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## POPULATION DENSITY OF PLANT-PARASITIC NEMATODES UNDER CONSERVATION AGRICULTURE AND CONVENTIONAL CROPPING SYSTEMS

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### ABSTRACT

The effects of tillage methods and crop residue management practices based on conservation and conventional agriculture systems were studied on the population density of plant parasitic nematodes in two different crop rotation systems. The experimental design was a split-plot arrangement in a randomized complete block with three replications. Three tillage systems (conventional tillage, minimum tillage and no-tillage) were assigned to main plots and three level of residue management (0, 30, and 60%) were assigned to sub plots. The first rotation system include wheat, maize, wheat, melon and wheat and the second rotation system include wheat, canola, wheat, Persian clover, tomato and wheat. The results of variance analysis on the population densities of plant parasitic nematodes showed that in the first rotation system, the effect of tillage on the population of *Filenchus* spp. was statistically significant but the effects of tillage, residue retention and the interaction between tillage × residue retention on the population of other plant parasitic nematodes and the total number of plant parasitic nematodes were not statistically significant. In the second rotation system, tillage had a significant effect on the population density of root lesion nematode (*Pratylenchus thornei*), *Filenchus* spp. and *Geocenamus* spp. Furthermore the effect of residue retention on the population of *Geocenamus* spp. and interaction between tillage × residue retention on the population of root lesion nematode (*Pratylenchus neglectus*) and *Geocenamus* spp. was significant but the effect of tillage, residue retention and the interaction between tillage × residue retention on the population of other plant parasitic nematodes and the total number of plant parasitic nematodes was not significant. The present study indicated that the conservation agriculture practices have no significant influence on the population density of major plant parasitic nematodes under the two crop rotation sequences and do not increase the risk of damage by nematodes.

**Keywords:** Agronomic practices, crop residue, farming systems, plant disease, rotation, tillage.

### INTRODUCTION

Increasing demands for food production due to expanding global population and climate change is putting tremendous pressure on the agricultural lands, natural resources and environment. Conservation agriculture (CA) is considered as a key route to sustainable and more productive cropping system. CA is based on three principles: minimal soil disturbance, continuous retention of crop residues or cover crops on

the soil surface and crop rotation (FAO, 2017; Hobbs et al., 2008; Verhulst et al., 2010). The CA practices is reported to have direct and indirect impact on many soil characteristics including physical, biochemical and biological soil quality (Verhulst et al., 2010). Over the past decade, CA adopted by many farmers in developed and developing regions of the world (Paulitz, 2006; Derpsch and Friedrich, 2010) and contributed to the sustainable intensification of agriculture by improving soil quality, resource use efficiency and productivity. However, diseases caused by residue-borne and soil-inhabiting pathogens, including plant pathogenic nematodes, remain major obstacles to wider adoption of conservation agriculture-based cropping systems.

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Nematodes are an important part of soil biological communities and an important group of plant pathogens. They play important roles in nutrient cycling of soil by feeding on bacteria, fungi and microfauna (Overstreet *et al.*, 2010). Crop rotations is reported to have many benefits to the agricultural systems including prevention of weed species from domination, reducing population densities of pests and pathogens, maintaining soil fertility, improving soil health and biological activity and producing higher yields (Nusbaum and Ferris, 1973; McLaughlin and Mineau, 1995; Florentín *et al.*, 2010; Duiker and Myers, 2006; Hobbs *et al.*, 2008). Crop rotations with nonhost plants play an important role in minimizing pest and disease build-up in CA systems (Trenbath, 1993) by breaking the life cycle of pathogen propagules in soil in the absence of the sources of nutrients (Reid *et al.*, 2001) and reducing the survival rate of pathogens in soil. Crop rotation has been used extensively as an efficient practice for controlling of a variety of important plant diseases; however, most studies on the role of crop rotation in disease control have looked at crop rotation in conventional agricultural systems.

