

Community Structure of Mites in PAH-contaminated Field Soils in Eastern China

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Abstract: Soil mites are good indicators for soil quality assessment, but they rarely have been used to indicate pollution levels of soil polycyclic aromatic hydrocarbons (PAHs). In former studies, mite community structure parameters (usually at group level) such as density, group richness, diversity index and evenness were mainly used as indicators. Since these indicators were established on coarse taxonomic resolution, the assessment effects have usually been found to be variable and unstable. The present study aims at finding more sensitive parameters of soil mite communities to PAHs among individual species, sex ratio and life history traits. Effects of PAH contamination on mite community structure were investigated in vegetable fields in Eastern China December, 2008, by contrasting the effects observed in the polluted soil with the reference study sites. Soil properties and heavy metals were examined as well as the PAH concentrations and mite community structure parameters. Total concentration of PAHs in polluted sites was ca. 18.7 to 26.8 mg kg⁻¹ dw soil, being 5.3 to 7.6 times of that in reference study site. Density and species richness of Macropylina, (Oribatida), in polluted sites were 3.2 to 8.5 and 1.0 to 2.2 times more than those in reference site, respectively. Oppositely, 1r-species density, male density and sex ratio of Mesostigmata were higher in reference site than in polluted sites by 0.9 to 1.1, 2.2 to 3.3 and 1.0 to 2.6, respectively, and negatively correlated with total PAH concentration. The results suggest that Macropylina and 1r-species, male individuals and sex ratio of Mesostigmata may be sensitive to soil PAH pollution directly or indirectly and can be further developed as indicators for assessment of soil pollution.

Key words: Acari; soil pollution; bio-indicator; species diversity; sex ratio; life history

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Polycyclic aromatic hydrocarbons (PAHs) are substances potentially genotoxic and carcinogenic, usually resulting from coke production, petroleum refining, fossil fuel combustion and other high-temperature industrial processes^[1], they are widespread in the environment, and their contamina-

tion is a serious problem on a large number of sites worldwide^[2]. PAHs can be produced from incomplete combustion. They are highly lipophilic semi-volatile organic compounds partitioning between the atmospheric gas-phase and particle-phase^[3] and the PAHs entering the airshed can be deposited directly onto

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soils or first absorbed by vegetation and then incorporated into the soil when vegetation enters the ground at the end of the growing season and decays^[4]. Soil PAH pollution is also serious in China. In Eastern China, the average concentrations of PAHs can be over 1000 ng g⁻¹ in soils around Shanghai city^[5] and Xiamen city of Fujian province^[6] and ca. 800 ng g⁻¹ in Jiangsu province^[7-8]. However, how PAHs affect soil ecosystems is still not very well understood.

Soil organisms may be useful in identifying clean-up priorities and monitoring environmental change because they provide objective metrics that integrate physical, chemical, and biological parameters^[9]. Soil invertebrates are popular candidates as biological indicators because of their role in essential ecological processes of soils including nutrient cycling and decomposition^[10-11]. The relative abundance and ubiquitous nature of soil invertebrates contribute to their usefulness in comparing their community structure among ecosystem types and land management practices^[12-13]. Soil mites are among the soil invertebrates having potential as biological indicators of soil health or conditions. Previous studies have shown that soil predatory mites belonging to Mesostigmata are sensitive to changes in management practices and type of land use^[12, 14-17]. Oribatida and Gamasina mites have also been evaluated as potential indicators of polluted soils by pentachlorophenol (PCP)^[18-19] and heavy metals^[12, 20-21].

Only a few studies have involved the effects of PAHs on soil mites. These studies have mainly focused on total populations of soil mites and the effects usually been found to be variable, and it is difficult to identify the mechanisms involved^[9, 22]. This is because of the coarse taxonomic resolution used in these studies, since different species within one family or order may have contrasting responses to PAH stress^[9, 20]. Therefore, more sensitive parameters of soil mite communities are required to provide better utility of mite communities as biological indicators.

