

Responses of soil microbial and nematode communities to aluminum toxicity in vegetated oil-shale-waste lands

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Abstract Both soil nematodes and microorganisms have been shown to be sensitive bioindicators of soil recovery in metal-contaminated habitats; however, the underlying processes are poorly understood. We investigated the relationship among soil microbial community composition, nematode community structure and soil aluminum (Al) content in different vegetated aluminum-rich ecosystems. Our results demonstrated that there were greater soil bacterial, fungal and arbuscular mycorrhizal fungal biomass in *Syzygium cumini* plantation, greater abundance of soil nematodes in *Acacia auriculiformis* plantation, and greater abundance of soil predatory and herbivorous nematodes in *Schima wallichii* plantation. The concentration of water-

soluble Al was normally greater in vegetated than non-vegetated soil. The residual Al and total Al concentrations showed a significant decrease after planting *S. cumini* plantation onto the shale dump. Acid extractable, reducible and oxidisable Al concentrations were greater in *S. wallichii* plantation. Stepwise linear regression analysis suggests the concentrations of water-soluble Al and total Al content explain the most variance associated with nematode assembly; whereas, the abundance of early-successional nematode taxa was explained mostly by soil moisture, soil organic C and total N rather than the concentrations of different forms of Al. In contrast, no significant main effects of either Al or soil physico-chemical characteristics on soil microbial biomass were observed. Our study suggests that vegetation was the primary driver on soil nematodes and microorganisms and it also could regulate the sensitivity of bio-indicator role mainly through the alteration of soil Al and physico-chemical characteristics, and *S. cumini* is effective for amending the Al contaminated soils.

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Introduction

The complex soil food web plays important role in soil carbon transformation and nutrient cycling (Coleman et al. 2004). Soil microbial communities are strongly influenced by soil-dwelling invertebrates such as nematodes and microarthropods (Scheu et al. 2005; Fu et al. 2005). Soil nematodes are important invertebrates in soil ecosystem with profound effects on microbial community and organic matter decomposition (Anderson et al. 1981; Freckman

1988; Yeates and Bongers 1999; Neher 2001; Fu et al. 2005). Many studies suggest that nematodes are useful bioindicators in soil and aquatic ecosystems, especially for assessment of ecosystem health (Bongers and Ferris 1999; Ferris and Bongers 2006; Shao et al. 2008). The relationship between indicator values to soil function and soil properties is well documented (Yeates et al. 1993; Goralczyk 1998; Bongers and Ferris 1999; Ekschmitt et al. 2001; Neher et al. 2012). Nematodes early in ecological succession (Fu et al. 2000) are relatively tolerant to contamination by heavy metal pollution in contrast to later successional taxa that are known to be sensitive to heavy metals (Bongers and Bongers 1998; Georgieva et al. 2002; Shao et al. 2008). Often, bacteria and bacterivorous nematodes are considered to be in an earlier stage of ecological succession than fungi and fungal-feeding nematodes. Omnivorous and predaceous nematodes are considered late-successional taxa (Neher 2001). Basal components of the detritus-based trophic system, soil microorganisms and nematodes that feed upon those microorganisms play an important role in soil formation, nutrient cycling and contaminant degradation; and they are sensitive to the changes in soil texture, moisture, temperature and nutrient status (Madsen et al. 1991; Fierer et al. 2003; Fu and Cheng 2004).

The topic of ecological linkages between aboveground and belowground biota has received much attention over the past decade (Van der Putten et al. 2001; Wardle et al. 2004). In general, plants are considered to play a key role in determining soil biota because they are the main food sources (i.e., leaf litter, dead roots and root exudates) to soil biota, which may exert a bottom-up control on soil organisms. Soil organisms are intimately tied to plant communities and indirectly by the decomposition of dead organic plant material (Sylvain and Wall 2011).

Oil shale mining results in severe ecological and environmental problems, including the production of waste materials, soil acidification and contamination by heavy metals. The combinations of these adverse physical and chemical features create a hostile environment to soil organisms (Xia 2004). The response of soil microbial communities and soil nematodes assemblages to soil contamination under the different management treatments would be a basis to evaluate the environmental pollution and ecological restoration. The metal of greatest concentration in oil shale waste is Al (>1,000 mg kg⁻¹) with small amounts of As, Cr, Cd, Pb, Ni, Mn, Zn and Cu (Xia et al. 2004). Aluminum toxicity is a major growth-limiting factor for crop cultivation in acid soils and many studies have demonstrated the suppressive effects in different plant species of soluble Al on root and shoot growth and water and nutrient uptake (Kochian 1995; Clark 1997; Rufyikiri et al. 2000). In addition, soil microorganisms or nematodes were more sensitive than roots as indicators of subtle chemical changes

in soil or water solution with Al toxicity (Williams and Dusenbery 1990; Joner et al. 2005). However, integrated studies about Al pollutants and their impact at soil food web levels (micro-food web: microbes and nematodes) in varied vegetated lands is not documented.