The impact of reduced and zero tillage on disease incidence and the population dynamics of soil borne plant pathogens including nematodes is not still well understood (Hobbs *et al.*, 2008; Raaijmakers *et al.*, 2009; Kassam *et al.*, 2009). The effect of tillage level on different plant pathogens varies greatly depending on their survival strategies and life cycle (Bockus and Shroyer, 1998), and soil conditions specially soil moisture and temperature (Krupinsky *et al.*, 2002). Zero tillage showed great potential for facilitating integrated pest management and biological control (Govaerts *et al.*, 2006). Increased soil fauna including microorganisms, earthworms and nematodes is reported under no-till and residue retention practices compared to conventional tillage practices. Reduced or zero tillage systems, can change the soil environment and gather crop residues that may contain pathogen propagule at the soil surface resulting in an increase, decrease, or no change in disease and pest incidence or severity depending on the cropping system and pathogen (Dordas, 2008; Ziadi *et al.*, 2013). Surveys of fungal diseases and nematodes in food legumes in ICARDA's long-term rotation trials showed that both soil borne pathogens and foliar diseases including cyst nematode and

Ascochyta blight could be a problem in conservation cropping system (Seid *et al.*, 2012).

Permanent soil cover from previous crop residues are important sources of food for soil organisms including plant pathogenic bacteria, fungi and nematodes which can cause major changes in the population of these pathogens and increase the risk of disease infection of subsequent crops, especially in the case of residue-borne pathogens that survive as resting structures or produces their fruiting bodies in the plant residues and in mono-cropping systems (Hobbs and Govaerts, 2010). On the other hand, increased soil microbial biomass due to increased level of crop residues can potentially discourage pathogen development as increased numbers of microorganisms compete for resources or cause inhibition through antagonism or the release of antibiotic (Weller *et al.*, 2002).

In general, many studies reported that CA practices may cause changes in the incidence, severity and type of weeds, pests and plant diseases. Few studies, however, have compared the effects of the three core principles of CA on the population of plant pathogenic nematodes. Therefore, it is clear that further research needs to be conducted to address this issue. In the present study, the main objective was to investigate the effect of conservation agricultural practices on the population density of plant pathogenic nematodes as compared to the conventional farming systems.

#### **MATERIALS AND METHOD**

**Experimental design:** The impacts of three tillage system, conventional tillage (CT), minimum tillage (MT) and no-tillage (NT) and three level of crop residue retention (no residue, 30% residue, and 60% residue) were determined on the population density of plant pathogenic nematodes under two different rotation sequences. The experiment was a randomized complete split-plot design with three replications. Three tillage systems were assigned as main plots and three level of residue retention as subplots. Planting area of each subplot was 150 m<sup>2</sup>(15m ×10 m) and planting area of each main plot was 450 m<sup>2</sup> (3 \*150 m<sup>2</sup>).The planting area of each replication of the experiment was 4050 m<sup>2</sup> (9 \*450 m<sup>2</sup>) and the total planting area of each rotation experiment was 12150 m<sup>2</sup> (3 \*4050 m<sup>2</sup>).The field experimental site is located at Toroq Agricultural Research Station (36°13' N, 59°40' E), in Mashhad,

Northeast of Iran. The permanent experimental plots were established in autumn 2012 and continued for five cropping season (2012-2017).

**Tillage treatments:** In the intensive, conventional tillage practice, a moldboard plow was used followed by an offset disc plough and then a leveller. Depending on the crop, a furrower was used to complete the seedbed preparation practices and then a seeding machine was used for planting. In the minimum tillage treatment a single furrow, 1-way disc plough or a chisel plough and then a furrower and finally a seeding machine was used. In the no-tillage treatment, depending on the crop and soil conditions a chisel plough or other tined implement followed by a light trailing cover harrow or a no-tillage direct seeding machine was used.

**Residue management treatments:** In the zero residue retained treatment, all the surface residue from the former crop was removed just after the harvest, leaving a bare soil surface. In the other two residue management treatments, 30% and 60 % of plant residues were left on the soil surface.

**Rotation systems:** The field experiments arranged under two rotation systems. Both rotation systems were based on the wheat and are considered as the most predominant rotation systems in the region. The first crop rotation system included bread wheat

(*Triticum aestivum* L.), maize (*Zea mays* L.), wheat, melon (*Cucumis melo* L.) and wheat and the second crop rotation sequence included wheat, canola (*Brassica napus* L.), wheat, white clover (*Trifolium repens* L.), tomato (*Solanum lycopersicum* L.) and wheat. Details of crop rotation sequences and agronomical practices for each crop are provided in Table 1.