The aim of the present study was to screen for more sensitive parameters of soil mite communities to PAHs in field condition in terms of species level, sex ratio and life history traits. Mites in PAH polluted and reference study sites were sampled and the community structures parameters between the polluted and reference sites were compared. We expected that: (1) mite

community composition parameters at species level, such as density, species richness, diversity index and evenness, will be higher in the reference than in polluted sites, i.e. community parameters at species level will be sensitive to soil PAH pollution; (2) biological parameters of life history features, such as *r* or *K* selected species, and sex features, such as male individuals and sex ratio, will be sensitive to the PAHs.

1 Materials and methods

1.1 Study sites and sampling design

The study sites are located in vegetable fields close to Wuxi city, Jiangsu province, Eastern China (31°36'08"-14° N, 120°28'38"-53° E). The PAH pollution source is a forge that has been producing small iron wares with soft coal since 1992 and the coal consumption is ca. 1 to 2 tons per day. Two PAH polluted and one reference study sites were investigated. The two polluted sites are 20 (Pol1) and 80 (Pol2) meters south of the forge and the reference site (Ref) is 500 meters east of the forge. In each study site, four plots each of ca. 5 m² were set up as four replicates with their centers 10 to 15 meters from each other. Seedlings of Chinese mustard (*Brassica chinensis*) were growing in the plots.

Sampling was conducted in December 2008. Three soil cores, each 20 cm² in area × 5 cm deep, were randomly sampled in each plot and then mixed together to give one composite sample for analysis of soil physicochemical properties and PAH compounds, giving a total of 12 samples for the three study sites, each with 4 plots (replicates). Three additional soil cores (20 cm² in area × 10 cm deep) were sampled from each plot adjacent to the previous cores and combined to give a second composite sample per plot for examination of soil mites.

1.2 Soil physical and chemical properties

The soil was a sandy loam (Typic Endoaquepts)^[23]. Soil texture was determined by the hydrometer method^[24], organic carbon content by dichromic oxidation followed by titration of excess K₂Cr₂O₇ with FeSO₄ (ISO 14235-1998), nitrogen by the Kjeldahl method^[24] (BUCHI KjelFlex K-360), pH by electrode in a 1:5 (m/V) suspension of soil in deionized water, and electrical conductivity by potentiometry in a 1:5 (m/V) suspension of the dissolved salts of the soil (METTLER TOLEDO Inlab Expert). Water content, water holding capacity and cation exchange capacity

were also determined^[24].

Because the forge was producing iron wares, heavy metals in the soil were also determined by atomic absorption spectrophotometry (Varian Spectra AAS-220)^[25].

Extraction of PAHs from the soil samples followed EPA methodology (3550C) with slight modifications. The soil samples were sieved through a 1 mm mesh and then lyophilized. 2 g of lyophilized soil was added to 10 ml of a mixture of re-distilled hexane and acetone solvent (1:1 v/v), extracted on an ultrasonic machine (KQ-400DB) at 40 kHz for 30 min, and centrifuged at 3000 rpm for 5 min. The procedure was repeated three times and the three extracts were combined and concentrated to 1 ml on a rotary evaporator under 350 mbr at 40 °C. The concentrated extract was passed through an SPE C18 column. The effluent was nitrogen dried, mixed with 2 ml of hexane and analyzed by GC-MS (Agilent 6890GC-5973NMS). An HP5-MS capillary column was used with helium as carrier gas. The oven temperature was programmed from an initial temperature of 60 to 120 °C at a rate of 30 °C/min, and then to 270 °C at rate of 5 °C/min. The injection was operated in splitless mode and the detector temperature maintained at 280 °C. MS detection was based on the selected ion monitoring (SIM) system (EPA method 8270). External and internal methods were used with standards of 16 PAHs and phenanthrene-D10 (Sigma-Aldrich), respectively.

1.3 Soil mites and data collected

Soil mites were extracted from the soil samples using modified Tullgren extractors with temperature increasing from 20 to 50 °C at a rate of 5 °C per day for 6 days^[26]. The mites extracted were identified and counted. All soil mites (total Acari) were identified to species level in the 4 suborders Oribatida, Mesostigmata, Prostigmata and Astigmata using taxonomic keys^[27-34].

