

A Soil Nematode Community Response to Reclamation of Salinized Abandoned Farmland

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Development from abandoned land to farmland after vegetation reestablishment for reclamation is an important salinization rehabilitation process in dryland ecosystems. While subsequent soil abiotic changes have been reported, few studies focused on how reclamation affected soil biota. Understanding the response of soil biota to reclamation is useful for evaluating the effect of agricultural management. We investigated soil physiochemical properties, the composition and structure of nematode communities, and nematode metabolic footprints in the control and reclaimed farmland. The results showed that soil properties were significantly altered by reclamation. In particular, reclamation significantly increased pH, organic carbon, total nitrogen, and microbial biomass carbon. Conversely, electrical conductivity was significantly decreased. Shannon and Simpson indices were affected by reclamation. Reclamation significantly increased Shannon index in the 10-20 cm soil layer. Reclamation significantly increased Simpson index in the 0-10 cm soil layer, while the change of value in the 10-20 cm soil layer was opposite. High basal index and fungal-based channel were found in the control. Total nematodes abundance increased due to reclamation, which included fungivores, herbivores, and omnivores-predators. More nematodes could store more biomass carbon in the reclaimed farmland. Reclamation had an effect on the structure and function of soil food web, and increased the metabolic footprints of varied trophic groups of nematodes. Nematode faunal analysis revealed that exogenous substances input led to the high level of communities structure, and the soil food web became mature in the reclaimed farmland. The nematode communities were affected by reclamation. Furthermore, pH, EC, SOC, TN, and MBC were key driving factors affecting the nematode communities. Therefore, reclamation could effectively enhance the structure and function of soil food web through bottom-up effects in the cotton fields in Xinjiang, China.

Key words: Faunal analysis, Metabolic footprint, Soil food web, Soil health, Land use.

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BACKGROUND

Salinization is a serious environmental problem that caused by natural or human activities, which threatens the sustainable production of crops and ecosystem sustainability in arid and semi-arid regions in the world. Arid desert climate is important condition to form salinized land. Salt content of soil parent material is important cause for salinization. Hydrogeology is the main driving force for salinization. Human factor is important condition for secondary salinization in irrigation area (i.e., Large amounts of water were diverted to reclaimed abandoned farmland. Reservoir leakage and imperfect irrigation and drainage system. Impact of agricultural measures.) (Zinck and Metternicht 2009; Zhang et al. 2009). Excessive salt accumulation affects the chemical and biological processes of soil, reduces the absorption of water and nutrients, and restricts crop growth (Barin et al. 2015). In order to ensure stabilized cultivated land and food security, it is necessary to reclaim the salinized abandoned farmland (Cheng et al. 2018).

Xinjiang is located in the northwest of China and belongs to arid region. However, there are 95% of population in the oasis where occupies only 7% land area of Xinjiang. Soil quality is crucial for agricultural production in oasis farmland. Excessive flood irrigation caused the near-surface groundwater level to rise. Evaporation aggravated salt accumulation in the soil surface due to the high salt content of shallow groundwater. Therefore, a large area of salinized land has formed in the region over time. Since 2000, drip irrigation technology has been widely applied in Xinjiang, which has enabled a large area of salinized abandoned farmland to be reclaimed. A common method is replanting crops to restore soil fertility in abandoned farmland. Reclamation and agricultural utilization not only improve soil quality, but also influence ecological processes. The evolution of soil ecosystems is an important aspect to assess the levels of agricultural utilization and management in the oasis process. In addition, as the most active part of the belowground ecosystem, soil organisms also respond positively to changes in the soil environment (Jangid et al. 2011). Previous studies have focused on changes in soil organic carbon, nutrients, salt, and soil structure during the oasis process (Li et al. 2006; Su and Yang 2008). However, it has not been reported from the perspective of soil fauna. In view of the important role of nematodes in the soil ecosystems, it is of great significance to study the changes of nematodes in the oasis process to reveal the ecological

processes of soil.

Nematodes are the most abundant and diverse group of metazoans in the soil. They are widely distributed in various habitats (Bongers and Bongers 1998), and occupy a central position in the detritus food web (Neher 2001). They play an important role in the decomposition of soil organic matter, nitrogen mineralization, and nutrient cycling (Griffiths 1994). In addition, the morphology or life history strategy of nematodes is highly related to their habitat characteristics (Cheng et al. 2018). Nematodes have the potential to be used as bioindicator of soil health because of they are sensitive to environmental changes (Bongers and Ferris 1999; Neher 2001; Yeates et al. 2009). The abundance and diversity of nematodes are used to infer soil process rates (Ettema 1998), soil functions (Yeates 2003), and the effects of disturbance on soil fauna (Wardle et al. 1995). The composition and ecological indices of nematode communities can indicate changes in the soil environment, which contribute to better understand the effects of external activities on soil nutrients, decomposition pathways, and the structure and function of soil food web (Bongers 1990; Ferris et al. 2001; Ferris 2010). During the reclamation process, the distribution and abundance of nematodes are determined by various factors such as salinity, fertilization, and tillage. The change of microbial communities composition caused by salinity may affect their predators (Llamas et al. 2008), such as free-living nematode communities, leading to changes in prey-predator balance and the structure of soil food web. Okada and Harada (2007) found that the abundances of total nematodes, bacterivores, fungivores, and omnivores were higher in soil with chemical and organic fertilizer than that in unfertilized soil. Wardle (1995) concluded from different studies that the abundance of total nematodes responded differently to tillage (stimulation or inhibition), and tillage reduced larger organisms. In addition, previous studies mainly focused on analyzing the ecological indices of composition and diversity of nematode communities from the perspective of nematode ecology. These indices do not provide much information about the magnitude or nature of ecosystem functions (Ferris 2010). Ferris (2010) extended the assessment of ecosystems by introducing the concept of nematode metabolic footprint (NMF). And this is an effective method for estimating the contribution of nematodes to ecosystem functions and services (Ferris et al. 2012). In this study, we use the nematode metabolic footprint to indicate how reclamation affects the soil food web.

The salinization process leads to changes in organisms in the soil, resulting in a decrease in soil productivity, because soil biota contributes to the growth and productivity of plants. Among the soil organisms affected by salinity, nematodes are the most prominent. They are affected by the low osmotic potential of the soil solution and a large number of toxic ions (Wu et al. 2015), leading to changes in ecological balance, presenting less structured and less complex nematode communities (Salamún et al. 2014), and high mortality (Poage et al. 2008). Soil salinity, organic carbon, and hydrocarbon content were key factors negatively affecting the density, biomass, and diversity of

nematodes (Mahmoudi et al. 2002). For example, *Tylenchus* and *Aphelenchoides* were tolerant to slight saline-alkaline soil, while a few species of *Dorylaimus* and *Tylencholaimus* were abundant in saline-alkaline soil (Ray and Das 1980). Usually, under high salt stress, the growth of many plants was threatened, and even some plants cannot survive (Steinhorst and Kudla 2019; Otlewska et al. 2020). Therefore, the reduction of vegetation abundance and diversity reduced the food sources of herbivores and microorganisms, and ultimately reduced the abundance and diversity of free-living nematodes (Rath et al. 2016; Steinhorst and Kudla 2019; Rath et al. 2019).

