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Changes of nematode community under monoculture wheat and wheat/jujube intercropping system in Xinjiang, Northwest China

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Summary

Nematode communities in the soils of wheat (*Triticum aestivum* Linn.) rhizosphere grown alone and grown in jujube (*Ziziphus jujuba* Mill.) orchard were investigated for three years in Hetian arid area, Xingjiang Uygur Autonomous Region, northwest of China. The results showed that eu-dominant families were Rhabditidae, Cephalobidae and Aphelenchidae among 15 families and 19 genera. Nematode abundance in wheat rhizosphere soil was smaller in wheat/jujube intercropping system, mainly because of lower numbers of bacterial feeders and fungal feeders. Besides, the nematode numbers of cp-1 and cp-2 (cp, colonizer-persister) guilds were significantly lower in wheat/jujube intercropping system than that in monoculture wheat system, due to the markedly lower numbers of Rhabditidae and Cephalobidae, although those of cp-3 and cp-4 guilds had no significant differences between monoculture and intercropping systems. Shannon-Weaver index (*H'*), genus dominance index (*Ig*) and structural index (*SI*), represented soil food web diversity and structure, had no differences between monoculture and intercropping systems. Significantly lower values of Wasilewska index (*WI*) and *PPI*/*MI* in monoculture wheat than in intercropping system. It was concluded that the soil status in monoculture wheat system exhibited better soil ecosystem in compared with wheat/jujube intercropping system.
Keywords: soil nematodes; colonizer-persister; biodiversity; maturity index; soil food web

Introduction

Although Xinjiang Uygur Autonomous Region account 17 % national territory with unique light and heat resources, still only 5 % arable land has been used effectively due to the desert climate (Liu *et al.*, 2012). The consensus of planted variety crops in the limited land was more popular for the local farmers. Wheat (*Triticum aestivum* Linn.) is large-scale cultivated in Xingjiang, the most important area of winter wheat production in China. Nowadays wheat/jujube (*Ziziphus jujuba* Mill.) intercropping system are widely employed because it is benefit for soil and water conservation and improvement of environment (Zhao & Lu, 2004).
Wheat/jujube intercropping system in Hetian improved total output and efficiency of land use, but decreased root length density and root surface area density (Zhang *et al.*, 2013). Intercropping wheat with jujube obviously improved soil physical properties, soil microorganisms and soil nutrient. In addition, the disease occurrence of dry-hot winds in wheat was decreased when grown in jujube orchard (Chang *et al.*, 2012). Other studies about intercropping focused on its important cultural practices in pest control, and found that natural enemies were build up and insect pests were reduced in intercropping castors (Rao *et al.*, 2012). In southern Africa, intercropping with sugar bean, peanut and sweet potato could reduce *Meloidogyne javanica* and *Pratylenchus zeae* infestation of sugarcane sett roots (Berry *et al.*, 2009). India farmers grew marigold plants in land bordering vegetable crops or between the rows in order to control plant parasitic nematodes in lower level (Siddiqui & Alam, 1987). However, there still is lack of data on long time changes of wheat rhizosphere soil nematode communities in wheat/jujube intercropping system.
Soil nematodes are very abundance and diverse in all soils, feeding on a wide range of soil organisms, the movement of soil nem-

atodes depend on the continuity of soil water, which can reflect disturbances rapidly (Yeates & Bongers, 1999). Besides, soil nematodes play a key role in the decomposition of soil organic matter, mineralization of plant nutrients as well as nutrient cycling, by regulating soil bacterial and fungal populations and their metabolites (Neher, 2001). It is apparently that soil nematode is one of the most influential and sensitive bioindicator of soil health. Interpretation of nematode community structure offer excellent opportunities to assess the condition of soils, and to monitor changes in the structure and function of the detritus food web of cultivation and land use changes (Bernard, 1992; Santorufo *et al.*, 2012). There are many studies on nematode communities as bioindicator, such as continuous cropping, organic cropping, fertilizer strategies, cultivation, rotation, different plant, grassland soil, urban soil and sand dune, soil recovery in tailings of a lead/zinc mine, chemical pollution, even river pollution (Ekschmitt *et al.*, 2001; Tomar *et al.*, 2009; Hu & Qi, 2010; Pan *et al.*, 2010; Villenave *et al.*, 2010; Ugarte *et al.*, 2013; Li *et al.*, 2014).