The following parameters or indices of soil mite communities were computed and compared between the reference and the polluted sites: 1) general mite community structure: density, species richness, Shannon diversity index and evenness^[16] of the 4 suborders Mesostigmata, Prostigmata, Astigmata and Oribatida as well as total Acari; 2) MGP groups of Oribatida: species in the three Oribatida groups Macropylina (M), Gymnonota (G) and Poronata (P) were sort-

ed and then MGP I index (proportion of species number of each group to that of the three groups together) and MGP II index (proportion of density of each group to that of the three groups together) were calculated^[35]; 3) life history classes of Mesostigmata: frequency of each class species (*K*- and *r*-selected species), and the maturity index (MI): weighted proportion of *K*-selected species in whole Mesostigmata community, with weights for *r*-values of 1, 2, 4, 1, 1 for *r*-selected species in families Ascidae, Digamasellidae, Eviphididae, Laelapidae and Ologamasidae, respectively and weighing factors for *K*-values 2 and 2 for *K*-selected species in Parholaspididae and Rhodacaridae, respectively^[12]; 4) sex ratio of Mesostigmata: male density and species number of male-dependent species and male to female sex ratio of Mesostigmata. Species whose males and females were both found in the same study site were used for the analysis so that the sex ratio for each species could be calculated. Correlations between the soil environmental factors (soil properties, heavy metals and PAHs) and the mite community parameters that were significantly different among the three study sites were analyzed.

1.4 Statistical analysis

Data between the study sites were compared generally by one-way ANOVA and significant differences among the mean values were detected using the Tukey HSD test. Density of the life history classes, the male density and the male to female sex ratio were analyzed by repeated ANOVA (using the species in the same class or the male-dependent species as multiple factors). Correlations between the environmental factors and the mite community parameters were analyzed with the data for plot by multiple regression analysis and Monte Carlo test. Statistical analyses were implemented using programs Statistica v. 7 (StatSoft, Tulsa, OK) and Canoco for Windows v. 4.5.

2 Results

2.1 Soil environmental factors

General soil properties of pH, electrical conductivity, cation exchange capacity, water content and water holding capacity were not significantly different among the three study sites (Table 1). Soil texture and nutrients were significantly different among the sites. Content of 2-0.02 in the reference site (Ref) exceeded that in the 80 meter polluted site (Pol2) by 23.0%, but

content of 0.2-0.002 mm particles in Ref was less than that in the 20 meter polluted site (Pol1) and Pol2 by 17.7% and 14.5%, respectively. Concentration of soil organic carbon in Ref exceeded that in Pol2 by 21.2%, but total nitrogen in Pol1 exceeded that in Pol2 by 23.3%.

Three heavy metals (Cu, Pb and Zn) were detected and the concentrations were low (Table 1); Cu and Pb concentrations were just over the criterion (35 mg/kg) of the lightest pollution and Zn below the lightest criterion (100 mg/kg) for crop soil in the Chinese National Standard. Cu and Pb were significantly

different among the three sites; Cu concentration in Pol2 exceeded that in Ref and Pol1 by 26.1% and 39.8%, respectively, while Pb in Pol1 was less than that in Ref and Pol2 by 46.2% and 36.1%, respectively.

13 of the 16 PAH compounds selected to be first controlled by the US EPA were detected in the soil samples; Naphthalenes of 2-ring and two 3-ring compounds acenaphthylene and acenaphthene were not detected (Table 1). 3-, 4-, 5- and 6-ring compounds accounted for 5.1%, 36.3%, 44.2% and 14.3% of the total PAH concentration, respectively. Total concen-

Table 1 F-values of one-way ANOVA and measurements (mean \pm se, $n=4$) of soil environmental factors (the soil properties, heavy metals and PAHs) for the reference (Ref) and the two polluted (Pol1 and Pol2) study sites