Therefore, to clarify how reclamation affects soil nematode communities and soil food web. The objectives of this study were: (i) to determine the changes in soil physicochemical property under reclamation, (ii) to determine the responses of soil nematode communities composition, nematode metabolic footprints to reclamation, and (iii) to explore the possible driving factors in the changes of nematode communities. We made the following hypotheses. First, reclamation might improve soil quality. Second, reclamation might have a positive effect on the abundance of soil nematodes. Third, reclamation might increase the metabolic activity of soil organisms, which was favorable for the stability and sustainability of soil food web.

MATERIALS AND METHODS

Study area

The study area is located in Shihutan Township, Xinjiang Province, China, which is on the alluvial plain of Manasi River Basin along the southern margin of the Junggar Basin (44°37'N, 86°08'E). This area has an arid continental climate. The annual temperature and accumulated temperature $\geq 10^{\circ}\text{C}$ are 6.6°C and 3,490°C, respectively. The annual rainfall, evaporation, and frost-free period are 110-200 mm, 1,500-2,000 mm, and 148-187 days, respectively. Cotton yields is 5,250 kg ha⁻¹ and continuous cotton cropping of cotton is common in this area.

Saline-alkaline is the collective term for saline soil and alkaline soil. It refers to soil with a salt content of more than 0.2%, or soil colloids that adsorb a certain amount of exchangeable sodium with a degree of alkalinity of more than 20%, which is harmful to the normal growth of crops. It is also known as saline soil.

In addition to mountains and deserts in Xinjiang, saline-alkali soil is generally distributed in plain areas. The arid climate and geological historical conditions promoted the general development of salinization in Xinjiang plains. Whether it is the type of salt accumulation or the composition of salt is extremely complex and diverse. Soil salt accumulation in Xinjiang has the following

remarkable characteristics: (1) Soil salinization is common, the degree of salt accumulation is high, and the distribution area is wide. (2) The composition of salt soil is complex, mainly including chloride, sulfate, soda, and nitrate. (3) The accumulation rate of salt is fast and the intensity of accumulation is high, which shows strong surface accumulation in southern Xinjiang. (4) The salt accumulates for a long time. In addition to modern salt accumulation, there is also a large area of the existence of residual saline-alkali soil. (5) In the ancient oasis irrigation areas in southern Xinjiang, low-lying land is mostly treated as dry salt drainage areas. In the oasis irrigation area of northern Xinjiang, lowland irrigation, which is easy to cultivate is selected, resulting in salt accumulation in the nearby micro-highland. In terms of the type and intensity of salt accumulation, there are great differences between northern and southern Xinjiang. The salt accumulation in northern Xinjiang is light, mainly sulfates. The salt accumulation in southern Xinjiang is heavy, mainly chlorides. Moreover, the salinized soil in most of Xinjiang has varying degrees of soda salinization. Affected by the general salt content of the soil parent material and the varying degrees of salinity in the groundwater of the irrigation area, once irrigation and drainage are unbalanced, it is easy to cause secondary salinization, and even if the land is well-treated, it is easy to cause salinity to return.

Experimental design and management practices

The reclaimed experiment employed a randomized complete block design with three replicates. The treatments were comprised of original abandoned farmland (control) and reclaimed farmland. The experimental site had been farmed for a long time before it was abandoned for 29 years due to severe salinization. In 2006, the abandoned farmland was reclaimed to plant cotton in the designated area, which was treated as reclaimed farmland treatment. In the past 10 years, cotton was continuously planted under plastic film mulch in the reclaimed farmland with drip irrigation. The area without cotton planting since 1996 was selected as the control treatment of pre-reclamation, which was not protected from anthropogenic activity. Each treatment had an area of 5.2×7 m with 2 m buffer rows around it, and the plots in each replication also had 2 m buffer rows.

Cotton was sown in April and harvested in October every year. The sowing density of cotton was 2.4×10^5 plants ha^{-1} . The rainfall was 138.7 mm during the growth period. Drip-irrigated was performed 10 to 12 times during the growth period. The total amount of irrigation reached $4,500 \text{ m}^3$ ha^{-1} . Nitrogen ($300 \text{ kg} \cdot \text{ha}^{-1}$) and phosphorus ($200 \text{ kg} \cdot \text{ha}^{-1}$) fertilizer were applied via drip-irrigation system at different growth stages of cotton. Before sowing, urea ($150 \text{ kg} \cdot \text{ha}^{-1}$) and calcium superphosphate ($450 \text{ kg} \cdot \text{ha}^{-1}$) were applied into the soil as basal fertilizers. After the cotton was harvested, the cotton straw ($6,000\text{-}7,500 \text{ kg} \cdot \text{ha}^{-1}$) was crushed and applied into the soil with plough.

The soils were classified as grey desert soil (Gong et al. 1988). Vegetation in the unfarmed area was sparse, with main species including *Tamarix chinensis* Lour., *Kalidium foliatum* (Pall.) Moq., *Karelinia caspia* (Pall.) Less, and *Seriphidium sawanense* (Besser ex Less.) Fourr. The vegetation was uniformly distributed across the field.

Soil sampling

Soil samples (soil layer: 0–10 cm and 10–20 cm; diameter: 2.5 cm) were collected from ten plots in each treatment using soil auger at the flowering stage of cotton on August 16, 2016. The plots were arranged by “S-pattern” across the entire area. Soils of each layer collected from the plots of each treatment were homogenized to obtain one composite soil sample as a representative soil sample. A total of 12 soil samples consisted of two treatments × two layers × three replicates, and the weight of each soil sample was approximately 500 g. Roots, rocks, and debris in all soil samples were removed by hand. The soil samples were stored individually in plastic bags and quickly taken back to the laboratory in dry ice boxes. Each soil sample was divided into two parts. One part was stored in refrigerator at 4°C for nematode analysis, and the other part was air-dried for soil physicochemical analysis.

Soil physicochemical properties

Soil pH (soil/water ratio of 1:5) and electrical conductivity (EC) (soil/water ratio of 1:2.5) were determined with the potentiometry (Jackson 1973) and electrode method (Rhoades 1996). Soil organic carbon (SOC) was determined with the potassium dichromate volumetric method-external heating method (Ciavatta et al. 1991). Total nitrogen (TN) was determined with the semi-micro Kjeldahl digestion method (McGill and Figueiredo 1993). Microbial biomass carbon (MBC) was determined with the chloroform fumigation extraction method (Vance et al. 1987). Soil moisture was determined with the gravimetric method (Yang et al. 2013).

Nematode extraction and identification

A modified cotton-wool filter method was used to extract nematodes from 100 g fresh soil (Townshend 1963). First, nematodes were counted using dissecting microscope (Motic, Group Co., Ltd., China). Second, the number of nematodes in 100 g fresh soil was converted to 100 g dry soil according to the soil moisture content. Nematode abundances were expressed as the number of nematode individuals per 100 g dry soil. One hundred nematode individuals (if there were fewer

than 100 nematode individuals, all nematode individuals were identified) were randomly selected and identified to the genus level using optical microscope (OLYMPUS CX41, Olympus Corporation, Tokyo, Japan) at $\times 100$ magnifications in each sample (Bongers 1994; Zhang et al. 2013). Nematodes were divided into four trophic groups: bacterivores (Ba), fungivores (Fu), herbivores (H), and omnivores-predators (OP) according to the trophic habits and esophageal morphology of nematodes (Yeates et al. 1993).