**Nematode sampling and analyses:** From each experimental plot, 10 soil samples were taken from the 0-30 cm depth in zig zag pattern using an auger of 2.5 cm in diameter. The 10 subsamples from each plot were thoroughly mixed together and a combined sample with approximately 1 Kg was transported to the Laboratory. Nematodes were extracted from 250 ml of each combined soil sample using sieving and centrifugal-flotation technique (Jenkins, 1964). One ml out of the 10 ml of the resulting suspension was used for measuring the population density of nematodes in 250 ml of soil samples using counting slides. Nematodes were identified at the genus level and the species level using valid nematodes systematic keys (Loof, 1978; Siddiqi, 1987; Handoo and Golden, 1989; Nickle, 1991; Hunt, 1993). Data were analyzed with MSTAT-C statistical software package. Means were compared using Duncan's Multiple Range Test (MRT).

Table 1. Details of two different wheat-based crop rotation sequences used in this study

Rotation system 1		Rotation system 2	
Growing season	Crop (cultivar)	Growing season	Crop (cultivar)
Oct. 2011-July 2012	Wheat (Parsi)	Oct. 2011-July 2012	Wheat (Parsi)
June 2013- Sep. 2013	Maize (Single cross 704)	Oct. 2012-June 2013	Canola (Hyola401)
Oct. 2013-June 2014	Wheat (Parsi)	Oct. 2013-June 2014	Wheat (Parsi)
June 2015- Sep. 2015	Melon (Khatouni)	Sep. 2014- May 2015	White Clover (Harati)
Oct. 2015- June 2016	Wheat (Parsi)	June 2015- Sep. 2015	Tomato (Petoeearly CH )
		Oct. 2015-June 2016	Wheat (Parsi)

**RESULTS**

**Population densities of plant pathogenic nematodes in rotation system 1:** In rotation system 1, seven species of plant pathogenic nematodes including two species of root-lesion nematode (*Pratylenchus thornei* and *Pratylenchus neglectus*), stem and bulb nematode (*Ditylenchus* spp.), the spiral nematodes (*Helicotylenchus* spp.), stunt nematodes (*Geocenamus* spp.), *Boleodorus* spp. and *Filenchus* spp. were detected and their populations were quantified. The results of analysis of

variance of population density of plant pathogenic nematodes in rotation system 1 showed that only the effect of tillage on population density of *Filenchus* spp. was statistically significant (P=0.05). The effect of tillage, crop residue retention and the interaction between tillage and residue retention on population density of other nematodes and total nematode numbers were not statistically significant. Nematode population densities under different tillage treatments were presented in Table 2. On average the total numbers of plant

pathogenic nematodes were higher in conventional tillage than reduced and no-till treatments but the effect of tillage was not statistically significant. Among the plant pathogenic nematodes, tillage only influenced the population density of *Filenchus* spp. The population density of *Filenchus* spp. was significantly higher under conventional tillage than reduced tillage but the difference between conventional tillage and no-till was not significant (Table 3). On average the population density of *Pratylenchus neglectus* was higher than *P. thornei* but the effect of tillage was not significant in none of the two species of root-lesion nematodes.

Population density of different species and the total number of plant pathogenic nematodes under different crop residue treatments were presented in Table 3. As the results show the effect of residue retention was not significant on the population of different species and total nematode numbers.

The interaction effect of different tillage methods and different levels of residue retention on the population density of plant parasitic nematodes under rotation

system 1 is presented in Table 4. Based on the results, the population of stem and bulb nematode (*Ditylenchus* spp.), was significantly higher in conventional tillage-60% residue retention than reduced tillage-no residue and no-till-60% residue retention. The population of spiral nematodes (*Helicotylenchus* spp.), was significantly higher in reduced tillage-30% residue retention than other treatments except conventional tillage-60% residue retention and reduced tillage-no residue. The interaction effect of tillage and residue retention on the population density of other plant parasitic nematodes was not significant. The highest and lowest population density was related to *Boleodorus* spp. and *Geocenamus* spp. respectively. The interaction effect of tillage and residue retention on the total number of plant parasitic nematodes in rotation system 1 was not significant. The highest number of plant parasitic nematodes was related to the conventional tillage- no residue treatment and the lowest density was related to no-till-60% residue retention treatment.