	F _{2,9}	Ref Mean	Se	Pol1 Mean	Se	Pol2 Mean	Se
General properties							
pH value	0.41	5.57	0.07	5.47	0.20	5.77	0.36
Electrical conductivity/($\mu\text{s}\cdot\text{cm}^{-1}$)	2.38	101.95	22.07	177.83	68.60	49.48	7.09
Cation exchange capacity/($\text{cmol}\cdot\text{kg}^{-1}$)	2.45	14.79	0.56	8.52	2.86	13.80	2.32
Water content/%	1.36	16.45	1.31	17.48	0.60	15.49	0.32
Water holding capacity/%	0.87	52.12	1.20	53.75	1.45	54.06	0.45
Texture/%							
2-0.02 mm	6.64*	35.41 ^a	1.90	30.82 ^{ab}	0.47	28.78 ^b	1.18
0.02-0.002 mm	4.42*	41.23 ^b	2.14	48.54 ^a	2.24	47.21 ^a	0.84
<0.002 mm	2.02	23.36	0.38	20.64	2.12	24.01	0.34
Nutrients/ ($\text{mg}\cdot\text{kg}^{-1}$ ·dry soil)							
Organic carbon	5.01*	14.16 ^a	0.77	13.31 ^{ab}	0.06	11.68 ^b	0.59
Total nitrogen	7.76*	1679.08 ^{ab}	34.23	1789.92 ^a	91.94	1451.76 ^b	43.18
Metals/ ($\text{mg}\cdot\text{kg}^{-1}$ ·dry soil)							
Cu	17.18***	40.39 ^b	2.64	36.41 ^b	0.73	50.91 ^a	1.52
Pb	14.10**	48.25 ^a	2.53	33.00 ^b	0.58	45.00 ^a	2.65
Zn	1.48	161.09	55.81	80.40	4.58	111.30	14.99
PAHs ($\text{mg}\cdot\text{kg}^{-1}$ ·dry soil)							
Three rings	8.94**	0.46 ^b	0.10	0.94 ^a	0.09	1.12 ^a	0.15
Fluorene	11.91**	0.00 ^b	0.00	0.02 ^a	0.00	0.02 ^a	0.00
Phenanthrene	8.74**	0.32 ^b	0.11	0.78 ^{ab}	0.08	0.95 ^a	0.13
Anthracene	0.12	0.14	0.03	0.14	0.00	0.15	0.01
Four rings	20.65***	1.26 ^b	0.01	6.35 ^a	0.95	10.19 ^a	1.42
Fluoranthene	19.51***	0.26 ^b	0.01	2.37 ^a	0.41	3.62 ^a	0.52
Pyrene	22.00***	0.17 ^b	0.00	0.79 ^b	0.12	1.80 ^a	0.28
Benz[a]anthracene	16.31***	0.49 ^b	0.00	1.42 ^a	0.19	2.08 ^a	0.29
Chrysene	19.65***	0.34 ^b	0.00	1.78 ^a	0.28	2.69 ^a	0.37
Five rings	19.04***	1.12 ^b	0.04	8.54 ^a	1.32	12.03 ^a	1.77
Benz[b]fluoranthene	19.15***	0.60 ^b	0.01	4.41 ^a	0.76	6.77 ^a	0.97
Benz[k]fluoranthene	19.48***	0.26 ^b	0.01	2.15 ^a	0.37	3.31 ^a	0.48
Benz[a]pyrene	8.17**	0.26 ^b	0.02	1.98 ^a	0.50	1.95 ^a	0.32
Six rings	10.80**	0.68 ^b	0.28	2.86 ^a	0.41	3.47 ^a	0.60
Indeno[1,2,3-cd]pyrene	4.95*	0.13 ^b	0.13	1.41 ^a	0.26	1.42 ^a	0.50
Dibenz[a,h]anthracene	0.38	0.31	0.18	0.44	0.15	0.51	0.18
Benz[g,h,i]perylene	20.97***	0.24 ^c	0.00	1.00 ^b	0.16	1.54 ^a	0.19

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Values with same small letters denote no significant difference among the three sites

trations of PAHs in Pol1 and Pol2 were 5.3 and 7.6 times of that in Ref, respectively. Except for anthracene and dibenz[a,h]anthracene, all the PAH compounds had significantly higher concentrations in Pol1 and Pol2 than in Ref, from 2 to 10 times for the different compounds.