Nematode ecological indices

The Shannon-Weiner index (H') (Shannon 1948) and Simpson index (λ) (Simpson 1949) were used to indicate the nematode diversity. The basal index (BI) was used to reflect the tolerance of opportunistic nematodes to soil disturbance. The channel index (CI) was used to indicate the predominant decomposition channels of soil. The enrichment index (EI) was used to assess the response of soil food web to available resources (Ferris et al. 2001). The structure index (SI) was used to indicate the changes in the structure of soil food web in the process of human disturbance or ecological restoration (Ferris et al. 2001).

Nematode biomass carbon and metabolic footprint

The average biomass (fresh weight) of each genus (W_t) was estimated according to the database at http://nemalex.ucdavis.edu/Ecology/nematode_weights.htm. (Sieriebriennikov et al. 2014). It was estimated that the dry weight of the nematodes accounted for 20% of its fresh weight, and the nematode biomass carbon accounted for 52% of its dry weight (Ferris 2010). The biomass carbon of nematodes was calculated according to the following formula (Ferris 2010):

$$W_t \times 20\% \times 52\%$$

Where W_t represents the fresh weight.

The metabolic footprint of nematodes was calculated according to the following formula (Ferris et al. 2012):

$$NMF = \sum \{N_t [0.1(W_t/m_t) + 0.273(W^{0.75})]\}$$

Where N_t represents the number of nematodes in the t -th genus, and W_t and m_t represent the fresh weight and c-p value in the t -th genus, respectively.

The metabolic footprints of nematodes consisted of enrichment footprint (efootprint) and structure footprint (sfootprint). Enrichment footprint refers to the metabolic footprint of nematodes with lower trophic levels (c-p value: 1-2) and rapid response to resource enrichment. Structure

footprint refers to the carbon metabolism process of nematodes with high c-p values (3-5), which have a regulating effect on soil food web (Ferris et al. 2012). The functional metabolic footprint (FMF) is represented by the total delineative region of efootprints and sfootprints, which is used to evaluate the soil food web (Ferris et al. 2012). The FMF was calculated according to the following formula:

$$FMF = (efootprint \times sfootprint)/2$$

Statistical analysis

Data distributions were checked prior to the transformation of data. The abundance of nematodes will be transformed using $\ln(x + 1)$ if it does not conform to the normal distribution prior to statistical analysis. Statistical analyses were performed using SPSS (version 19.0, IBM Corp., Armonk, New York, USA). Two-way ANOVA (general linear model) was used to analyze the effects of reclamation, soil layers, and their interactions. Significance tests were performed using multiple comparison tests at $P < 0.05$. The structure of nematode communities was analyzed by principal component analysis (PCA) using Canoco software (version 4.5) based on the abundance of nematode genera (ter Braak and Šmilauer 2012). The relationships of soil nematode communities with each environmental variable were determined using the redundancy analysis (RDA) in R software (version 3.4.3, R Development Core Team, New Zealand) with vegan package (Sheik et al. 2012). R was used to generate the figures.

RESULTS

Soil physicochemical properties

Two-way analysis of variance showed that reclamation had a significant effect on soil pH, EC, SOC, TN, and MBC ($P < 0.01$) (Table 1), soil layer had a significant effect on TN ($P < 0.01$), and their interaction had a significant effect on EC, TN, MBC ($P < 0.01$), and SOC ($P < 0.05$). In both soil layers, SOC, TN, and MBC in the reclaimed farmland were significantly higher than that in the control, while EC was lower in the reclaimed farmland ($P < 0.01$). Reclamation only significantly increased pH in the 0-10 cm soil layer compared with control ($P < 0.01$).

Table 1. Soil physicochemical properties in the abandoned and reclaimed soils

	0-10 cm		10-20 cm		reclamation	layer	reclamation × layer
	abandoned farmland	reclaimed farmland	abandoned farmland	reclaimed farmland			
pH	8.51 ± 0.02B	8.82 ± 0.11A	8.64 ± 0.09	8.79 ± 0.07	< 0.01	ns	ns
EC (µs/cm)	1012.00 ± 9.00A	196.37 ± 6.78B	970.33 ± 11.06A	263.00 ± 26.85B	< 0.01	ns	< 0.01
SOC (g/kg)	3.55 ± 0.19B	12.16 ± 0.27A	2.84 ± 0.68B	12.60 ± 0.14A	< 0.01	ns	< 0.05
TN (g/kg)	0.25 ± 0.02B	0.95 ± 0.02A	0.19 ± 0.03B	1.15 ± 0.01A	< 0.01	< 0.01	< 0.01
C/N	14.09 ± 1.16	12.85 ± 0.12	15.09 ± 5.47	10.99 ± 0.25	ns	ns	ns
MBC (mg/kg)	28.24 ± 4.56B	163.84 ± 12.14A	79.39 ± 9.23B	116.08 ± 8.60A	< 0.01	ns	< 0.01

Notes: Values are means ± standard deviation ($n = 3$). EC, SOC, TN, C/N and MBC represent electrical conductivity, soil organic carbon, total nitrogen, carbon nitrogen ratio, and microbial biomass carbon, respectively. Different capital letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.01$). ns indicates no significant differences.

Nematode ecological indices

The influences of reclamation, soil layer ($P < 0.05$), and their interaction ($P < 0.01$) on the Shannon index were significant (Fig. 1a). In the 10-20 cm soil layer, the Shannon index in the reclaimed farmland was significantly higher than that in the control ($P < 0.01$). The influences of soil layer ($P < 0.05$) and their interaction ($P < 0.01$) on the Simpson index were significant (Fig. 1b). In the 0-10 cm soil layer, the Simpson index in the reclaimed farmland was significantly higher than that in the control ($P < 0.05$). However, in the 10-20 cm soil layer, the Simpson index in the reclaimed farmland was significantly lower than that in the control ($P < 0.01$). Reclamation had a significant effect on the basal index ($P < 0.01$) (Fig. 1c). The basal index in the control was significantly higher than that in the reclaimed farmland in both soil layers ($P < 0.01$). Their interaction had a significant effect on the channel index ($P < 0.05$) (Fig. 1d). In the 10-20 cm soil layer, the channel index in the control was significantly higher than that in the reclaimed farmland ($P < 0.01$).