Jujube trees and wheat are of importance as the main economic and food crops in southern Xinjiang. However, no systematic study has been carried out on the nematodes characters of wheat/jujube and sole jujube systems. So the present study was aimed to characterize the nematode community structure of wheat grown alone field and grown in jujube orchard field to provide an evaluation criterion for further clarifying the mechanism of its difference between monoculture and inter-cropping systems. The objectives of our study in detail were to: 1) Characterize and compare nematode communities in wheat rhizosphere soils under monoculture and intercropping system 2) Evaluate soil food web condition in wheat rhizosphere by means of nematode faunal response to different cropping systems; 3) Determine the soil status of wheat rhizosphere that planted in jujube orchard.

Material and Methods

This study was conducted at a wheat field which was set up in 2009 at Hetian Agricultural Scientific Research Institute (37°12' N, 79°94' E), Agro-Tech Extension and Service Center of Hetian Prefecture, Xinjiang Uygur Autonomous Region, China. Average annual precipitation is only 35 mm, the potential evaporation is up to 2480 mm. The annual sunshine time is 2470 – 3000 h, with total solar radiation is 6627 MJ m^{-2} $year^{-1}$, and annual mean temperature is 13.7 °C, with effective temperature more than 10 °C reach to 4200 °C. The region has a typical arid climate. And the soil at the site is classified as an Arenosol in the classification system of the Food and Agriculture Organization (FAO).

The experimental design was a single factor field experiment, comprising three replicates of wheat grown alone and three replicates of wheat grown in 6-year-old jujube orchard. Wheat was sown in October, harvest in June next year. Sole wheat was planted 0.15 m inter-row and within rows distance. Intercropping system was designed as a replacement series, including 12 rows wheat of 0.15 m inter-row distance plus 2 rows of 6-year-old jujube trees, the distance between jujube trees and nearest wheat row was 0.6 m. The 6-year-old jujube tree occupied 40 % of the intercropped area and wheat 60 %. The density of intercropped wheat was 11,250,000 plants ha^{-1}. All the three pairs of the treatments plots (12 m × 40 m) have undergone the same management consistently. All plots were given identical applications of urea (N) at 450 kg ha^{-1}, and diammonium phosphate (P) at 30 kg ha^{-1}. All the P fertilizer and a half of the N were broadcast evenly and incorporated into the top 20 cm of the soil prior to sowing. The remaining half of the N fertilizer was applied at the elongation stage. Irrigation was carried out on four occasions on March 25, April 14, May 2, and May 20. Each irrigation application consisted of 90 mm (900 m^3 ha^{-1}). The irrigation practice followed that recommended to farmers by local agronomists. Soil samples were collected during the growing season in May 23, 2011, May 29, 2012 and May 25, 2013, with the wheat in the booting stage and jujube in the sprouting-leaf unfolding stage. Three soil replicate samples (altogether thirty cores) were collected per each treatment plot (12m × 40 m), that means per wheat alone and per wheat grown in jujube orchard, with the checkerboard sampling method. Soils from ten cores were mixed to constitute one sample. Each core was taken by using geotome (i. d. 25 mm) to a depth of 20 cm.

The samples were stored in individual plastic bags, and immediately transferred to a 4 °C cold storage. All samples were processed within 7 days of collection. Soil moisture was determined by drying the samples at 105 °C for 8 h. Soil pH value was measured in a paste of air-dried soil to solution at a ratio of 1:2 in KCl (1 mol/L) by glass electrode. Soil organic matter content was determined by burning dried soil in a muffle furnace at 490 °C for 8 h. Total nitrogen was measured following micro-Kjeldahl digestion (Bremner & Mulvaney, 1982). The experiment soil fundamental state was shown in Table.1.