2.2 General mite community structure

More than 600 soil mites of Acari were collected representing 54 species, of which 27 belonged to Oribatida, 14 to Mesostigmata, 11 to Prostigmata and 2 to Astigmata suborder, but 32 could not be assigned with certainty to described species (Table 2). Species over 5% of the total abundance were *Scheloribates oryzae* (17.3%), *Tectocephus velatus* (10.5%), *Oppeia nova* (9.8%), *Rostrozetes foveolatus* (8.3%), *Rhodacarus* sp. (6.8%), *Rhizoglyphus callae* (6.7%) and *Epilohmannia pallida pacifica* (6.3%). Two Oribatida species *Epilohmannia pacifica* ($F_{2,9} = 11.43$, $P < 0.01$) and *Hypochthonius rufulus* ($F_{2,9} = 4.61$, $P < 0.05$) were

significantly different among the three study sites, with densities of the two species in Pol2 being much higher than that in Ref and Pol1. Species richness of Oribatida was significantly different among the three study sites ($F_{2,9} = 9.61$, $P < 0.01$); the species richness in Pol2 (8.50 ± 0.87) exceeded that in Ref (5.25 ± 0.63) and Pol1 (4.25 ± 0.63 species number per plot) by 61% and 100%, respectively.

2.3 MGP groups of Oribatida

Similar to *Oribatida*, both density ($F_{2,9} = 15.40$, $P < 0.01$) and species richness ($F_{2,9} = 8.31$, $P < 0.01$) of Macropylina of Oribatida in Pol2 (density 3167 ± 518 , species richness 4.00 ± 0.71) exceeded those for density in Ref (750 ± 323) and Pol1 (333 ± 118 ind./m²) by 322% and 851%, respectively and those for species richness in Ref (2.00 ± 0.41) and Pol1 (1.25 ± 0.25 species number per plot) by 100% and 220%, respectively.

2.4 Life history classes of Mesostigmata

Table 2 Species, group (M: Macropylina, G: Gymnonota, P: Poronata, K: K-selected, r: r-selected) and density (ind.·m⁻², mean \pm se, n = 4) of Oribatida, Mesostigmata, Prostigmata and Astigmata of soil Acarina in the reference (Ref) and the two polluted (Pol1 and Pol2) study sites