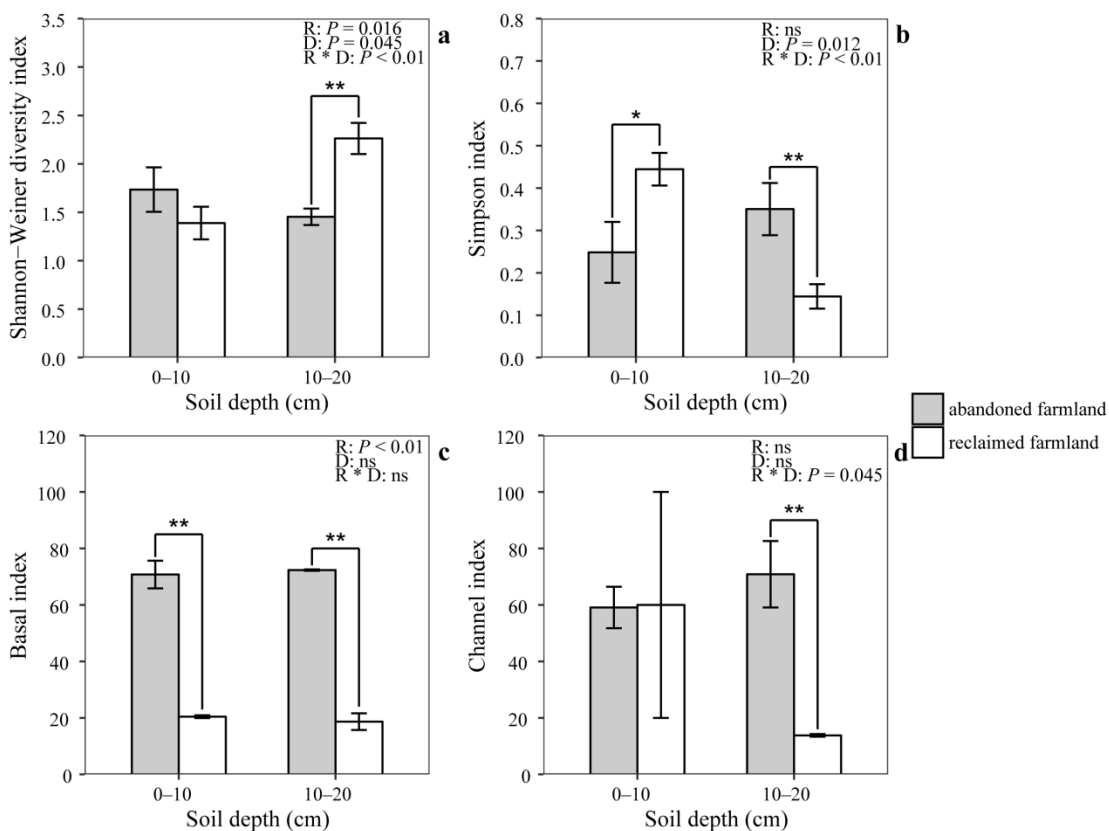


Fig. 1. Variation of nematode ecological indices for abandoned farmland and reclaimed farmland. Values are means \pm standard deviation ($n = 3$). The effects of reclamation and layer from the two-way ANOVA are revealed for each figure. R represents reclamation, D represents soil layer, and R*D represents the interaction between reclamation and soil layer. The * indicates significant

differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.05$) and ** at the $P < 0.01$ level. ns indicates no significant differences.

Nematode abundance and biomass carbon

In this study, 25 and 28 nematode genera were identified in the control and reclaimed farmland in both soil layers, respectively (Table S1). The influences of reclamation on the abundances of total nematodes, H, OP ($P < 0.01$), and Fu ($P < 0.05$) were significant (Fig. 2). Their interaction had a significant effect on the abundances of total nematodes ($P < 0.05$) and H ($P < 0.01$). In the 0-10 cm soil layer, the abundances of total nematodes ($P < 0.05$), H, and OP ($P < 0.01$) in the reclaimed farmland were significantly higher than that in the control. In the 10-20 cm soil layer, the abundances of H ($P < 0.05$) and OP ($P < 0.01$) in the reclaimed farmland were also significantly higher than that in the control.

In addition, the influences of reclamation on the biomass carbon of total nematodes, Fu, H, OP ($P < 0.01$), and Ba ($P < 0.05$) were significant (Table 2). The influences of soil layer on the biomass carbon of total nematodes, Ba, OP ($P < 0.05$), and H ($P < 0.01$) were significant. Their interaction had a significant effect on the biomass carbon of total nematodes, Ba, and H ($P < 0.01$). The biomass carbon of Fu ($P < 0.05$), H, and OP ($P < 0.01$) were, respectively, 197%, 2552%, and 467% in the reclaimed farmland significantly higher than that in the control in the 0-10 cm soil layer. The biomass carbon of total nematodes, Ba, OP ($P < 0.01$), Fu, and H ($P < 0.05$) were, respectively, 424%, 609%, 404%, 55%, and 114% in the reclaimed farmland significantly higher than that in the control in the 10-20 cm soil layer.

Table 2. Soil nematode biomass carbon (μg per 100 g dry soil)

	0-10 cm		10-20 cm		reclamation	layer	reclamation \times layer
	abandoned farmland	reclaimed farmland	abandoned farmland	reclaimed farmland			
Nematode-C	11.00 \pm 5.10	26.28 \pm 8.31	8.97 \pm 2.32B	47.00 \pm 5.14A	< 0.01	< 0.05	< 0.01
Ba-C	8.54 \pm 5.32	5.02 \pm 3.08	5.47 \pm 1.78B	38.79 \pm 5.12A	< 0.05	< 0.05	< 0.01
Fu-C	1.15 \pm 0.20b	3.41 \pm 1.65a	1.50 \pm 0.21b	2.33 \pm 0.41a	< 0.01	ns	ns
H-C	0.50 \pm 0.11B	13.26 \pm 2.43A	1.45 \pm 0.53b	3.10 \pm 0.65a	< 0.01	< 0.01	< 0.01
OP-C	0.81 \pm 0.07B	4.59 \pm 1.15A	0.55 \pm 0.50B	2.77 \pm 0.25A	< 0.01	< 0.05	ns

Notes: Values are means \pm standard deviation ($n = 3$). Nematode-C, Ba-C, Fu-C, H-C and OP-C represent the biomass carbon of total nematodes, bacterivores, fungivores, herbivores and omnivores-predators, respectively. Different lowercase letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.05$). Different capital letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.01$). ns indicates no significant differences.