The soil nematodes were extracted from 100g fresh soil, using sugar flotation and centrifugation method (Barker *et al.*, 1985). Extracted nematodes were killed at 60 °C and fixed in 5 % FA (Formalin acetic acid) for genus identification (Griffiths *et al.* 1990). The number of nematodes was counted and individuals were identified to genus level, using an upright differential interference microscope (LEICA).

The characteristics of the nematode communities were described by the following approaches: 1) total nematode abundance (individuals per 100g dry soil). 2) The classification of trophic groups based on known feeding habits or stoma and esophageal morphology was assigned to: a) Bacterial feeders (BF), b) Fungal feeders (FF), c) Plant feeders (PF), and d) Omnivores-predators

Table 1. Soil environmental conditions under wheat grown alone and grown in jujube orchard field

Sample	Soil moisture (%)	pH value	Organic matter (g/kg)	Total N (g/kg)	C/N
W	10.99	8.00	14.32	0.87	16.49
JW	13.81	8.11	19.69	0.69	28.48

W: wheat grown alone, JW: wheat grown in jujube orchard

(Om) (Bongers, 1990; Yeates *et al.*, 1993). 3) The classification of nematode colonizer-persister (cp) values based on life strategies according to Bongers (Bongers, 1990; Bongers & Bongers, 1998; Bongers, 1999). 4) Diversity index: a) Shannon-Weaver index (*H'*): $H' = -\sum Pi \ln Pi$, where *pi* is the proportion of each taxon in the total population (Shannon & Weaver, 1949), b) Genus dominance index (*Ig*): $Ig = \sum Pi^2$, where *pi* is the proportion of individuals in the *i*-th taxon (Simpson, 1949), c) Evenness index (*J'*): $J' = H' / \ln(S)$ (Pielou, 1975), d) Richness index (*SR*): $SR = (S\text{-}1) / \ln(N)$, where *S* is the number of taxa and *N* is the number of individuals identified (Yeates & King, 1997), e) Trophic diversity index (*TD*): $TD = 1 / \sum Ti^2$, where *Ti* is the proportion of the trophic group *i* in the nematode community (Heip *et al.*, 1988). 5) Functional index: a) Nematode Channel Ratio (*NCR*): $NCR = BF / (BF + FF)$ (Yeates 2003), b) Channel index *(CI)*, Basal Index (*BI*), Structure Index (*SI*), Enrichment Index (*EI*) according to Ferris et al. (Ferris *et al.* 2001, Ferris&Matute 2003), c) Wasilewska Index (*WI*): $WI = (BF + FF) / PP$, the ratio of bacterial feeders plus fungal feeders to plant parasites (Wasilewska 1994). 6) Maturity index: a) Plant parasites index (*PPI*): $PPI = \sum ui\ fi'$, where *vi* is the c-p value for the plant-parasitic nematodes to the *i*-th nematode genus and *fi* is the proportion of genus in the plant-parasitic nematode community (Bongers 1990, Bongers *et al.* 1997), b) Maturity index (*MI*): $MI = \sum ui\ fi$, where *vi* is the c-p value for free-living nematodes to the *i*-th nematode genus and *fi'* is the proportion of the genus in the free-living nematode community (Bongers, 1990; Bongers, 1999), c) $PPI/MI = \sum ui\ fi' / \sum ui\ fi$ (Bongers *et al.* 1997), d) $\sum MI = \sum ui\ Pi$, modified maturity index, where *vi* is the c-p value for free-living and plant parasitic nematodes to the *i*-th nematode genus and pi is the proportion of the genus in the whole nematode community (Yeates, 1994).

All the data were analyzed through ANOVA by IBM SPSS Statistic v18.0 software package. Mean value (three replicates × three years, n = 9) were compared by paired sampled *t*-test, and difference at $p < 0.05$ level were considered as statistically significant.