No.	Species	Group	Ref _r	Se	Pol1	Se	Pol2	Se	No.	Species	Group	Ref _r	Se	Pol1	Se	Pol2	Se
	Oribatida		3500	1404	3792	2626	9292	1596	28	<i>Euparholaspulus primoris</i>	2K	500	500	42	42	0	0
1	<i>Brachychochthonius elsosneadensis</i>	M	42	42	0	0	250	160	29	<i>Proparholaspulus suzukii</i>	2K	125	80	125	125	83	83
2	<i>Cyrtthermannia parallela</i>	M	0	0	42	42	0	0	30	<i>Rhodacarus</i> sp.	2K	167	118	375	172	1167	844
3	<i>Epilohmannia pallida pacifica</i>	M	42	42	0	0	1542	448	31	<i>Cheiroseius</i> sp.	1r	125	125	0	0	0	0
4	<i>Epilohmannoides</i> sp.	M	542	284	0	0	500	245	32	<i>Gamasellus</i> sp.	1r	458	229	83	83	333	152
5	<i>Haplochthonius</i> sp.	M	0	0	0	0	42	42	33	<i>Gamasellus tianmuensis</i>	1r	250	160	250	198	0	0
6	<i>Heminothrus yamasakii</i>	M	0	0	167	167	0	0	34	<i>Hypoaspis queerlandicus</i>	1r	292	197	250	160	208	158
7	<i>Hypochthonius rufulus</i>	M	0	0	42	42	583	259	35	<i>Asca</i> sp.	1r	42	42	0	0	0	0
8	<i>Lohmannia</i> sp.	M	42	42	0	0	0	0	36	<i>Cheiroseius nepalensis</i>	1r	83	83	0	0	0	0
9	<i>Malaconothrus</i> sp1.	M	0	0	0	0	208	208	37	<i>Gamasiphis pulchellus</i>	1r	42	42	42	42	83	83
10	<i>Malaconothrus</i> sp2.	M	0	0	83	48	0	0	38	<i>Gamasiphis</i> sp.	1r	0	0	0	0	42	42
11	<i>Palaeacarus</i> sp.	M	0	0	0	0	42	42	39	<i>Digamasellus perpusillus</i>	2r	42	42	0	0	0	0
12	<i>Pseudocryptacarus</i> sp.	M	42	42	0	0	0	0	40	<i>Evimirus</i> sp 1.	4r	42	42	0	0	42	42
13	<i>Sellnicknochthonius</i> sp.	M	42	42	0	0	0	0	41	<i>Evimirus</i> sp 2	4r	0	0	42	42	83	48
14	<i>Eremobelba japonica</i>	G	0	0	0	0	83	48		Prostigmata		250	48	250	108	625	343
15	<i>Eremobodes</i> sp.	G	42	42	0	0	0	0	42	Alicorhagiidae		42	42	0	0	42	42
16	<i>Karenella</i> sp.	G	42	42	0	0	0	0	43	<i>Bdella</i> sp.		0	0	0	0	42	42
17	<i>Oppeia nova</i>	G	0	0	333	226	2125	1375	44	Calyptostomidae sp.		42	42	0	0	0	0
18	<i>Tectocephus velatus</i>	G	167	118	1125	1017	1333	649	45	<i>Eustigmacus</i> sp.		0	0	0	0	42	42
19	<i>Ceratozetella</i> sp.	P	0	0	42	42	0	0	46	<i>Mahunlania secunda</i>		42	42	42	42	0	0
20	<i>Ceratozetes</i> sp.	P	42	42	0	0	0	0	47	<i>Pulaeus</i> sp.		0	0	0	0	42	42
21	<i>Fissurobates</i> sp.	P	0	0	42	42	0	0	48	<i>Robustochelates</i> sp.		0	0	0	0	83	83
22	<i>Lamellobates</i> sp.	P	167	167	0	0	0	0	49	<i>Scirula</i> sp.		0	0	42	42	0	0
23	<i>Pergalumna</i> sp.	P	42	42	0	0	0	0	50	<i>Scutacarus</i> sp.		0	0	125	125	0	0
24	<i>Rostrozetes foveolatus</i>	P	208	208	292	172	1583	1148	51	<i>Spinibdella</i> sp.		42	42	0	0	125	80
25	<i>Scheloribates latipes</i>	P	0	0	292	292	0	0	52	<i>Tarsonemus granarus</i>		83	83	42	42	250	250
26	<i>Scheloribates oryzae</i>	P	2000	1502	1333	1074	1000	514		Astigmata		1542	1487	125	80	250	250
27	<i>Sphaerozetes</i> sp.	P	42	42	0	0	0	0	53	<i>Rhizoglyphus callae</i>		1500	1445	83	83	83	83
	Mesostigmata		2167	785	1208	229	2042	1163	54	<i>Tyrophagus putrescentiae</i>		42	42	42	42	167	167

Majority of Mesostigmata species were classified as 2K- (3 species) and 1r- (8 species) selected species, 2r and 4r were only 1 and 2 species, respectively, while 1K and 3K species were not found (Table 2). 1r-species number in Ref (7 species) was almost twice those in the two polluted sites (4 species in both sites). 2r-species were only found in Ref. By repeated ANOVA, density of 1r-species (using the 8 species as multiple factors) was significantly different among the three study sites ($F_{12,8} = 6.22$, $P < 0.01$); the density in Ref exceeded that in Pol1 and Pol2 by 107% and 94%, respectively. Values of the maturity index were variable among Ref (0.30), Pol1 (0.43) and Pol2 (0.25).

2.5 Sex ratio of Mesostigmata

7 species of Mesostigmata whose male and female individuals were found simultaneously at least in one study site were used for the analysis (Table 3). Of the 7 species, males of 5 species were found in Ref but males of only 2 and 3 species in Pol1 and Pol2, respectively. Similar to the 1r-species, by repeated ANOVA, male density ($F_{14,6} = 4.66$, $P < 0.05$) and male to female sex ratio ($F_{14,6} = 8.27$, $P < 0.01$) (both using the 7 species as multiple factors) were significantly different among the three study sites; the density of single species on average in Ref exceeded that in Pol1 and Pol2 by 221% and 328%, respectively and the relevant values for the sex ratio were 97% and 255%.