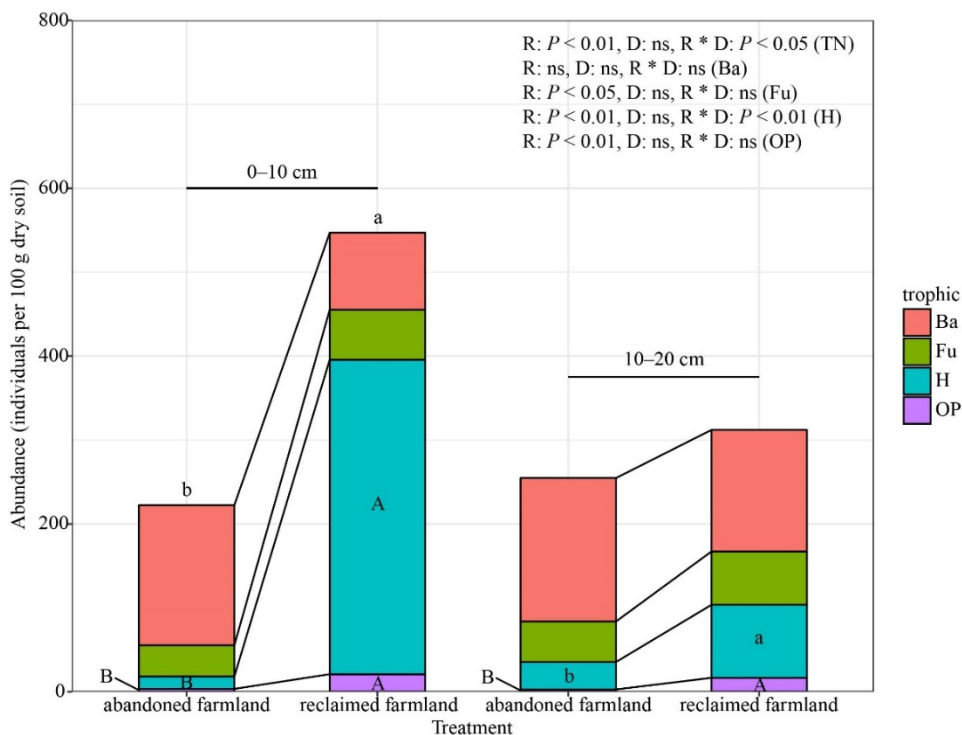


Fig. 2. Soil nematode abundance. Values are means \pm standard deviation ($n = 3$). TN, total nematodes; Ba, bacterivores; Fu, fungivores; H, herbivores and OP, omnivores-predators. R represents reclamation, D represents soil layer, and R*D represents the interaction between reclamation and soil layer. Different lowercase letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.05$). Different capital letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.01$). ns indicates no significant differences.

Nematode faunal analysis and metabolic footprint

The metabolic footprint characteristics of soil food web showed that the EI and SI were lower in the control than that in the reclaimed farmland in the 10-20 cm soil layer, and the SI was lower in the control than that in the reclaimed farmland in the 0-10 cm soil layer (Fig. 3). In both soil layers, plots in the control were in quadrant D, indicating the soil nutrient status was poor, the disturbance was the highest and the C/N ratio was high, which caused environmental stress and the degraded soil food web. There were clear differences in the metabolic footprint characteristics between 0-10 cm and 10-20 cm soil layers in the reclaimed farmland. The plot in the 0-10 cm soil layer was in quadrant C, which indicated that the soil nutrient status was poor, the disturbance was less, the soil food web was in a structured status, and the C/N ratio was high. The nematode faunal profiles indicated that the soil nutrient was rich, the disturbance was less, the soil food web was stable and mature, and the C/N ration was low in the 10-20 cm soil layer for the location of plot in quadrant B.

The influences of reclamation on Fu, H, OP, and sfootprint were significant ($P < 0.01$) (Table 3). Soil layer had a significant effect on the metabolic footprint of Ba, H, OP, efootprint ($P < 0.05$),

and sfootprint ($P < 0.01$). Their interaction had a significant effect on the metabolic footprint of Ba, H, efootprint, sfootprint ($P < 0.01$), and OP footprint ($P < 0.05$). The metabolic footprint of Fu ($P < 0.05$), H, OP, and sfootprint ($P < 0.01$) in the reclaimed farmland were significantly higher than that in the control in the 0-10 cm soil layer. Similar changes were found in the 10-20 cm soil layer with the metabolic footprints of H ($P < 0.05$), Ba, OP, efootprint, and sfootprint ($P < 0.01$) in the reclaimed farmland were significantly higher than that in the control. The FMF in the reclaimed farmland were significantly higher than that in the control in the 0-10 cm ($P < 0.05$) and 10-20 cm soil layers ($P < 0.01$).

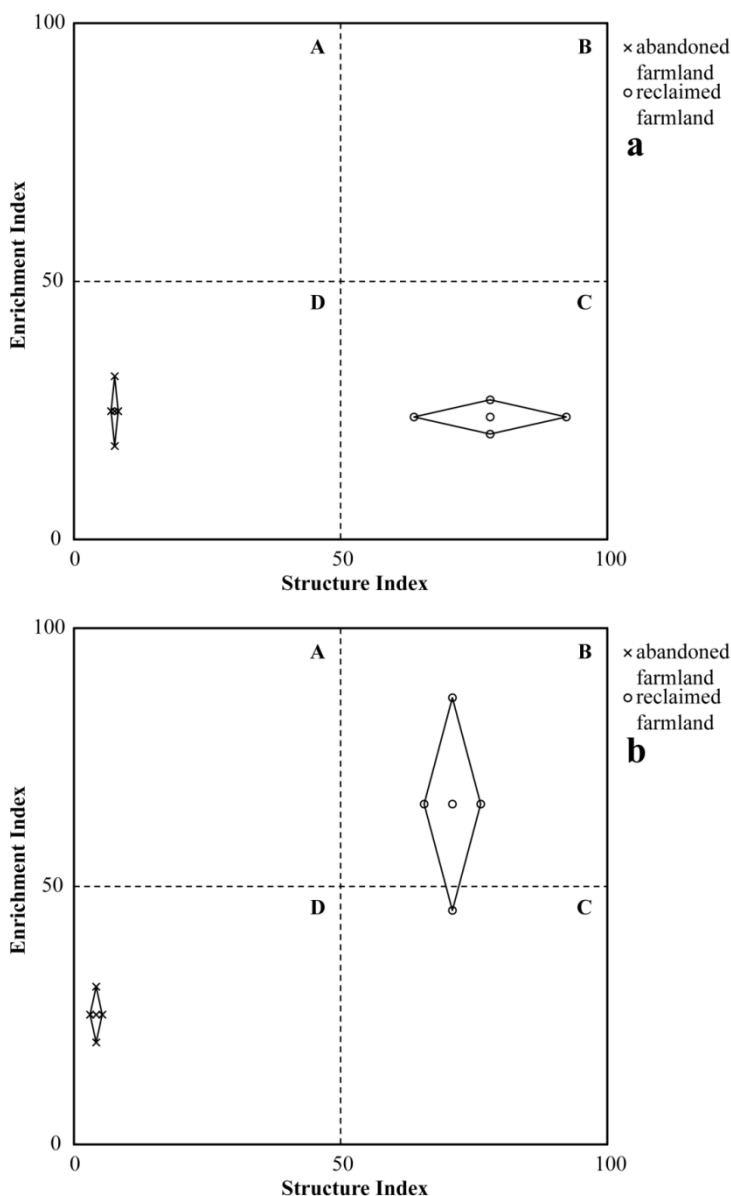


Fig. 3. Functional metabolic footprints of nematode communities in the abandoned farmland and reclaimed farmland for different soil layers. (a) and (b) represent the 0-10 cm and 10-20 cm soil layer, respectively. The EI represents the enrichment footprint and the SI represents the structure footprint. The functional metabolic footprint is described as follows: (SI, EI), (SI-0.5sfootprint/k, EI), (SI, EI+0.5efootprint/k), (SI+0.5sfootprint/k, EI) and (SI, EI-0.5efootprint/k). The adjusted k value is 2.5. The functional metabolic footprint is the total area of the enrichment and structure footprint.

