 ns

**Correlation is significant at the 0.01 level (2-tailed).

*Correlation is significant at the 0.05 level (2-tailed).

ns no significant.

The plant-parasitic nematode population levels under organic amendment treatments might have changed for many reasons, including changes in soil properties, nutrients released to plants, increase in predators and parasitic microorganisms, toxic metabolites released from organic amendments breakdown, or health of the host crop (Oka, 2000). Thoroughly decomposed organic matter (humus) improves the soil texture and the ability of soil to retain water and plant nutrients. Additionally, humus promotes a better soil structure by acting as glue that causes soil particles to form aggregates.

In this study three soil texture classes were identified. Loam, sandy loam and sandy clay loam with the latter soil type showing the highest mean population levels and the highest proportion of infested samples. Probably, the higher population levels of *P. loosi* in lighter soils (sandy loam) can be explained by a better nematode mobility. Contrary, the oxygen levels and porosity are lower in heavier soils and as a result; the mobility of *P. loosi* in this soil types is lower. This result is inconsistent with other studies that reported the biggest damages by the tea root lesion nematode in clay soils with a poor drainage and high level of soil acidity (Gnanapragasam & Sivapalan, 1991). However, our findings are supported by Wallace (1973) who suggested that some *Pratylenchus* species are associated with lighter sandy soils (with higher availability of oxygen). It is evident that numbers of plant parasitic nematodes vary with soil texture as well as pH (Olabiyi *etal.*, 2009; Robinson, 2005).

Thus maximum pathogenicity and reproduction of root-lesion nematodes observed in silty clay soils. Contrary lowest numbers of root lesion nematodes and alluvial soils whereas were recorded in red soils with low soil porosity (Sundararaju & Jeyabaskaran, 2003). Similarly, Endo (1959) showed that sandy loam soils favoured the infestation and reproduction of *Pratylenchus brachyurus* while clay loam soils were least suitable. It was also reported that the pathogenicity of *P. neglectus* on rangeland grasses was lower in fine silt loam than in fine loam soils (Griffin, 1996).

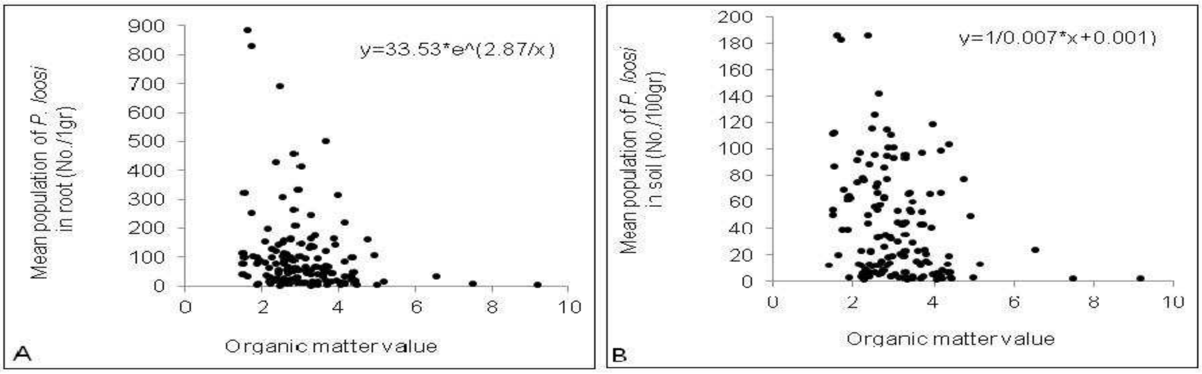

Fig. 1. Relationship between soil organic matter content and (A): mean population level of *Pratylenchus loosi* per 1 g of root; (B): mean population level of *P. loosi* per 100 g of soils.