Approaches to ecosystem restoration after oil shale mining mainly include: (1) planting trees or shrubs directly on the untreated waste dump, (2) covering the waste dump with top soil followed by planting trees or shrubs, and (3) treating the waste dump with chemical methods followed by re-vegetation (Xia et al. 2004). However, the contributions of linkages between vegetation and soil organisms in the recovery process and the indicator function of microorganisms in soil overlaid with oil shale waste are poorly understood.

Maoming petro-chemical company, affiliated with the China petro-chemical corporation, is regarded as one of the China's largest nationally graded petro-chemical companies with respect to mining and refining oil from oil shale. However, the company discharged large amounts of oil shale waste into the surrounding environment from the 1970s through the 1990s. Oil shale waste has become one of the most conspicuous environmental issues in this region. The discarded solid waste not only occupied large areas of land, but also resulted in off-site pollution from its drainage. A huge oil shale deposition site (667 ha area with depth >10 m) is situated in the north suburb of Maoming city, approximately 10 km from the city center. From 1971 to 1992, this tract of land was used to receive oil shale waste and oil shale-containing surface soils, recovered from oil shale excavation, separation, combustion, and refining.

Since 2000, restoration projects sponsored by different governmental agencies have been conducted in this area, mainly restoring the vegetation by planting various pioneer tree species onto the shale waste. Five sites have been established, each with a different vegetation type: monoculture plantation of *Syzygium cumini* (site 1), monoculture plantation of *Acacia auriculiformis* (site 2), monoculture plantation of *Schima wallichii* (site 3), naturally recovered grassland (site 4), and oil shale waste (site 5). Density of canopy trees in all plantation sites exceeded 70 %. Composition of understory vegetation varied among the three plantation types. *Eupatorium odoratum*, *Dicranopteris linearis*, and *Blechnum orientale* were the dominant understory species in the *S. cumini* plantation, covering 10–20 % of the soil surface. Understory vegetation covered 10 % of the soil surface in the other two plantation types. Dominant understory species were *Borreria latifolia*, *Clerodendron cryptophyllum*, and *Bridelia tomentosa* in the *A. auriculiformis* plantation and *B. latifolia*, *E. odoratum*, and *C. cryptophyllum* in the *S. wallichii* plantation. The grassland contained *D. linearis*, *Imperata cylindrical*,

E. odoratum, and *B. latifolia* which covered the soil surface by 80–90 %. Understory vegetation coverage is <0.5 % in the control.

We analyzed Al toxicity, soil microbial community, and soil nematodes assemblages under the different management treatments in soil overlaid with oil shale waste. Our primary objective was to quantify the response of the microbial and nematode communities to toxic concentrations of aluminum in vegetated oil-shale-waste lands.

Materials and methods

Site description and soil sampling

Maoming city is located in the southwest of Guangdong province (21°25′–22°42′N and 110°21′–111°46′E), China. Its geographical position is transitioning from the tropics through the low subtropics. The climate of the region is marine monsoon. The mean annual temperature is 23.2 °C, and the mean monthly maximum and minimum temperatures are 28.4 °C (July) and 15.5 °C (January), respectively. The mean annual precipitation is 1,567 mm, with distinct dry (from October to March) and wet (from April to September) seasons. The regional native vegetation is tropical rain forest but it has been mostly converted into other land uses since the 1960s (Xia 2004). Soil moisture ranged from 13.5 to 30.4 % across all sites, it was greatest in the *S. wallichii* and least in the grassland site. Soil pH was in the range of 4.0–4.4 across all sites and it was greater in the *S. wallichii* plantation, *S. cumini* plantation and grassland sites than that in the *A. auriculiformis* plantation and oil shale waste sites. Total soil organic C was in the range of 1.9–7.0 % across all sites. Soil total N ranged from 0.1 to 0.4 % across all sites. The soil C/N ratio was the greatest in the oil shale waste site (Table 1).

In each site (vegetation type), we set three transects for sample collection. The area of each transect was approximately 400–600 m². In June 2008, 12 soil cores (5 cm in diameter) were taken from randomly selected locations

within each transect at 0–5 cm depth. Three soil cores were combined to form one composite sample. In total, there were four composite samples from each transect. As a result, 60 composite samples (five sites × three transects × four composite samples) were taken. Litter was removed from soil surface before the soil samples were taken.

Soil analyses

Soil pH was determined in 1:2.5 (w/v) soil solutions, and soil moisture was measured gravimetrically by drying fresh soil at 105 °C to constant weight (Liu et al. 1996). Soil organic C was determined by the dichromate oxidation method and total N was measured with an ultraviolet spectrophotometer after Kjeldahl digestion (Liu et al. 1996). Total soil Al concentration was determined by inductively-coupled plasma atomic emission spectrometry (ICP-AES) after acid digestion (McGrath and Cunliffe 1985). The sequential extraction scheme was determined with the modified BCR program (Quevauviller et al. 1993); the metals were divided into five fractions: the water-soluble fraction (directly available for plant uptake), the acid extractable fraction (exchangeable metal and carbonate-associated fraction, potentially available for plant uptake), the reducible fraction (Fe and Mn oxide-associated, potentially available for plant uptake), the oxidisable fraction (bound to organic matter, potentially available for plant uptake), and the residual fraction (insoluble form) (Walter et al. 2006).