Table 2. Effect of tillage methods on the population of plant parasitic nematodes in rotation system 1

Nematode species	Conventional tillage	Reduced tillage	No-till
<i>P. neglectus</i>	22.22 <sup>a</sup>	13.33 <sup>a</sup>	16.67 <sup>a</sup>
<i>P. thornei</i>	6.733 <sup>a</sup>	7.82 <sup>a</sup>	2.37 <sup>a</sup>
<i>Filenchus</i> sp.	31.11 <sup>a</sup>	18.89 <sup>b</sup>	21.11 <sup>ab</sup>
<i>Ditylenchus</i> sp.	41.11 <sup>a</sup>	16.73 <sup>a</sup>	22.24 <sup>a</sup>
<i>Geocenamus</i> sp.	2.378 <sup>a</sup>	1.289 <sup>a</sup>	1.28 <sup>a</sup>
<i>Boleodorus</i> sp.	66.67 <sup>a</sup>	50.00 <sup>a</sup>	43.33 <sup>a</sup>
<i>Helicotylenchus</i> sp.	1.28 <sup>a</sup>	7.88 <sup>a</sup>	0.20 <sup>a</sup>
Total	171.5 <sup>a</sup>	116.0 <sup>a</sup>	107.2 <sup>a</sup>

Means with the same letter in each row are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

Table 3. Effect of residue retention management on the population of plant parasitic nematodes in rotation system 1

Nematode species	Residue retention (%)		
	0	30	60
<i>P. neglectus</i>	21.11 <sup>a</sup>	15.56 <sup>a</sup>	15.56 <sup>a</sup>
<i>P. thornei</i>	5.64 <sup>a</sup>	5.64 <sup>a</sup>	5.64 <sup>a</sup>
<i>Filenchus</i> spp.	22.22 <sup>a</sup>	27.78 <sup>a</sup>	21.11 <sup>a</sup>
<i>Ditylenchus</i> spp.	25.60 <sup>a</sup>	26.67 <sup>a</sup>	27.82 <sup>a</sup>
<i>Geocenamus</i> spp.	1.28 <sup>a</sup>	2.37 <sup>a</sup>	1.28 <sup>a</sup>
<i>Boleodorus</i> spp.	71.11 <sup>a</sup>	47.78 <sup>a</sup>	41.11 <sup>a</sup>
<i>Helicotylenchus</i> spp.	3.48 <sup>a</sup>	4.60 <sup>a</sup>	1.28 <sup>a</sup>
Total	150.5 <sup>a</sup>	130.4 <sup>a</sup>	113.8 <sup>a</sup>

Means with the same letter in each row are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

Table 4. Interaction between tillage method and crop residue retention on the population of plant parasitic nematodes in rotation system 1 at Toroq research station

Tillage methods	Residue retention(%)	<i>P. neglectus</i>	<i>p. thornei</i>	<i>Filenchus</i> spp	<i>Ditylenchus</i> spp.	<i>Geocenamus</i> spp.	<i>Boleodorus</i> spp.	<i>Helicotylenchus</i> spp.	Total
Conventional tillage	0	23.33 <sup>a</sup>	6.73 <sup>a</sup>	23.33 <sup>a</sup>	36.67 <sup>ab</sup>	3.46 <sup>a</sup>	86.67 <sup>a</sup>	0.20 <sup>b</sup>	180.40 <sup>a</sup>
	30	20.00 <sup>a</sup>	6.73 <sup>a</sup>	43.33 <sup>a</sup>	33.33 <sup>ab</sup>	3.46 <sup>a</sup>	63.33 <sup>a</sup>	0.20 <sup>b</sup>	170.40 <sup>a</sup>
	60	23.33 <sup>a</sup>	6.73 <sup>a</sup>	26.67 <sup>a</sup>	53.33 <sup>a</sup>	0.20 <sup>a</sup>	50.00 <sup>a</sup>	3.46 <sup>ab</sup>	163.70 <sup>a</sup>
Reduced tillage	0	16.67 <sup>a</sup>	6.73 <sup>a</sup>	16.67 <sup>a</sup>	10.13 <sup>b</sup>	0.20 <sup>a</sup>	73.33 <sup>a</sup>	10.07 <sup>ab</sup>	133.80 <sup>a</sup>
	30	13.33 <sup>a</sup>	6.73 <sup>a</sup>	20.00 <sup>a</sup>	20.00 <sup>ab</sup>	0.20 <sup>a</sup>	50.00 <sup>a</sup>	13.40 <sup>a</sup>	123.70 <sup>a</sup>
	60	10.00 <sup>a</sup>	10.00 <sup>a</sup>	20.00 <sup>a</sup>	20.07 <sup>ab</sup>	3.46 <sup>a</sup>	26.67 <sup>a</sup>	0.20 <sup>b</sup>	90.40 <sup>a</sup>
No-till	0	23.33 <sup>a</sup>	3.46 <sup>a</sup>	26.67 <sup>a</sup>	30.00 <sup>ab</sup>	0.20 <sup>a</sup>	53.33 <sup>a</sup>	0.20 <sup>b</sup>	137.2 <sup>a</sup>
	30	13.33 <sup>a</sup>	3.46 <sup>a</sup>	20.00 <sup>a</sup>	26.67 <sup>ab</sup>	3.46 <sup>a</sup>	30.00 <sup>a</sup>	0.20 <sup>b</sup>	97.13 <sup>a</sup>
	60	13.33 <sup>a</sup>	0.20 <sup>a</sup>	16.67 <sup>a</sup>	10.07 <sup>b</sup>	0.20 <sup>a</sup>	46.67 <sup>a</sup>	0.20 <sup>b</sup>	87.33 <sup>a</sup>
Total	156.65	50.77	213.34	240.27	14.84	480.00	28.13	1184.06	