Table 2. Families and respective genera of nematodes found in wheat rhizosphere by grown alone and in jujube orchard

Families	W	JW	Genera	W	JW
Rhabditidae	+++++	+++++	*Rhabditis*	X	X
			Diploscapter	X	X
Cephalobidae	+++++	+++++	*Cephalobus*	X	X
			Eucephalobus	X	X
			Acrobeles	X	X
			Acrobeloides	X	X
Plectidae	-	+	*Wilsonema*	O	X
Monhysteridae	-	+	*Monhystera*	O	X
Prismatolaimidae	+	-	*Prismatolaimus*	X	O
Alaimidae	++	+	*Alaimus*	X	X
Aphelenchoididae	+++	+	*Aphelenchoides*	X	X
Aphelenchidae	++++	+++++	*Aphelenchus*	X	X
Diphtherophoridae	-	+	*Diphtherophora*	O	X
Tylencholaimidae	+	-	*Tylencholaimus*	X	O
Mononchidae	+	-	*Mononchus*	X	O
Dorylaimidae	++	+	*Dorylaimus*	X	X
Tylenchidae	+++	++++	*Tylenchus*	X	X
Paratylenchidae	+	+	*Paratylenchus*	X	X
Pratylenchidae	+	+	*Hirschmanniella*	X	X

Note: X = present; O = absent. W: wheat grown alone, JW: wheat grown in jujube orchard. +++++: Eu-dominant (≥10%); ++++: Dominant (5 % – 10 %); +++: Sub-dominant (2 % – 5 %); ++: Resident (1 % – 2 %); +: Sub-resident (0 % – 1 %); -: Absent (0%)

Results

A total number of 15 families and 19 genera were present in W and JW, with only 3 families (Rhabditidae, Cephalobidae, and Aphelenchidae) occurring in common (>10 %). Bacterial feeders dominated nematode communities in both genus composition and proportion (85.22 % in W and 81 % in JW), consisting of ten genera, *Rhabditis*, *Diploscapter*, *Cephalobus*, *Eucephalobus*, *Acrobeles*, *Acrobeloides*, *Wilsonema*, *Monhystera*, *Prismatolaimus*, and *Alaimus*. However, *Wilsonema* and *Monhystera* were not found in W, while *Prismatolaimus* was not found in JW. Bacterial feeders that significantly contributing to this trend were Rhabditidae and Cephalobidae, which belong to eu-dominant families. Fungal feeders consisted of four genera, *Aphelenchoides*, *Aphelenchus*, *Diphtherophora*, and *Tylencholaimus*, respectively. Their contributions to the total nematode communities were 9.92 %, 11.56 % under W and JW. *Diphtherophora* was not found in W, while *Tylencholaimus* was not found in JW. Fungal feeders that significantly contributed to this trend were Aphelenchidae and Aphelenchoididae. Aphelenchidae was eu-dominant family in JW, but dominant in W; Aphelenchoididae was sub-resident family in JW, and sub-dominant in W (Table 2). Plant feeders consisted of three genera, *Tylenchus*, *Paratylenchus*, and *Hirschmanniella*, Their contributions to the total nematode communities were 3.41 %, 6.82 % under W and JW, respectively. Omnivore-predators consisted of *Mononchus* and *Dorylaimus*, occupied 1.45 %, 0.62 % of the total nematode communities under W and JW, and *Mononchus* was not found in JW (Table 2).

Rhabditidae, Cephalobidae, and Aphelenchidae were the most abundant families in W soil nematode community, thus were responsible for the trend of different treatment changes. Cephalobidae occupied 54 % of total nematodes in W and increased to 59 % in JW; Rhabditidae occupied 29 % in W and reduced to 22 % in JW; Aphelenchidae occupied 8 % in W and increased to 10 % in JW. Other families account for only 9 % of total nematodes in W and JW (Fig. 1). Mean density of Rhabditidae and Cephalobidae were significantly higher in W than JW, while that of Aphelenchidae did not significant differed between W and JW (p>0.05) (Table 3).