Phospholipid fatty acids (PLFA) analysis was conducted using the method described by Bossio and Scow (1998) and measured by gas chromatography. The biomass of bacteria was determined using the sum of fatty acids *iso* 15:0, *anteiso* 15:0, 15:0, *iso* 16:0, 16:1 ω 9c, *iso* 17:0, 17:0, *anteiso* 17:0, 17:0cy, 18:1 ω 5c and 19:0cy. The biomass of fungi was determined as the sum of 18:1 ω 9c, 18:2 ω 6c and 18:2 ω 9c. It was reported that PLFA 16:1 ω 5 has high specificity to AM fungi (Olsson et al. 1995). The biomass of arbuscular mycorrhizae was determined by 16:1 ω 5c (Bossio et al. 1998; Mikola and Setälä 1998; Kowalchuk et al. 2004).

Table 1 Soil physico-chemical characteristics of different sites

Site no.	Vegetation type	Soil moisture (%)	Soil pH	Total soil organic C (%)	Total soil N (%)	Soil C/N ratio
1	<i>S. cumini</i>	19.8 ± 1.8b	4.4 ± 0.2a	2.5 ± 0.6b	0.1 ± 0.0bc	20.3 ± 3.0bc
2	<i>A. auriculiformis</i>	26.2 ± 2.9a	4.1 ± 0.0b	7.0 ± 1.9a	0.4 ± 0.1a	19.3 ± 1.5bc
3	<i>S. wallichii</i>	30.4 ± 2.2a	4.4 ± 0.0a	3.8 ± 1.1ab	0.2 ± 0.0bc	21.0 ± 2.5ab
4	Grassland	13.5 ± 4.7c	4.3 ± 0.1a	1.9 ± 0.6b	0.1 ± 0.0c	18.4 ± 2.9c
5	None (oil shale waste)	22.6 ± 0.7b	4.0 ± 0.0b	4.4 ± 0.2a	0.2 ± 0.0b	27.1 ± 2.1a

Data are mean ± SE

Different letters indicate significant differences among sites

Nematode analyses

For each composite soil sample, nematodes were extracted from 50 g of fresh soil using Baermann funnels (Barker 1985). After fixation in 4 % formalin solution, nematodes were counted under an inverted microscope and the first 100 individuals encountered were identified to genus or family and classified into trophic groups (plant-feeders, bacterial-feeders (Ba), fungal-feeders (Fu), predators and omnivores) (Yeates et al. 1993) and functional guilds (Ba1, Ba2, Ba3, Ba4, Fu2, Fu3, Fu4, etc.) (Ferris et al. 2001). The functional guild designation is defined by the nematode's trophic behavior and by its response to the environment as a colonizer (c-p1 to c-p2) or persister (c-p3 to c-p5) (Bongers and Bongers 1998). All nematodes were identified when the nematode were fewer than 100 individuals in a sample.

Nematode faunal analysis, based on a weighted matrix classification of life traits and feeding habits, provides a qualitative measure of the soil food web. The structure index (SI) is based on the relative weighted abundance of disruption-sensitive guilds representing structure (s); the enrichment index (EI) on opportunistic bacterial- and fungal-feeding nematodes representing enrichment (e). At the base of both indices are taxa tolerant of adverse conditions and basal (b) to all nematode assemblages. SI and EI were calculated using the following equations: $SI = 100(s/(s + b))$ and $EI = 100(e/(e + b))$, respectively, the s, e and b component are calculated as $\sum k_s n_s$, $\sum k_e n_e$, $\sum k_b n_b$, respectively, where k_i are weights applied to the indicator importance of each functional guild of nematodes. When SI is plotted against EI for a sample of nematodes, the resulting graph can be divided into four quadrats that are descriptive of food web characteristics (Ferris et al. 2001; Ferris and Matute 2003).

Statistical analysis

One-way ANOVA were performed to determine the effect of site type on nematode community, microbial community, and soil chemistry. Ad hoc means comparisons among the five site types were performed by least significant difference (LSD). Linear stepwise regression analysis was performed to quantify the response of the soil microbial biomass or soil nematode community assemblage to soil physico-chemical characteristics (SC), microbial community (MB), and concentrations of different form of Al (AL). To avoid collinearity among related variables, principal component analyses (PCA) were performed separately for each SC, MB and AL variables. PCA axes (proxies for SC, AL and MB) were used as independent variables in the linear stepwise regression analysis. To meet assumptions of normality, results of fungal PLFAs and total PLFAs were

transformed as $\ln(x + 1)$. Statistical significance was determined at $P < 0.05$. All statistical analyses were performed using SPSS software (SPSS Inc., Chicago, IL).

Results

Soil microbial PLFAs

Total microbial biomass was the greatest in the *S. cumini* plantation and *A. auriculiformis* plantation and the least in the *S. wallichii* plantation site. Bacterial biomass was the greatest in the *S. cumini* plantation site; no significant differences were found among other sites. Fungal biomass was also the greatest in the *S. cumini* plantation and the least in the *S. wallichii* plantation site. Biomass of arbuscular mycorrhizal fungi was only detected in the *S. cumini* plantation site (Fig. 1).