Means with the same letter in each column are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

#### Population densities of plant pathogenic nematodes in rotation system 2

In the rotation system 2, in addition to the nematodes detected in rotation system 1, the pin nematode (*Paratylenchus* spp.) was also detected and quantified. The results of analysis of variance showed that the effect of tillage on population density of the root-lesion nematode, *Pratylenchus thornei*, *Filenchus* spp. and *Geocenamus* spp. was statistically significant ( $P=0.05$ ). The effect of crop residue retention on the population density of *Geocenamus* spp. ( $P=0.01$ ) and the interaction effect of tillage  $\times$  residue retention on population density of the root-lesion nematode, *Pratylenchus neglectus* ( $P=0.05$ ) and *Geocenamus* spp. ( $P=0.01$ ) were statistically significant but the effect of tillage, residue retention and their interaction on the population density of other nematodes and total nematode numbers were not statistically significant. The population density of plant pathogenic nematodes under different tillage treatments were presented in Table 5. On average the highest density of nematodes was related to no-tillage treatment and the lowest density was related to conventional tillage, however the effect of tillage was not significant. The population density of *Pratylenchus thornei* in no-till treatment was statistically higher than conventional tillage and reduced tillage treatments but tillage had no effect on population density of *Pratylenchus neglectus*. The population density of *Filenchus* spp. was statistically

higher in reduced tillage than no-till and conventional tillage. Tillage had no effect on the population density of other nematode species and total nematode numbers.

Population density of plant pathogenic nematodes under different crop residue treatments in rotation system 2 were presented in Table 6. Based on the results, the population of *Geocenamus* spp. in no-residue treatment was statistically higher than the conventional tillage and reduced tillage treatments. Crop residue retention had no effect of the population of other nematode species and total nematode numbers.

The interaction effect of tillage and residue retention on the population density of plant parasitic nematodes under rotation system 2 is presented in Table 7. The result showed that interaction between tillage and residue retention significantly influenced the population of both species of root-lesion nematode, *Pratylenchus neglectus* and *P. thornei* and also *Geocenamus* spp. On average the population of *P. thornei* was higher than *P. neglectus*. The interaction of tillage  $\times$  residue retention on the population density of other nematode species and total number of plant parasitic nematodes in rotation system 2 was not significant. The highest and lowest population density was related to *Boleodorus* spp. and *Helicotylenchus* spp. respectively. The highest number of plant parasitic nematodes was related to the conventional tillage-60%

residue retention and the lowest density was related to no-till-no-residue treatment.