The mean density of total nematode in W was higher than that in JW, mainly reflected in the greater number of bacterial feeders in W. No significant difference of the mean density of fungal feeders and omnivores-predators were found between W and JW. Whereas, plant feeders were increased in JW in comparison with those in W. In W soil mean number of cp-1 colonizers was 1.79 times greater than that in JW, in addition, the mean number of cp-2 groups was 1.20 times greater than that in JW. On the other side,

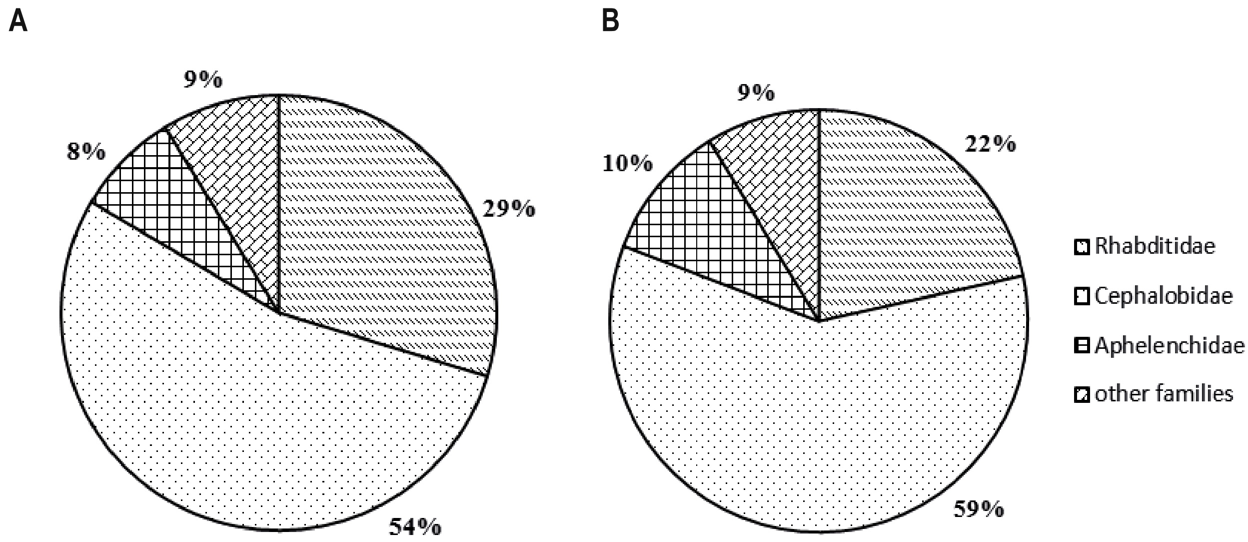

Fig. 1. Percentage of most abundant families in wheat rhizosphere soil nematodes by grown alone (A) and grown in jujube orchard (B)

there was no significant difference found in cp-3 and cp-4 groups (Table 3).

No significantly effects were found in Shannon index (*H'*), genus dominance (*Ig*), evenness (*J'*), nematode channel ratio (*NCR*), channel index (*CI*), basal index (*BI*), structure index (*SI*), enrichment index (*EI*), plant parasitic nematode maturity index (*PPI*), maturity index (*MI*) and modified maturity index (∑*MI*). But significant effects were found in species richness (*SR*), trophic diversity (*TD*), Wasilewska index (*WI)*, and the ratio of plant parasitic nematode maturity index to maturity index (*PPI/MI*). *SR* and *TD* were higher in JW than W, while *WI* and *PPI/MI* were lower in JW than W (Table 4).

Discussion

Soil nematode communities varied in wheat rhizosphere grown alone and grown in jujube orchard. *Wilsonema* (cp-2) and *Monhystera* (cp-2) *Diphtherophora* (cp-3) were only found in JW, while *Prismatolaimus* (cp-3) *Tylencholaimus* (cp-4) *Mononchus* (cp-4) was only found in W. In addition, *Wilsonema* is known as aquatic nematodes favoring wetter habitats and mulch-influenced environments (Porazinska *et al.* 1999). Besides, *Wilsonema* species live in soils that is rich in organic matter, and some of them actually prefer wet sites whereas others tolerate drier soils. So the two factors could act in favour of *Wilsonena* in JW with more organic matter (Table 1).