Nematode assemblage

The total nematode density was the greatest in the *A. auriculiformis* plantation site; no significant differences were found among other sites. However, the abundance of predatory nematodes and plant-feeding nematodes was the greatest in *S. wallichii* plantation site. The abundance of bacteria-feeding and fungi-feeding nematodes was the greatest in *A. auriculiformis* plantation; the abundance of fungi-feeding nematodes was the lowest in *S. cumini* plantation. No significant differences of the abundance of omnivores were found among different sites (Table 2).

The graphic presentation of "weighted faunal analysis" showed that *S. cumini*, *S. wallichii*, *A. auriculiformis* and grassland were mapped in quadrat B with the oil shale waste site mapped in quadrat A. Nematode community composition was similar among all vegetated sites and contrasted with the control (Fig. 2).

Soil Al concentration

The water-soluble Al concentration was greater in the *S. cumini* plantation site than that in oil shale waste site. No significant differences of water-soluble Al concentrations were found among the other sites. Acid extractable Al concentrations were greatest in the *S. wallichii* plantation, moderate in the *S. cumini* plantation, *A. auriculiformis* plantation and grassland sites. Aluminum concentrations at the *S. wallichii* plantation and oil shale waste site were similar. Reducible Al concentrations were the greater in the *S. wallichii* plantation site than the other four sites. Oxidizable Al concentrations were greater in the *S. wallichii* plantation than that in *S. cumini* plantation and grassland sites but were similar among the *S. wallichii* plantation,

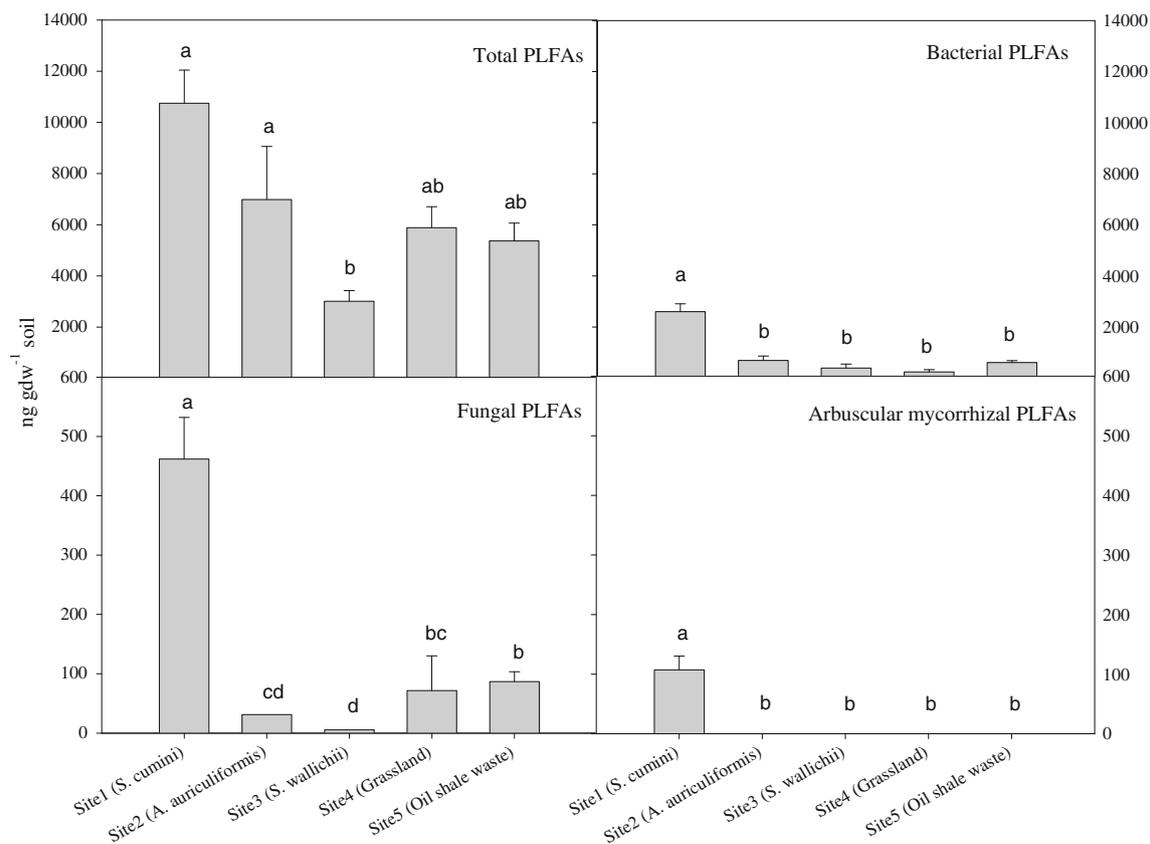


Fig. 1 Soil microbial community characteristics. Data are mean \pm SE. Means followed by different *letters* indicate significant differences among sites