Table 5. Effect of tillage methods on the population of plant parasitic nematodes in rotation system 2

Nematode species	Conventional tillage	Reduced tillage	No-till
<i>P. neglectus</i>	16.67 <sup>a</sup>	21.11 <sup>a</sup>	28.89 <sup>a</sup>
<i>P. thornei</i>	2.378 <sup>b</sup>	3.46 <sup>b</sup>	12.27 <sup>a</sup>
<i>Paratylenchus</i> spp.	2.378 <sup>a</sup>	6.77 <sup>a</sup>	2.40 <sup>a</sup>
<i>Filenchus</i> spp.	1.28 <sup>b</sup>	9.00 <sup>a</sup>	2.40 <sup>b</sup>
<i>Ditylenchus</i> spp.	6.73 <sup>a</sup>	11.20 <sup>a</sup>	4.55 <sup>a</sup>
<i>Geocenamus</i> spp.	3.48 <sup>a</sup>	4.55 <sup>a</sup>	38.93 <sup>b</sup>
<i>Boleodorus</i> spp.	22.22 <sup>a</sup>	33.40 <sup>a</sup>	20.04 <sup>a</sup>
<i>Helicotylenchus</i> spp.	4.55 <sup>a</sup>	4.57 <sup>a</sup>	0.20 <sup>a</sup>
Total	59.71 <sup>a</sup>	94.09 <sup>a</sup>	109.7 <sup>a</sup>

Means with the same letter in each row are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

Table 6. Effect of residue retention management on the population of plant parasitic nematodes in rotation system 2

Nematode species	Residue retention (%)		
	0	30	60
<i>P. neglectus</i>	27.78 <sup>a</sup>	21.11 <sup>a</sup>	17.78 <sup>a</sup>
<i>P. thornei</i>	5.66 <sup>a</sup>	6.75 <sup>a</sup>	5.68 <sup>a</sup>
<i>Paratylenchus</i> spp.	3.48 <sup>a</sup>	6.77 <sup>a</sup>	1.28 <sup>a</sup>
<i>Filenchus</i> spp.	4.57 <sup>a</sup>	2.40 <sup>a</sup>	5.71 <sup>a</sup>
<i>Ditylenchus</i> spp.	6.73 <sup>a</sup>	11.18 <sup>a</sup>	4.57 <sup>a</sup>
<i>Geocenamus</i> spp.	36.71 <sup>a</sup>	6.75 <sup>b</sup>	3.51 <sup>b</sup>
<i>Boleodorus</i> spp.	24.44 <sup>a</sup>	22.29 <sup>a</sup>	28.93 <sup>a</sup>
<i>Helicotylenchus</i> spp.	1.28 <sup>a</sup>	3.48 <sup>a</sup>	4.55 <sup>a</sup>
Total	110.7 <sup>a</sup>	80.76 <sup>a</sup>	72.04 <sup>a</sup>

Means with the same letter in each row are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

Table 7. Interaction between tillage method and crop residue retention on the population of plant parasitic nematodes in rotation system 2

Tillage methods	Residue retention (%)	<i>P. neglectus</i>	<i>P. thornei</i>	<i>Paratylenchus</i> spp.	<i>Filenchus</i> spp.	<i>Ditylenchus</i> spp.	<i>Geocenamus</i> spp.	<i>Boleodorus</i> spp.	<i>Helicotylenchus</i> spp.	Total
Conventional tillage	0	0.20 <sup>b</sup>	13.33 <sup>bc</sup>	3.46 <sup>a</sup>	3.46 <sup>a</sup>	6.73 <sup>a</sup>	10.07 <sup>b</sup>	20.0 <sup>a</sup>	3.46 <sup>a</sup>	60.73 <sup>a</sup>
	30	6.7 <sup>ab</sup>	20.00 <sup>bc</sup>	3.46 <sup>a</sup>	0.20 <sup>a</sup>	10.0 <sup>a</sup>	0.20 <sup>b</sup>	23.3 <sup>a</sup>	3.46 <sup>a</sup>	67.40 <sup>a</sup>
	60	0.20 <sup>b</sup>	16.67 <sup>bc</sup>	0.20 <sup>a</sup>	0.20 <sup>a</sup>	3.46 <sup>a</sup>	0.20 <sup>b</sup>	23.33 <sup>a</sup>	6.73 <sup>a</sup>	51.00 <sup>a</sup>
Reduced tillage	0	6.7 <sup>ab</sup>	43.33 <sup>a</sup>	6.80 <sup>a</sup>	10.07 <sup>a</sup>	10.0 <sup>a</sup>	6.73 <sup>b</sup>	36.67 <sup>a</sup>	0.20 <sup>a</sup>	120.5 <sup>a</sup>
	30	3.46 <sup>ab</sup>	10.00 <sup>c</sup>	10.07 <sup>a</sup>	6.800 <sup>a</sup>	16.80 <sup>a</sup>	6.73 <sup>b</sup>	26.80 <sup>a</sup>	6.80 <sup>a</sup>	87.47 <sup>a</sup>
	60	0.20 <sup>b</sup>	10.00 <sup>c</sup>	3.46 <sup>a</sup>	10.13 <sup>a</sup>	6.80 <sup>a</sup>	0.20 <sup>b</sup>	36.73 <sup>a</sup>	6.73 <sup>b</sup>	74.27 <sup>a</sup>
No-till	0	10.07 <sup>ab</sup>	26.67 <sup>abc</sup>	0.20 <sup>a</sup>	0.20 <sup>a</sup>	3.46 <sup>a</sup>	93.33 <sup>a</sup>	16.67 <sup>a</sup>	0.20 <sup>a</sup>	150.8 <sup>a</sup>
	30	10.07 <sup>ab</sup>	33.33 <sup>ab</sup>	6.80 <sup>a</sup>	0.20 <sup>a</sup>	6.73 <sup>a</sup>	13.33 <sup>b</sup>	16.73 <sup>a</sup>	0.20 <sup>a</sup>	87.40 <sup>a</sup>
	60	16.67 <sup>a</sup>	26.67 <sup>abc</sup>	0.20 <sup>a</sup>	6.80 <sup>a</sup>	3.46 <sup>a</sup>	13.13 <sup>b</sup>	26.73 <sup>a</sup>	0.20 <sup>a</sup>	90.87 <sup>a</sup>
Total		53.67	200.0	34.0	37.46	67.44	143.32	226.96	27.18	790.44