2.6 Correlation between mite community parameters and soil environmental factors

Correlations of the 6 mite community structure parameters (Oribatida species richness, Macropylina density, Macropylina species richness, the 1r-species density, the male density and the sex ratio) and the 7 soil environmental factors (2-0.02 and 0.02-0.002 mm

content of soil texture, organic carbon, total nitrogen, Cu, Pb and total PAHs) were analyzed. These mite parameters and the environmental factors were chosen because they were significantly different among the study sites. Multiple regression analysis was applied and the 7 environmental factors were used as multiple variables. Oribatida species richness, Macropylina density and Macropylina species richness were significantly positively correlated with Cu concentration (Table 4), that is, they generally increased with Cu increasing in range of the occurred concentration (ca. 30-55 Cu mg kg⁻¹ dw soil). The coefficient (R) of Macropylina density with Cu was highest among the 3 parameters and the effect of Cu explained (λA) accounted for majority of the total effect of all the 7 environmental factors together explained (0.92). Oppositely, the 1r-species density, male density and sex ratio were significantly negatively correlated with the concentration of total PAHs (Table 4). Among the 3 parameters, the male density had the highest negative coefficient and the λA value, while 1r-species density had the second highest negative coefficient but its λA was lower than that of the sex ratio.

3 Discussion

3.1 Soil environmental factors

The general soil properties of pH, electrical conductivity, cation exchange capacity, water content and water holding capacity were not significantly different between the reference and polluted study sites, suggesting that the difference of the mite community structure among the study sites were not caused by these properties. Soil texture (2-0.02 and 0.02-0.002 mm) and soil nutrients (organic C and total N) were significantly different between the study sites, but the differences were not very strong and were not corre-

Table 3 Density (ind. m⁻², mean \pm se, $n = 4$) of males (M) and male to female ratio (M/F) of the 7 species of Mesostigmata and the average analyzed by repeated ANOVA (using the 7 species as multiple factors) among the reference (Ref) and the two polluted (Pol1 and Pol2) study sites

Species	Ref				Pol1				Pol2			
	M	Se	M/F	Se	M	Se	M/F	Se	M	Se	M/F	Se
<i>Euparholaspulus primoris</i>	125	125	0.5	0.5	0	-	0	-	0	-	-	-
<i>Proparholaspulus suzukii</i>	0	-	0	-	42	42	1.0	1.0	42	42	1.0	1.0
<i>Rhodacarus</i> sp.	0	-	0	-	0	-	0	-	42	42	0.04	0.04
<i>Cheiroseius</i> sp.	42	42	0.5	0.5	0	-	-	-	0	-	-	-
<i>Gamasellus</i> sp.	208	158	2.5	2.5	0	-	0	-	42	42	0.33	0.33
<i>Gamasellus tianmuensis</i>	83	83	0.5	0.5	125	80	1.5	1.5	0	-	-	-
<i>Hypoaspis queerlandicus</i>	83	83	1.0	1.0	0	-	0	-	0	-	0	-
Average	77 ^a	74	0.71 ^a	0.71	24 ^{ab}	17	0.36 ^{ab}	0.36	18 ^b	18	0.20 ^b	0.20

Values with different small letters denote significant difference among the study sites. Statistical results refer to the text

Table 4 Coefficients (*R*) of correlation between mite community parameters and soil environmental factors by Multiple regression analysis and effect of each factor explained of total effect (λA) and *F* values by Monte Carlo test (permutation number 499)