Table 2 Abundance of nematode trophic groups for different sites

	Abundance (ind 100 g ⁻¹ dry soil)					
	B*	Fu*	H*	Om*	P*	T*
Site 1 (<i>S. cumini</i>)	300.4 \pm 102.7b	73.5 \pm 5.4b	29.6 \pm 13.9b	53.4 \pm 27.2a	0.2 \pm 0.2c	457.1 \pm 118.3b
Site 2 (<i>A. auriculiformis</i>)	823.6 \pm 209.2a	241.6 \pm 74.5a	45.1 \pm 23.6b	105.2 \pm 53.1a	0.0 \pm 0.0d	1215.5 \pm 297.5a
Site 3 (<i>S. wallichii</i>)	290.0 \pm 66.4b	192.2 \pm 54.6ab	121.8 \pm 16.3a	159.0 \pm 53.5a	65.8 \pm 56.4a	828.8 \pm 109.4ab
Site 4 (Grassland)	149.0 \pm 40.0b	129.8 \pm 41.0ab	41.3 \pm 12.6b	121.3 \pm 41.5a	5.5 \pm 3.0b	446.9 \pm 138.0b
Site 5 (Oil shale waste)	205.3 \pm 69.7b	202.8 \pm 39.1ab	27.7 \pm 1.3b	38.8 \pm 16.3a	2.5 \pm 2.2bc	477.1 \pm 131.5b

Data are mean \pm SE

Different *letters* indicate significant differences among sites

* H, P, Om, B, Fu and T refer to plant feeders, predators, omnivores, bacterial feeders, fungal feeders, and total respectively

A. auriculiformis plantation and oil shale waste sites. The residual Al concentrations and total Al concentrations showed a similar trend and it was greater in the oil waste site than that in the *S. cumini* plantation site and there were no significant differences among the other sites (Fig. 3).

Relationships between soil biota and environmental variables

AL2 (water-soluble Al and total Al) was most important in determining the abundance of bacteria-feeding nematodes.

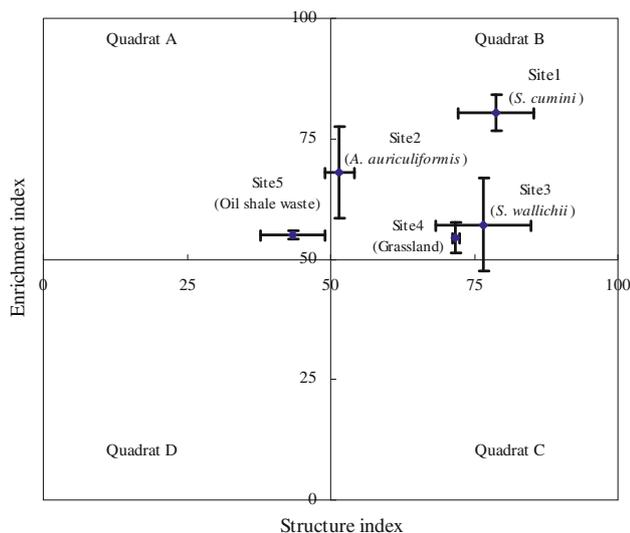


Fig. 2 Faunal analysis for all sites. Means and SE for enrichment and structure indices are shown

Water-soluble Al and total Al was related positively with the abundance of bacteria-feeding nematodes ($P = 0.043$). The MB2 (total PLFAs) was related negatively with the abundance of plant-feeding nematodes ($P = 0.002$). AL1 (acid extractable Al, reducible Al and oxidisable Al) was related positively with the abundance of predators ($P < 0.001$). Abundance of early-colonizing nematode taxa (c-p2) was determined by SC1 (soil moisture, soil organic C and soil total N) ($P = 0.031$) rather than different form of Al. Abundance of later-successional nematode taxa (c-p4) was best determined by both AL2 (water-soluble Al and total Al) and MB2 (total PLFAs) and it was related negatively with water-soluble Al, total Al and total PLFAs ($P < 0.001$). Abundance of late-successional nematode taxa (c-p5) nematodes was not related to any environmental variables because of the infrequent occurrence of functional guilds belonging to the c-p5 guild. The microbial biomass did not appear to be affected by environmental variables (Tables 3, 4).

Discussion

Impacts of aluminum toxicity on microbes and nematodes

Toxic concentrations of aluminum had an effect on nematode communities and the microbial biomass did not appear to be affected by soil characteristics and Al toxicity.