Means with the same letter in each column are not significantly different from each other at  $\alpha=0.05$  based on Duncan's Multiple Range Test

## DISCUSSION

**Tillage effect on nematode density:** Tillage effects on density of different nematode species was different based on the crop rotation sequence and the level of residue retention. In the rotation system 1 (wheat, maize, wheat, melon and wheat ), tillage influenced the population density of *Filenchus* spp. and in the rotation system 2 (wheat, canola, wheat, Persian clover, tomato and wheat), tillage had a significant effect on the population density of root lesion nematode (*Pratylenchus thornei*), *Filenchus* spp. and *Geocenamus* spp., however, tillage had no significant effect on the population density of other nematode species and total number of plant parasitic nematodes in both crop rotation systems. The impact of decreased level of tillage intensity from conventional tillage to reduced tillage and no-till treatments was also dependent to nematode species and crop rotation sequence. The population density of *Filenchus* spp. in the rotation system 1, was significantly lower in reduced tillage than the conventional tillage and no-till treatments, however in the rotation system 2, the population density of this species was significantly higher in reduced tillage than the conventional tillage and no-till treatments. The population density of *Geocenamus* spp. and *P. thornei*, the root-lesion nematode, was significantly increased when tillage intensity decreased from conventional tillage to no-till.

The impact of tillage individually or in combination with other conservation agriculture practices have been studied by other researchers but controversial results has been reported. Some studies report an increase in the population density of s plant parasitic nematodes, while others report a decrease.

An early study by López-Fando and Bello (1995) on the effect of tillage and crop rotation on the soil nematode fauna showed that the populations of endo-parasitic plant nematodes including *Heterodera avenae* and *Pratylenchus* spp., decreased in no-tillage systems compared to conventional tillage. Westphal et al. (2009) studied the effect of tillage on the population density of *Heterodera glycines*, the soybean cyst nematode, in the maize-soybean rotation system and reported that decreasing in the population densities of *H. glycines* was associated with reducing tillage intensity from plowing + secondary tillage to no-tillage practice. Ramakrishna and

Sharma (1998) reported that no-tillage increases the population density and diversity of beneficial soil microorganisms that can reduce the population of plant pathogenic organisms in soil.