Table 3. Soil nematode metabolic footprints ($\mu\text{g C kg}^{-1}$ soil)

	0-10 cm		10-20 cm		reclamation	layer	reclamation \times layer
	abandoned farmland	reclaimed farmland	abandoned farmland	reclaimed farmland			
Ba footprint	28.84 \pm 16.26	16.33 \pm 8.65	20.20 \pm 6.65B	102.89 \pm 13.03A	ns	< 0.05	< 0.01
Fu footprint	4.53 \pm 0.39b	11.02 \pm 4.94a	5.95 \pm 0.82	8.48 \pm 1.53	< 0.01	ns	ns
H footprint	1.83 \pm 0.25B	49.65 \pm 9.34A	5.16 \pm 2.03b	11.48 \pm 2.55a	< 0.01	< 0.05	< 0.01
OP footprint	1.88 \pm 0.32B	11.01 \pm 2.56A	1.35 \pm 1.24B	6.64 \pm 0.23A	< 0.01	< 0.05	< 0.05
efootprint	33.75 \pm 16.35	16.57 \pm 11.02	26.97 \pm 8.20B	102.96 \pm 12.90A	ns	< 0.05	< 0.01
sfootprint	3.33 \pm 0.48B	71.44 \pm 14.46A	5.68 \pm 0.76B	26.52 \pm 0.66A	< 0.01	< 0.01	< 0.01

Notes: Values are means \pm standard deviation (n=3). Ba, bacterivores; Fu, fungivores; H, herbivores; OP, omnivores-predators; efootprint, enrichment footprint and sfootprint, structure footprint. Different lowercase letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.05$). Different capital letters indicate significant differences between abandoned farmland and reclaimed farmland within the same soil layer ($P < 0.01$). ns indicates no significant differences.

Differences in the nematode community and its influencing factors

The first and second principal components explained 85.1% of the total variance in the composition of nematode communities (68.5% and 16.6% for the PC1 and PC2 axes, respectively) (Fig. 4). PCA analysis showed that the composition of nematode communities in the control and reclaimed farmland in both soil layers were divided into two groups: (i) control and (ii) reclaimed farmland. “Control” and “reclaimed farmland” could be further separated into two soil layers. Each soil layer formed a distinct cluster. The nematode communities of control (i.e., group 1) and reclaimed farmland (i.e., group 2) were well separated by PC1. In the 0-10 cm and 10-20 cm soil layers, the nematode communities in the reclaimed farmland were well separated by PC2. Overall, these results suggested that (i) there were similarities in the composition of nematode communities within same treatment or soil layer and (ii) reclamation was the main reason for the differences in nematode communities.

RDA analysis showed that six environmental factors interpreted 80.56% of the distribution of nematodes (permutation test $P = 0.001$) (Fig. 5), indicating that the result of ordination could accept the interpretation of environmental factors on species distribution. The RDA1 and RDA2 axes explained 63.23% of the total variation. The RDA1 and RDA2 axis explained 44.11% and 19.12%, respectively. Significance tests for each environmental factor showed that the variations in the composition of nematode communities had a positive relationship with the pH, EC, SOC, TN, and MBC ($P < 0.01$) (Table 4).

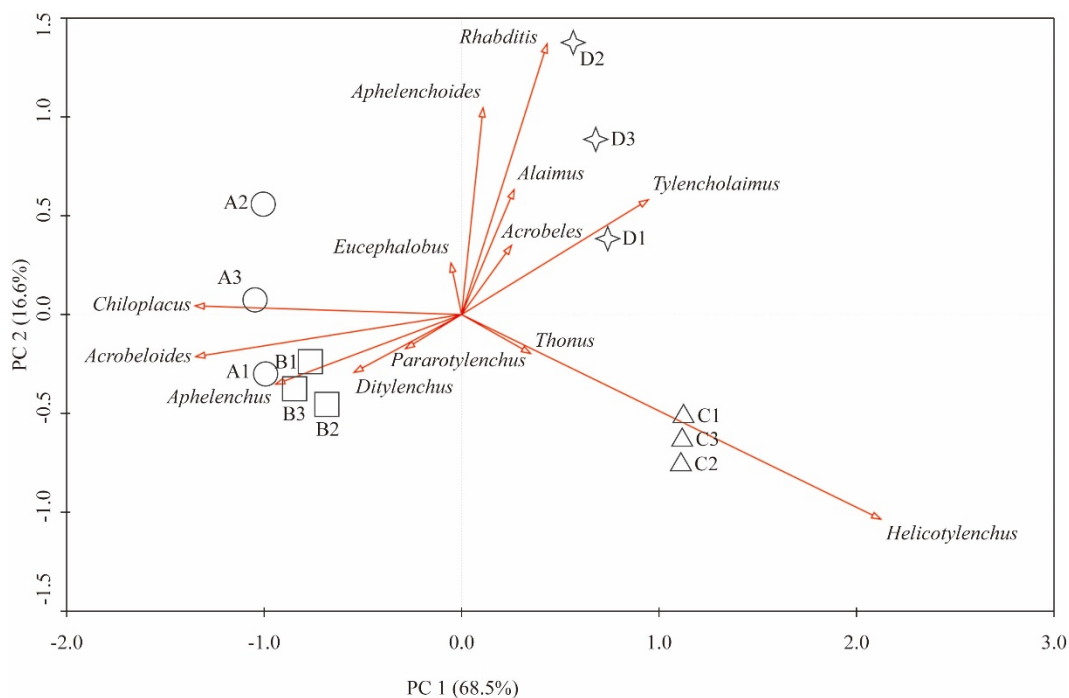


Fig. 4. Principle component analysis based on the relative abundance of nematode genus (> 1%) in the abandoned farmland and reclaimed farmland for different soil layers. A1-A3 and C1-C3

represent abandoned farmland and reclaimed farmland in the 0-10 cm soil layer, respectively. B1-B3 and D1-D3 represent abandoned farmland and reclaimed farmland in the 10-20 cm soil layer, respectively.