The differences in response to Al toxicity between soil microbial and nematode communities in varied vegetated

lands possibly because functional groups of microbial communities indicated by the PLFA biomarkers differ in their importance and activity so that estimates of bacterial, fungal and total biomass do not reflect activity. Furthermore, plant specific differences as well as seasonal changes in plant physiology have an influence on aluminum toxicity in acid forest soils (Collignon et al. 2012). Our sampling time represents a single point in time and differences of seasonal changes in plant physiology of various plants probably also contribute to differences. The concentration of water-soluble Al (bioavailable) was from 74 to 446 mg kg⁻¹ dry soil in the habitats with vegetation and it was greater in vegetated land than non-vegetated land (it was only 5 mg kg⁻¹ dry soil), especially under the fast-growing *S. cumini* plantation, which was highly toxic and exceeded background values of healthy soils (Xia et al. 2004). The toxicity of Al solution on nematodes has been tested in previous studies. The LC50 concentrations of Al ion to *Caenorhabditis elegans* are 79, 2, 1.9 and 1.8 mg L⁻¹ at 24, 48, 72 and 96 h, respectively (Williams and Dusenbery 1990). In the present study, we found that the water-soluble Al negatively affected nematode assemblage although it accounted for a small proportion of the soil total Al which was normally ignored in most published literature. However, no direct toxic effects of Al on communities of both nematode and microorganism were observed in terms of normally tested forms of Al, i.e., acid extractable Al, reducible Al, oxidisable Al etc. although they might cause potential toxic effects in the long run.

The negative association between concentration of Al in soil and abundance of late-successional nematodes is consistent with the findings in previous studies (Zullini and Peretti 1986; Bongers 1990; Korthals et al. 1996; Bongers and Ferris 1999). For example, Zullini and Peretti (1986) and Korthals et al. (1996) found that late successional nematodes, omnivores and predators, were the most sensitive group to heavy metals. Omnivorous and predatory nematodes are considered to be highly sensitive to environmental disturbance because of their large body size, long life cycles and low reproduction rate (Bongers 1990; Bongers and Ferris 1999). Previous studies showed contrasting results when using c-p2 nematodes as bio-indicator of Cu and Pb contamination (Georgieva et al. 2002; Sánchez-Moreno et al. 2006), suggesting that using the c-p2 nematodes to assess the effects of heavy metal contamination is in doubt. No correlation was found between early-successional nematodes and water-soluble Al and total Al in the present study, the abundance of early-successional nematodes might be determined by soil moisture, soil organic C and soil total N rather than different form of Al. On one hand, the Al toxicity could lead to the reduction of high trophic-level nematodes directly; on the other hand, the abundance of low trophic-level (i.e. primary colonizers)