In the current study the highest numbers accumulation of *P. loosi* were observed in lighter soils (sandy loam) with 5 to 10% clay, 60 to 65% sand and 10 to 15% silt. Similarly, the numbers increased as soil sand content increased and decreased as soil clay content increased. This shows that the tea root lesion nematode tends to prefer light to medium soils.

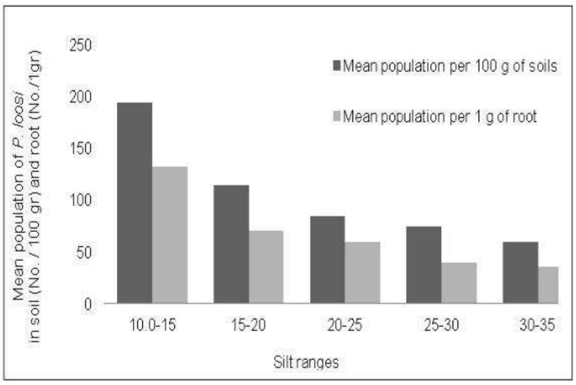

Fig. 2. Mean population changes of Pratylenchus loosi in 100 gr of soil and 1 gr of root in different soil silt ranges.

Contrary Thompson *et al* (2010) reported that maximum population levels and damage of *P. thornei* were observed in heavier soils (48% clay, 37% sand) and for *P. neglectus* in lighter soils (39% clay, 45% sand). Another study in Iran reported that there was a significant positive correlation between the soil sand content and the population density of both *P. neglectus* and *P. thornei* (r=0.43) and a significant negative correlation with soil clay content (r=-0.41) (Ghaderi *et al*, 2010). Similarly, soil type significantly influenced infection and severity of root necrosis caused by *P. coffeae* in banana (Sundararaju & Jeyabaskaran, 2003).

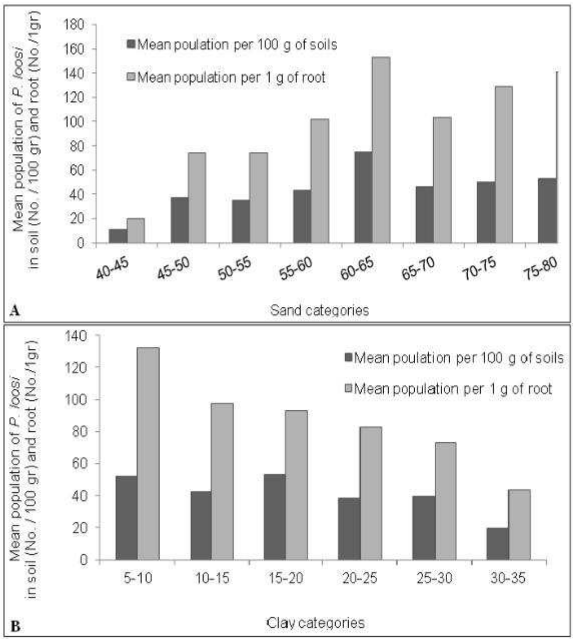

Fig. 3. A. Mean population of *Pratylenchus loosi* per 100 g of soil and 1 g of root in different soil sand categories. **B.** Mean population of *P. loosi* per 100 g of soil and 1 g of root in different soil sand categories.

In Iran, tea is cultivated in sandy loam, sandy clay loam and loam soils and with organic matter contents between 1.4 to 9.2%. In our study with 183 soil samples from Northern Iran, the highest mean population density and the highest percentage of infestation with *P. loosi* was observed in sandy loam soils. Most samples having organic matter contents between 2 to 4%. Highest mean population density of *P. loosi* was observed in soils with less than 4% organic matter. Furthermore, the mean population density of *P. loosi* was higher in lighter soils (sandy loam) than medium (loam) and heavier soils (sandy clay loam). Given that the soil texture is naturally pre-defined only the manipulation of soil with organic matter contents with manures (animal manure and tea wastes) or other forms of soil amendments (Calcite and Dolomite) could be used for the management of tea root lesion nematode in Iran.

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