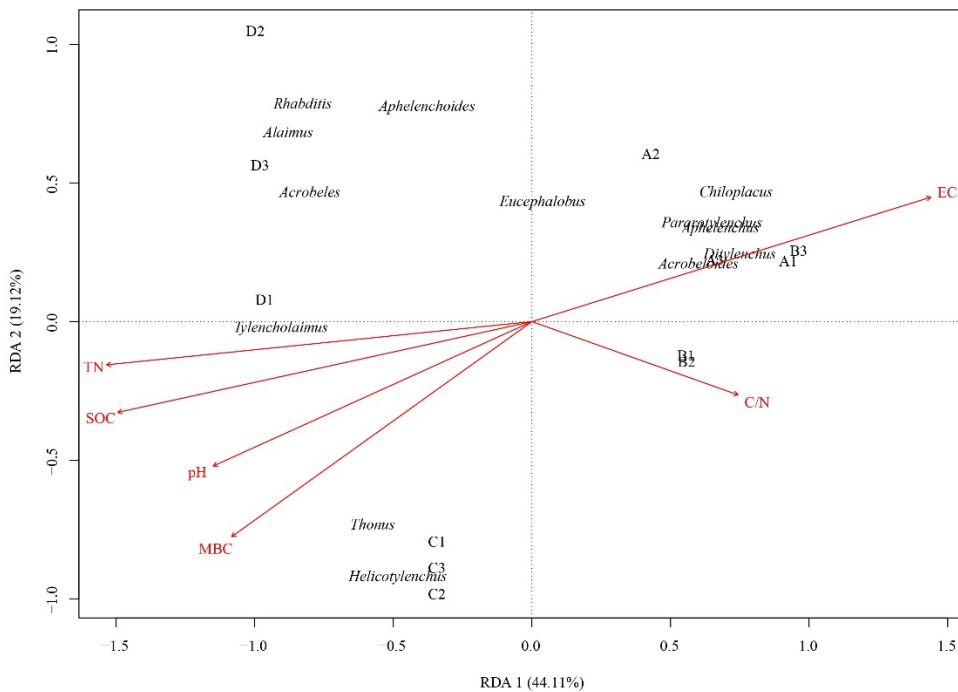


Fig. 5. Redundancy analysis of the dominant nematode genera and soil physicochemical properties in the abandoned farmland and reclaimed farmland for different soil layers. A1-A3 and C1-C3 represent abandoned farmland and reclaimed farmland in the 0-10 cm soil layer, respectively. B1-B3 and D1-D3 represent abandoned farmland and reclaimed farmland in the 10-20 cm soil layer, respectively. EC, electrical conductivity; SOC, soil organic carbon; TN, total nitrogen; C/N, carbon nitrogen ratio and MBC, microbial biomass carbon.

Table 4. Significance test of environmental factors on the distribution of nematode communities

	RDA1	RDA2	R ²	P value
pH	-0.882	-0.471	0.688	0.007**
EC	0.937	0.349	0.946	0.001***
SOC	-0.967	-0.257	0.955	0.001***
TN	-0.992	-0.129	0.954	0.001***
C/N	0.928	-0.374	0.258	0.256
MBC	-0.768	-0.641	0.820	0.002**

Notes: ** $P < 0.01$; *** $P < 0.001$.

DISCUSSION

The effect on soil physicochemical properties

The replanting of crops had a significant effect on soil physicochemical properties, which caused by the fertilization and straw return to the farmland during reclamation (Yang et al. 2018). Reclamation significantly decreased EC was due to the drip irrigation and plastic film mulch employed during planting. Drip irrigation and plastic film mulch could lead to the movement and accumulation of salt to the deep soil layer with water. Zhang et al. (2017) also found the same phenomenon during reclaimed process. However, reclamation significantly increased SOC, which could be attributed to the straw return and fertilization during reclaimed process. Study showed that straw return and fertilization could increase soil organic matter input, carbon sequestration potential, and SOC pool, thus making farmland soil "carbon sink" (Chen et al. 2015). In this study, cotton was planted and straws were returned to the field after reclamation. Vegetation roots and residues were the main source of soil organic matter. Long-term straw return and drip irrigation under plastic film mulch could control soil moisture and temperature, which were conducive to decompose the straw by microorganism and increase SOC (Zheng et al. 2015). Besides, TN was increased due to reclamation, which was attributed to straw return and fertilization improved soil fertility (Zu et al. 2014). Straws provided sufficient C source for microbial biomass construction and reproduction and promoted microbial activity, which was conducive to form MBC (Muhammad et al. 2006).

The effect on nematode abundance

The abundance of total nematodes was lower in the control than that in the reclaimed farmland. Study found that soil organic matter, alkali-hydrolyzable nitrogen, and total nitrogen had significantly positive effects on organism abundance in both saline and non-saline soils (Wu et al. 2015). Almost all soil organisms need carbon-based food sources to provide the nutrients they need, including nitrogen and phosphorus. Soil organic matter is the main source of soil nutrients and carbon, so it is very important to the soil biota. Nematode abundance was closely related to organic matter in the soil (Wall et al. 2002). Increasing organic matter provided abundant food source for nematodes. The application of organic-inorganic fertilizers and long-term agricultural cultivation resulted in plant root residues into the soil, increasing the organic matter in the farmland soil in reclaimed process. On the other hand, the abandoned farmland was not conducive to nematodes survival due to its low organic matter and limited food sources, resulting in low nematodes abundance.

The reclaimed farmland showed a greater abundance of nematodes compared to the control, suggesting that salinization negatively affects nematodes. Similar results were found by Wu et al. (2015), Su et al. (2016), and Pen-Mouratov et al. (2010), who found that salinity significantly

decreased nematodes abundance. This reduction in nematodes abundance could be due to high EC in the control, as seen in Wu et al. (2015). However, there could be a second pathway in which salinity negatively affected the abundance of nematodes, since salinity reduces microbial biomass and respiration in the soil due to inhibit the decomposition of organic matter, thus, a reduction in the release of nutrients that may cause a reduction in the abundance of nematodes (Su et al. 2016). The difference in nematodes abundance between control and reclaimed farmland is caused by the low osmotic potential of the soil solution and ion toxicity or imbalanced ion absorption in the soil (Wu et al. 2015).

In the present study, EC was higher in the control than that in the reclaimed farmland. Many studies reported that salinity reduces microbial activity and biomass, and changes the structure of microbial communities (Rietz and Haynes 2003; Yuan et al. 2007; Saviozzi et al. 2011; Yan and Marschner 2013), thus affecting the nematode communities. Some studies reported that soil salinity is an important factor affecting the abundance and structure of soil nematode community (Thurston et al. 1994; Gooseff et al. 2003; Moorhead et al. 2003). Pen-Mouratov et al. (2011) found that EC was negatively correlated with nematodes abundance. Vicente et al. (2015) observed a negative correlation between omnivorous and EC in semiarid regions in Brazil. In addition, nematodes abundance was negatively correlated with salinity in arid desert oasis, indicates that salt is determinant of nematodes abundance (Su et al. 2012). The high salt content observed in salinized abandoned farmland destroys soil characteristics, creating a toxic environment for microorganisms (Wu et al. 2015), which could explain the decrease in nematodes abundance found in this study on salinized abandoned farmland.

The changes of nematode trophic groups were closely related to the dynamics of microbial communities affected by salinization. Studies showed that bacterial communities were dominate in the salinized soil (Pankhurst et al. 2001). Bacterivores were dominant trophic group in the control, indicating that bacteria were the main food resource of nematodes, and the energy pathway was mainly dominated by bacteria in the salinized soil. Bacterivores and herbivores were dominant trophic groups in the reclaimed farmland. A large amount of organic matter nourished abundant bacteria in the farmland, and bacterivores fed on bacteria as nutrient source (BulluckIII et al. 2002). Herbivores were the largest trophic group in the surface soil in the reclaimed farmland. This result was consistent with study by Ou et al. (2005) who found herbivores were dominant trophic group in maize fields of yellow brown soil. While the feeding behavior of herbivores accelerated the transportation of nutrients from plants to soil and promoted the accumulation of organic matter (van der Putten and van der Stoel 1998). Omnivores-predators were the lowest trophic groups in both soils. High trophic level organisms (i.e., omnivores-predators) responded slowly to their prey (Mikola and Setälä 1998), suggesting that the matter and energy flow were bottom-up in the soil

food web. The abundance of fungivores and herbivores in the reclaimed farmland were higher than that in the control was related to fertilization (Vestergård 2004), plant residues returned to the field (Sánchez-Moreno et al. 2006), and increased in vegetation roots (van Capelle et al. 2012). And the high abundance of omnivores-predators indicated that the stability of soil environmental has been improved. Because of omnivores-predators were very sensitive to environmental disturbances (Ruan et al. 2012). Abandoned farmland soil was frequently exposed and eroded by salinization, and its stability was lower than that in the reclaimed farmland.