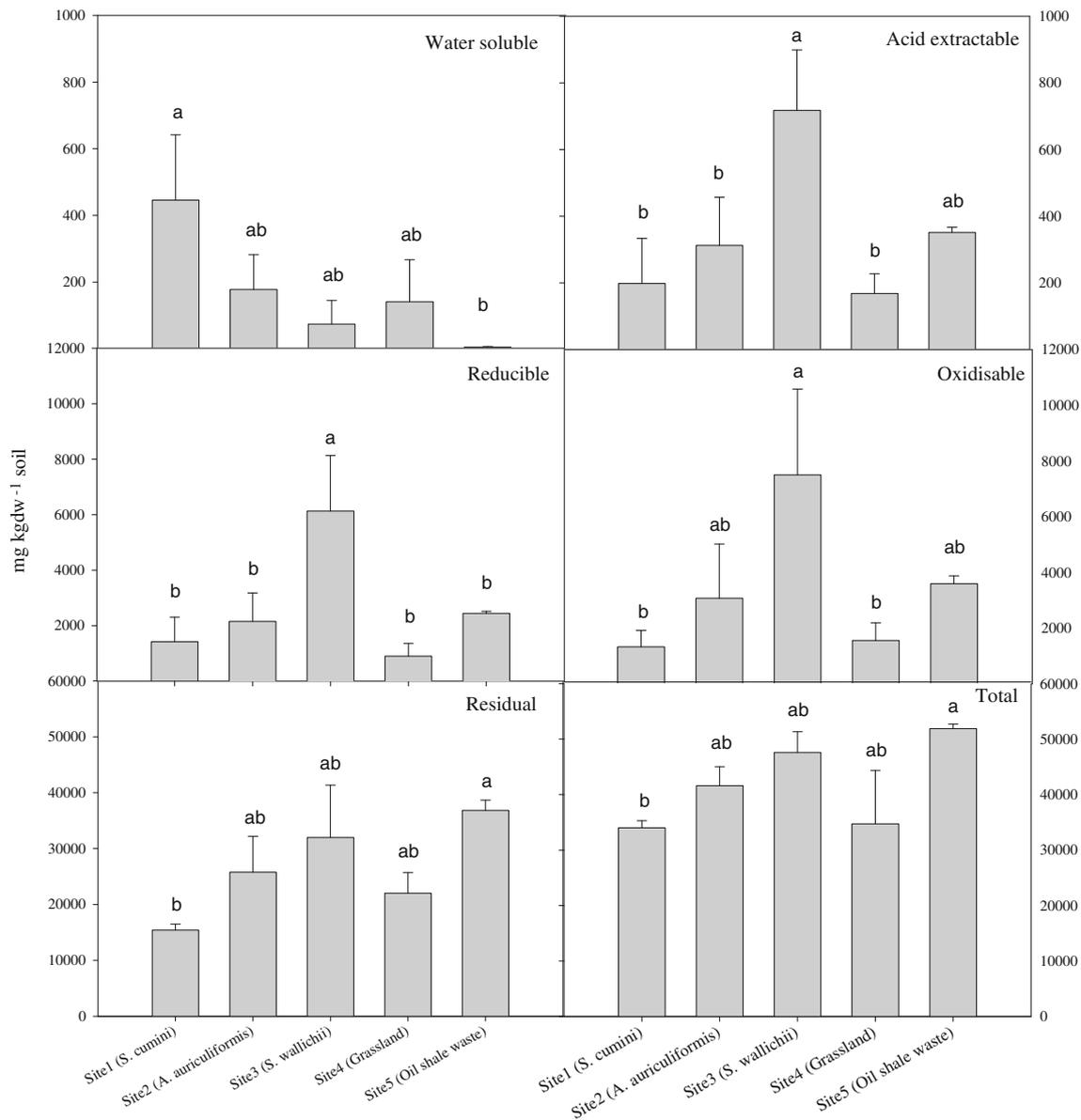


Fig. 3 Concentrations of different forms of Al for different sites. Data are mean \pm SE. For each panel, *different letters* indicate significant differences among sites

nematodes would also be decreased due to the reduction of the plant photosynthates affected by Al toxicity. However, the abundance of late-successional nematodes was not related to any environmental variables due to infrequent occurrence of functional guilds belonging to the top predator guild. Some results remain uncertain, such as the negative relationship between total PLFAs and the abundance of plant-feeding nematodes. Nematodes grazing on soil microbes, and they are also grazed by other soil biota (such as some predacious mites). Interactions among soil organisms seem to contribute to the complexity of the some results at the different vegetated sites.

Possible mechanisms of effect of Al toxicity on soil microbial and nematode communities in different vegetation sites

Presence and type of vegetation affects soil communities because plants are the major food sources (i.e., leaf litter, dead roots and root exudates) to soil biota (Zhao et al. 2011). The greater bacterial, fungal and AM fungal biomass in the soils of the *S. cumini* plantation, greater abundance of nematodes in the soils of the *A. auriculiformis* plantation and greater abundance of predatory nematodes in the soils of *S. wallichii* plantation suggest that soil microorganisms and

Table 3 The stepwise linear regression parameters of variables in each pathway using linear regression model and weighting of each variable in the three suites of variables (microbial, physico-chemical, nematode communities) using principal component analysis (PCA)

Parameters	Principal components	
	PCA1 (SC1)	PCA2 (SC2)
Soil physico-chemical characteristics (SC)		
Soil moisture (SM)	0.774	0.113
Soil pH	-0.106	-0.873
Total soil organic C (SOC)	0.922	0.289
Total soil N (TN)	0.951	0.066
Soil C/N ratio (C/N)	0.176	0.859
Cumulative percent (%)	55.355	24.549
Concentrations of different forms of Al (AL)	PCA1 (AL1)	PCA2 (AL2)
Water-soluble (Ws)	-0.379	-0.818
Acid extractable (Ae)	0.926	0.316
Reducible (Red)	0.963	0.225
Oxidisable (Oxi)	0.952	0.27
Residual (Res)	0.626	0.646
Total	0.127	0.914
Cumulative percent (%)	73.037	16.766
Microbial biomass (MB)	PCA1 (MB1)	PCA2 (MB2)
Total PLFAs (TP)	0.381	0.924
Bacterial PLFAs (BP)	0.856	0.458
Fungal PLFAs (FP)	0.915	0.382
Arbuscular mycorrhizal PLFAs (AP)	0.925	0.36
Cumulative percent (%)	88.14	10.03

The principal components whose regression coefficient estimates are statistically significant (the most important variables) are given and shown in bold

nematodes are intimately linked to plant communities. Nematode community composition was similar among all vegetated sites and contrasted with the oil shale waste site. Plants species seems to be the factor that most strongly determines effect of Al toxicity on soil microbial and nematode communities based on the results of planting different plant species on the waste lands. Yang et al. (2012) found that root organic acid exudates may affect composition of microbial communities and the organic acids secreted by various plants were different. In addition, soil microbes can also produce organic acids, low pH and Al mobilization can affect the turnover of microbial populations (Fierer and Jackson 2006). In the present study, we speculate that root organic acids secreted by various plants affect the composition, activity and turnover rate of microbial communities; subsequently, soil nematodes in the food chain would be inevitably affected by the changes of soil microbes.