Table 3. Density of (a) total nematode populations, (b) nematode feeding groups, (c) colonizer-persister groups, and (d) abundant families in wheat rhizosphere by grown alone and grown in jujube orchard

	2011		2012		2013		Mean		
	W	JW	W	JW	W	JW	W	JW	Sig.
(a) Total nematodes	2187	1164	2052	1950	2462	1809	2234	1641	*
(b) Feeding groups:									
Bacterial feeders	1885	945	1685	1556	2151	1485	1907	1329	*
Fungal feeders	252	153	213	217	193	187	219	186	ns
Omnivore-predatorS	0	5	80	20	11	7	30	11	ns
Plant feeders	50	61	74	157	107	130	77	116	*
(c) Colonizer-persister									
cp - 1	682	78	239	371	1142	706	688	385	*
cp - 2	1492	1072	1613	1515	1300	1093	1468	1227	*
cp - 3	13	9	4	32	9	3	9	15	ns
cp - 4	0	5	196	32	11	7	69	15	ns
(d) Abundant families									
Rhabditidae	682	78	239	371	1142	706	688	385	*
Cephalobidae	1203	867	1342	1173	1008	778	1184	939	*
Aphelenchidae	198	143	190	182	121	175	170	167	ns

ns = No significant difference, * refers to significant difference between the W and JW at 0.05 level by paired sample t test.
W: wheat grown alone, JW: wheat grown in jujube orchard

Table 4. Nematode community parameters: diversity index (a), functional index (b), maturity index (c) under grown alone and grown in jujube orchard

	W	JW	Sig.
Diversity index (a)			
H'	1.55	1.44	ns
Ig	0.29	0.36	ns
J'	0.66	0.59	ns
SR	1.50	1.68	*
TD	1.36	1.48	*
Functional index (b)			
NCR	0.90	0.88	ns
CI	9.99	17.09	ns
BI	32.43	44.86	ns
SI	12.43	5.50	ns
EI	63.25	53.90	ns
WI	31.20	14.21	*
Maturity index (c)			
PPI	3.07	2.08	ns
MI	1.74	1.79	ns
PPI/MI	1.71	1.17	*
∑MI	1.78	1.81	ns

ns = No significant difference, * refers to significant difference between the W and JW at 0.05 level by paired sample t test. W: wheat grown alone, JW: wheat grown in jujube orchard

Prismatolaimus (cp-3) *Tylencholaimus* (cp-4) *Mononchus* (cp-4) was only found in W, it is indicated that the wheat rhizosphere soil in wheat/jujube intercropping system was less suitable for some genera with cp-3 and cp-4 guilds. This result is consistent with previous study which suggested that the density of cp-3, cp-4, cp-5 nematodes were negatively correlating with the pollutants (Shao *et al.*, 2008; Korthals *et al.*, 2009).

The lower number of total nematodes in JW, mainly reflected in less abundance of bacterivores and less abundance of cp-1 and cp-2 groups in JW. Rhabditidae and Cephalobidae were the most abundance families in wheat rhizosphere nematode community in monoculture wheat system and jujube/wheat intercropping system, occupying above 20 % and 50 %, respectively. The absolute abundance of *Rhabditis* and *Acrobeloides* were higher in long-term turfgrass management practices and a field grown tomatoes (Nahar *et al.*, 2006; Cheng *et al.*, 2008). Moreover, *Acrobeles* and *Acrobeloides* were also more numerous in tomatoes grown in conventional than in organic systems (Ferris *et al.*, 1997). At the generic or family level, Rhabditidae was the enrichment opportunists, showed a typical r-selected behavior, with sudden and temporal population increase (within 3 – 4 weeks), high fecundity, large amount of eggs and high standard metabolism under food-rich conditions (Bongers & Bongers, 1998). Cephalobidae belonged to cp-2 category which was associated with dry conditions in the soil environment (Griffiths *et al.*, 1995). *Rhabditis* and *Acrobeloides* were co-existing species, which had a significant effect on ecosystem function through the creation of additional consumer, decomposer, and symbiotic organism niches. They should be influenced by the root exudates of jujube trees, how and which root exudates influence the soil microorganism and nematodes need the further studies. Lower numbers of Rhabditidae and Cephalobidae means lower fertility of soil and slower decomposition rate of soil food web, which is not benefit for the plant growth.