Severe reduction in population of plant parasitic nematodes including the rice-root nematode (*Hirschmannella oryzae*) and the stunt nematode (*Tylenchorhynchus* spp.) reported in no-tillage compared to conventional tillage in the rice-wheat rotation system in India (Singh et al., 2005)

Ahmed et al. (2012) compared the effect of conventional tillage and zero-tillage systems under the long-term rotation trials on the incidence of soybean cyst nematodes (*Heterodera glycines*) on chickpea and lentil, and reported that tillage had no significant effect. Mashavakure et al. (2018) studied the effect of tillage and fertilizer treatments on soil and root borne nematodes of maize and reported that plant-parasitic nematodes showed differential responses to different tillage systems. Significant interaction effect of tillage treatments on the population density of *Pratylenchus* in different crop rotation systems is also reported by Okada and Harada (2007).

Increase in nematode population has been reported in the rice-wheat rotation sequence due to adoption of no-till systems (Duveiller et al., 2004). Govaerts (2007) reported that the farmer practice in Mexico including conventional tillage with continuous maize monoculture and residue removal dramatically increased the population of the root-lesion nematode, *P. thorne* but nematode populations were low at Zero tillage in the same cropping system.

**Residue retention effect on nematode density:** The effect of residue retention on the population density of nematodes species was different in the two rotation sequence. Residue retention influenced the population of *Geocenamus* spp. and interactions of tillage and residue retention effects were found for the population of root lesion nematode (*Pratylenchus neglectus*) and *Geocenamus* spp. in rotation sequence 1, but the effect of tillage, residue retention and their interaction on the population of other nematodes and total nematode numbers were not significant.

There has been no general trend on the effect of residue retention alone or in combination with other conservation agricultural practices on the population

density of soil microorganisms including plant pathogenic fungi and nematodes in literatures. The effect of different levels of crop residue retention, different levels of tillage intensity and crop rotations on nematode populations has been studied by Govaerts *et al.* (2007), in the CIMMYT long-term sustainability field trials and concluded that residue retention in comparison with residue removal and zero tillage in comparison with conventional tillage reduced the population density of the root-lesion nematode, *P. thornei* due to higher number of microbial diversities in both maize and wheat crops. *P. thornei* is reported as an economically important nematode on wheat that may yield losses of up to 40% in north and central Mexico (Nicol and Ortiz-Monasterio, 2004).

Several authors have asserted that residue retention and no-till practices compared to conventional treatments plays a direct role in increasing the population of soil fauna including bacteria, actinomycetes, fungi, earthworms and nematodes and their biological activity in soil (Lupwayi *et al.*, 2001; Spedding *et al.*, 2004). It is generally argued that higher diversity of soil microbial communities increase the soil health and the capacity of soil to respond more efficiently to agricultural interventions under different environmental conditions including an increased level of resilience (Kibblewhite *et al.*, 2008). However, several studies have found that conservation agriculture practices are associated with an increase in plant parasitic nematodes and fungal diseases that preserve in the stubble. On the other hand, there are evidences that appropriate rotation sequences can break the life cycles of stubble-borne disease and some pests (Thierfelder *et al.*, 2013). The peanut and soybean root-knot nematode (*Meloidogyne* spp.), reniform nematode (*Rotylenchulus reniformis*) and soybean cyst nematode (*Heterodera glycines*) infestations may decline following the non-host bahiagrass (Johnson *et al.*, 2000; Katsvairo *et al.*, 2006). Improving the soil biological activity using organic amendments, plant residue and tillage practices decreased the population density of root-knot nematode (*Meloidogyne incognita*) (Stirling and Eden, 2008).

In general it can be concluded that, regular monitoring of plant diseases including plant parasitic nematodes is required under CA farming systems. A variety of suitable agronomic practices including appropriate rotation sequence, field hygiene and sanitation and nutrient

balance can help reduce the pressure of plant disease under CA.

#### CONCLUSION

The conservation agricultural practices are influencing the population density of plant parasitic nematodes; however their effects depend on the nematode species and the crop rotation sequence. In general, it is difficult to develop control methods for the soil-borne plant pathogens, including nematodes, so the effect of conservation agricultural practices on major soil-borne pathogens should be studied at local level before introducing a new cropping system to medium and large scale farmers. The results of this study indicated that the conservation agriculture farming system have no significant influence on the population density of major plant parasitic nematodes under the wheat, maize, wheat, melon, wheat and wheat, canola, wheat, Persian clover, tomato, wheat rotation sequences and do not increase the risk of damage by nematodes.

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|-------------------|---|--|
| Reza Aghnoum      | : | Conduct research and write manuscript. |
| Ahmad Z. Fizabadi | : | Review manuscript and conduct research |