The effect on nematode ecological indices

Shannon index in the reclaimed farmland was slightly lower than that in the control in the 0-10 cm soil layer. Because of the surface soil after reclamation was largely disturbed (i.e., tillage, fertilization, irrigation et al.). However, Shannon index in the reclaimed farmland was significantly higher than that in the control in the 10-20 cm soil layer. The input of exogenous substances provided abundant food sources for nematodes. Simpson index in the reclaimed farmland was significantly higher than that in the control in the 0-10 cm soil layer. Because of vegetation increased in the reclaimed surface soil, and herbivores (*i.e.*, *helicotylenchus*) abundance increased rapidly. The dominant population of nematode was relatively single, which was not conducive to the stability of nematode communities. Simpson index in the reclaimed farmland was significantly lower than that in the control in the 10-20 cm soil layer. Herbivores were evenly distributed among different genera, resulting in the dominance was decreased. Basal index reflects the response of soil food web to nutrient resources (Ferris et al. 2001). High basal index indicated that the nematode communities were mainly composed of the tolerant species of nematodes with lower trophic levels in the disturbed ecosystem and the ecosystem was in degraded state. Basal index in the reclaimed farmland was significantly lower than that in the control in both soil layers, indicating that the health condition of ecosystem in the reclaimed farmland was better than that in the control (Berkelmans et al. 2003). Channel index reflects the decomposition pathways in the soil food web. Channel indices were greater than 50 in both soil layers in the control, indicating that the fungal decomposition pathway was dominant. Spatial analysis of reclaimed and undisturbed sites showed that fungal biomass was significantly lower in reclaimed soil than that in undisturbed soil (Mummey et al. 2002). The surface soil of reclaimed farmland also showed the same higher value. The fungal decomposition pathway is characterized by low decomposition rate (Yeates and Boag 2004) and slow nutrient cycling. However, the bacterial decomposition pathway was dominant and high nutrient cycling in the 10-20 cm soil layer in the reclaimed farmland. When cotton grew vigorously, there were few litters in the upper layer and developed roots in the deeper layer. Studies

showed that the plant residues were decomposed by microorganism contributed to the growth of bacterivores (Chen et al. 2007).

Both EI and SI were low in the control in both soil layers due to environmental disturbances, which caused environmental stress and the degraded soil food web. Low EI and high SI in the surface soil in the reclaimed farmland indicated that the disturbance was less, and the soil food web was in a stable and structured state. In the 10-20 cm soil layer, both EI and SI were high, indicating that the disturbance was less, and the soil food web was in a more mature state. Human agricultural management measures led to abundant nutrients and opportunistic nematodes been enriched (Villenate et al. 2003). The environmental homeostasis of reclaimed farmland resulted in the structure of communities was good, indicating healthy and well-managed ecosystem. SI was primarily determined by omnivores-predators. However, omnivores-predators required longer generation cycle than bacterivores and fungivores (Liang et al. 2009). High SI was due to high omnivores-predators abundance in the reclaimed farmland, which indicated that the food web had more trophic-level contact in the habitat (Ferris and Matute 2003).

The effect on nematode metabolic footprint

Nematode footprint characterizes the carbon metabolism. It not only reflects the response of nematode communities to resources, but also reflects the functions and services provided by nematodes. They show the connectivity of the soil food web and the relationship between predator and prey (Ferris et al. 2012). Enrichment footprint refers to rapid response to resource enrichment (Ferris et al. 2012). High enrichment footprint in the reclaimed farmland in the 10-20 cm soil layer indicating an increase in the supply of resources. In the reclaimed farmland, the prey meets the needed of the predators and maintains the metabolic balance of the system by improving reproduction and metabolic activity to maintain high productivity and metabolic activity (Ferris 2010). Low enrichment footprint in the control in both soil layers and in the 0-10 cm soil layer in the reclaimed farmland, indicating that the prey abundance was low, which resulted in insufficient resource supply, and ultimately decreased the number of predators. Structure footprint refers to resources output and the metabolic activity of nematodes with high colonizer-persister groups, which have a regulating effect on the soil food web. High structure footprint in the reclaimed farmland in both soil layers indicated that the metabolic activity of predator was increased, which may be related to increased predation pressure. Similarly, the omnivores-predators footprint also showed the same changes. In both soil layers, the functional metabolic footprint in the reclaimed farmland was higher than that in the control. High functional metabolic footprint indicated that massive carbon was used for the reproduction of nematodes (Zhang et al. 2012).

The relationship between soil environment and nematodes

PCA and RDA analyses showed that the structure of nematode communities were different between control and reclaimed farmland in both soil layers. There were significant differences in nematode communities among different habitats. The results showed that the agricultural management measures changed the soil biota compared with initial state.

Reclamation changed soil properties and had an effect on nematode communities. So, the soil physicochemical properties were important driving forces. RDA analysis showed that pH, EC, SOC, and MBC were important in affecting nematode communities, which was consistent with the results of previous studies (Fiscus and Neher 2002; Ruan et al. 2012). SOC and TN directly or indirectly affect nematode communities and even soil food web by changing plant physiology or microbial activity (Jiang et al. 2013). High pH was associated with high herbivores and omnivores-predators abundance (Nielsen et al. 2015). Study showed that EC was negatively correlated with the abundance of total nematodes, indicating that salinity was an important factor to affect the composition and abundance of nematode communities (Su et al. 2018).

CONCLUSIONS

This study greatly improves our understanding of the effects of reclamation on soil nematode communities. Soil physicochemical properties could be significantly enhanced and soil developed toward maturity during reclamation. Reclamation has a positive correlation with nematode communities. Reclamation decreased nematode diversity in the 0-10 cm soil layer, but significantly increased diversity in the 10-20 cm soil layer. Nematode resistance to disturbance was reduced by reclamation. The decomposition channel of soil food webs was changed from fungal to bacterial during reclamation. Reclamation increased the abundance of total nematodes, fungivores, herbivores, and omnivores-predators. Reclamation had an effect on the structure and function of soil food web, and had a significant effect on the resistance of soil organisms due to environmental changes. Nematode trophic footprints responded differently to reclamation, providing information on carbon and energy entering or exiting the soil food web. Reclamation could store more biomass carbon, and eventually form C sink. There were significant changes in the composition and structure of nematode communities in the control and reclaimed farmland. pH, EC, SOC, TN, and MBC had a significant effect on nematode communities. Therefore, reclamation is beneficial to improving the stability of soil ecosystem and promoting the restoration and reconstruction of

abandoned farmland.

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Supplementary materials

Table S1. Mean relative abundance of nematode genus (proportion) (per 100 g dry soil) in the abandoned farmland and reclaimed farmland for different soil layers. (download)