Soil food web structure was strongly affected by soil organic matter and total N. The abundance of nematodes

Table 4 The regressions of microbial biomass (with all principal components in the SC and AL), nematode abundance of trophic groups and guilds (with all principal components in the SC, AL and MB) using stepwise linear regression model

Parameters	Variable	r^2	P
Total PLFAs	-	-	-
Bacterial PLFAs	-	-	-
Fungal PLFAs	-	-	-
AM Fungi	-	-	-
Bacteria-feeding nematodes	-AL2(ws_Al, tot_Al)	0.279	0.043
Plant-feeding nematodes	-MB2(tot_PLFAs)	0.521	0.002
Fungi-feeding nematodes	-	-	-
Predators	AL1(Ae_Al, Red_Al, Osi_Al)	0.71	<0.001
Omnivores	-	-	-
Cp-1	-	-	-
Cp-2	SC1(SM, SOC, TN)	0.312	0.031
Cp-3	-	-	-
Cp-4	-AL2-MB2(ws_Al, total_Al, tot_PLFAs)	0.691	0.001
Cp-5	-	-	-
Abundance of total nematodes	-	-	-

ws_Al water-soluble Al, tot_Al total Al, tot_PLFAs total PLFAs, Ae_Al acid extractable Al, Red_Al reducible Al, Osi_Al oxidisable Al, SM soil moisture, SOC soil organic carbon, TN total soil nitrogen

was greatest, but its structure index (SI) value was less in *A. auriculiformis* plantation than those in other sites, and we considered the Al toxic effects on soil food web was compensated by the status of soil fertility. The soil organic matter and total N contents were the highest in *A. auriculiformis* plantation compared to other sites. Apparently, higher contents of soil organic matter and soil N in N-fixing *A. auriculiformis* plantation contributed to the highest abundance of nematodes, especially the early-successional bacterivorous nematodes. We believe that toxic effects of Al on soil nematodes can be mitigated by soil enrichment and the linkages of soil biological communities and vegetation may complicate the direct effects of Al toxicity on soil nematodes or microbial communities.

Although, there are greater SI values in the nematode assemblage and greater abundance of predatory nematodes in the *S. wallichii* plantation, biomass of microbial organisms was less at this site. In addition, most plant-feeding nematodes have negative effects on ecosystem services and there is greater abundance of plant-feeding nematodes in the *S. wallichii* plantation, suggesting that the soil health status is not very well at this site.

In the *S. cumini* plantation, the water-soluble fraction of Al was the greatest; however, the results of nematode faunal

analysis and greater bacterial, fungal and AM fungal biomass indicate a better soil food web condition. In addition, the residual Al concentrations and total Al concentrations showed a significant decrease after planting *S. cumini* plantation onto the shale dump, which suggests that *S. cumini* tree species is effective for amending the Al contaminated soils. AM fungi have been reported to be effective in alleviating Al toxicity to plants by enhancing the nutrient and water acquisition in metal-contaminated soils although it may not degrade the metal toxicity directly (Clark 1997; Meharg and Cairney 2000; Rufyikiri et al. 2000; Wu 2008). Planting trees associated with mycorrhizal fungi seems to be a robust and promising bioremediation strategy for many polluted soil sites (Banitz et al. 2011). We speculate that the greater AM fungi biomass in *S. cumini* plantation could be responsible for the better soil food web condition.

Sensitivity of indicator values

It seems that vegetation was the primary driver for soil microorganisms and nematodes in the Al contaminated ecosystems mainly through the alteration of soil Al and other soil physico-chemical characteristics. In the present study, in the *S. cumini* plantation, the greater AM fungi biomass could alleviate Al toxicity; the trophic interactions appear to be unaffected by Al contamination. In that case, our results were consistent with the findings of previous studies that demonstrate nematodes and other soil organisms (such as microorganisms, tardigrades or mites) are similar in environmental sensitivities and indicator roles (Sánchez-Moreno et al. 2008, 2009). Al toxicity could lead to a reduction of high trophic-level predatory nematodes in soil ecosystems due to their sensitivity to the environmental contamination in the *A. auriculiformis* plantation (Bongers 1990; Bongers and Ferris 1999). In contrast, better soil nutrient status could contribute to the increased abundance of low trophic-level nematodes. In that case, nematodes seem to be more suitable for indicating the soil health status than some high trophic-level soil organisms (such as collembolans or mites). Although, there are greater SI values in the nematode assemblage in the *S. wallichii* plantation, biomass of microbial organisms was less at this site, suggesting that nematodes are more suitable for indicating the soil health status than primary decomposers (such as soil microorganisms).

The accumulated evidence suggests that sensitivity of indicator values change with aluminum concentration and vegetation could regulate the bio-indicator role of soil food web. In particular, an approach with an integrative analysis of soil microbes, nematodes and other high-level soil organism would be a more robust reflection of soil condition.

Limited scope of extrapolation

The results came from one region and needs to be confirmed by testing in other regions. Furthermore, our sampling time represents a single point in time, whereas soil microbial and nematode communities might fluctuate due to the changes of seasons. In addition, the Baermann funnel for extraction might bias against larger and/or more sedentary nematodes. However, the results clearly show integrated studies between pollutants and their impact at soil food web levels are interesting and linkages between vegetation and the soil organisms could be important in mitigating the effects of Al toxicity.

Conclusion

We investigated the relationship among nematode community structure, soil microbial community composition and soil aluminum content in varied vegetated aluminum-rich lands, the present study concludes that linkages of soil biological communities and vegetation may mitigate the negative impacts of soil Al and enhance the positive effects of soil fertility on the soil food web. Vegetation could regulate the bio-indicator role of soil food web. Moreover, planting trees associated with mycorrhizal fungi, i.e., *S. cumini*, seems to be a promising bioremediation strategy for Al contaminated soils.

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