Shannon index (*H'*) associated with genus dominance (*Ig*) indicated the diversity of soil detrital food web, with *H'* giving more weight to rare species, while *Ig* giving greater weight to common species (Freckman & Ettema, 1993). In detail, higher *H'* and lower *Ig* indicated greater diversity (Simpson, 1949). *J'* and *SR* indicate the evenness and species richness (Yeates & King, 1997). Trophic diversity (*TD*) describes diversity of functional groups within the nematode community (Freckman & Ettema, 1993). Values of *TD* and *SR* were significantly increased in JW, mainly because of lower percentages of bacterivores. No significant differences in the rest of diversity indices between the two systems. It is concluded that wheat/jujube intercropping system had relatively less affect on the diversity in wheat rhizosphere soil food web.

Ecological indices such as basal index (*BI*), channel index (*CI*), structural index (*SI*), and enrichment index (*EI*) may provide insight to whether the nematode community structure is in stressed, nutrients enriched, stable structured or decomposition environments, and provide information on the dynamics of the soil food web (Ferris *et al.*, 2001). Moreover, nematode channel index (*NCR*) indicated the decomposition processes in soil (Yeates, 2003). Unfortunately, these indices were not sensitive to show any difference between wheat monoculture system and wheat/jujube intercropping system in wheat rhizosphere soil. The values of Wasilewska index (*WI*) could present substantial changes in the trophic structure of the nematode community, in this study was greater than 14, which were higher than tillage and wheat residue field (4 – 12) (Liang *et al.*, 2005), soybean field (1.01 – 8.80) (McSorley & Frederick, 1996), and Gansu wheat field (0.40) (Liu *et al.* 2006). Bongers *et al.* (1997) reported that *PPI*/*MI* were higher in the intensively managed agricultural systems than natural undisturbed habitats, and effects of slight nutrient disturbances are indicated by a value of up to 1.2. Lower *PPI*/*MI indicate* barren soil food web and less plant productions, and showed that wheat/jujube intercropping system could emaciate the soil conditions. Lower values of *PPI*/*MI* suggested less plant production underground in JW, in line with decreased root length density and root surface area density in JW (Zhang *et al.*, 2013).

Most studies on intercropping have focused on resource utilization, including water, light, and nutrients (McIntyre *et al.*, 2001; Inal *et al.*, 2007). It has been shown that the interspecific interactions in the rhizosphere facilitate N and P uptake in intercropping systems, resulting in substantial yield advantage compared with sole cropping (Li *et al.*, 2010). Our obtained result also showed that wheat/jujube intercropping system increased the content of soil organic material (Table 1). It has been reported that jujube/wheat intercropping significantly decreased growth and yield of wheat and jujube in south Xinjiang, China, however, both of Land equivalent ratio (LER) were greater than 1.0, that means intercropping is advantageous (Song *et al.*, 2007; Zhang *et al.*, 2013). The difference between monoculture jujube and wheat/jujube intercropping system in jujube rhizosphere soil need further studies (in preparation).

Conclusion

Wheat grown in jujube orchard could negatively affect the nematode community in wheat rhizosphere soil. Wasilewska index (*WI*) and *PPI*/*MI* could provide useful information about the effects of monoculture and intercropping system on soil nematode fauna, lower *WI* and *PPI*/*MI* in wheat/jujube intercropping system might indicate the less fertile soil food web with fungal decomposition channels than wheat monoculture system.

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