	Oribatida species richness			Macropylina density			Macropylina species richness			1r-species density			Male-dependent density			Male to female sex ratio		
	<i>R</i>	λA	<i>F</i>	<i>R</i>	λA	<i>F</i>	<i>R</i>	λA	<i>F</i>	<i>R</i>	λA	<i>F</i>	<i>R</i>	λA	<i>F</i>	<i>R</i>	λA	<i>F</i>
Texture																		
2-0.02 mm	-0.31	0.21	4.48	-0.48	0	0.14	-0.27	0.01	0.23	0.56	0.07	1.05	0.58	0.02	0.53	0.02	0	0.04
0.02-0.002 mm	-0.06	0.13	3.66	0.17	0.05	3.03	-0.03	0	0.05	-0.49	-	-	-0.48	0.02	0.62	0.03	0.03	0.57
Nutrient																		
Organic carbon	-0.3	0.04	0.68	-0.62	0.01	0.77	-0.38	0.08	2.37	0.46	0.01	0.21	0.28	-	-	-0.02	0.13	2.91
Total nitrogen	-0.46	0.01	0.32	-0.77	0.05	2.29	-0.64	0.02	0.67	0.21	0.08	1.30	0.17	0.02	0.63	0.06	0.03	0.58
Heavy metal																		
Cu	0.61	0.37*	5.76	0.86	0.77**	33.47	0.76	0.62**	16.15	-0.24	-	-	0.02	0.06	1.48	-0.09	0.05	1.13
Pb	0.23	0.05	1.12	0.41	0.02	0.98	0.35	0.01	0.3	0.19	0.14	2.62	0.56	0.08	2.5	0.39	0	0.02
PAH																		
Total PAHs	0.51	0.02	0.29	0.63	0.02	1.09	0.49	0.06	1.92	-0.70	0.39**	6.43	-0.76	0.63**	16.79	-0.51	0.47*	8.9

P* < 0.05, *P* < 0.01. - variables not to improve fit

lated with the mite community structure, suggesting that these soil parameters could be factors causing the difference of the mite community structure but might not be main factors.

For heavy metals in the soils, Cu, Pb and Zn were detected. Cu and Pb were significantly different between the study sites and Cu significantly (but positively) correlated with the mite community structure, suggesting that heavy metals, especially Cu, could be factors associated with the mite community structure. However, the concentrations of the metals were low, Cu and Pb concentrations being just above and Zn below the criterion of the lightest pollution category in the Chinese National Standard, and therefore the concentrations would not be high enough to cause negative effects on the soil mite community structure.

The total PAH concentrations were in the range of 2.93 to 33.17 mg/kg, which are comparable to the concentrations of 5.28 to 80.64 mg/kg in another study [22]. PAHs can be produced from incomplete combustion, which are highly lipophilic semi-volatile organic compounds partitioning between the atmospheric gas-phase and particle-phase [3] and the PAHs entering the airshed can be deposited directly onto soils or first absorbed by vegetation and then incorporated into the soil when vegetation falls to the ground at the end of the growing season and decays [4]. The concentrations of PAHs in the polluted sites were much higher than those in the reference site and negatively correlated with the mite community structure parameters related to Mesostigmata, suggesting that

PAHs could be the main factor causing the differences of the mite community structure parameters, directly or indirectly [36], in the present study.

Therefore, in the present study, it seems that joint effects of PAHs, heavy metals, soil nutrients and soil texture caused the differences of the mite community structure parameters between the reference and polluted study sites, with the PAHs being the main factor.

3.2 General mite community structure and parameters of Oribatida

Densities of all species, Shannon diversity and evenness indices of total mites (Acari) and the 4 sub-orders (Oribatida, Prostigmata, Astigmata and Mesostigmata) were not significantly different but species richness of Oribatida was significantly different between the reference and the polluted sites, suggesting that the species richness is more sensitive than the density, Shannon diversity and evenness indices in the present study [37-38].

Density and species richness of Macropylina were also significantly different among the three study sites. The Oribatida species richness being consistent with Macropylina species richness may be due to Macropylina accounting for majority (almost half) species of Oribatida. In Pol2, Cu concentration was highest but organic C and total N were lowest among the three study sites and concentrations of PAHs were much higher than those in the reference site, which may simultaneously cause the increase of Oribatida. Microcosm studies on another organic pollutant, PCP, showed that different body form of oribatids had dif-

ferent sensitivities. Numbers of small soft-bodied mite species increased in the higher PCP concentration treatment, while no change in numbers of large and more sclerotized Oribatida mites were found^[18,39]. They explained the increase in these mites in the higher PCP concentration by the efficient utilization of the high bacterial production and/or lowered resource competition from other microbivores. This is also consistent with the prediction of^[40] and^[41] that small body size and short life-cycle were favoured in stressed soil. In the present study, most small soft-bodied species of Oribatida were in Macropylina and Majority of species of Macropylina were small soft-bodied. Of 13 species of Macropylina, 8 species (*Brachychochthonius elsosneadensis*, *Epilohmannia pacifica*, *Malaconothrus* sp.1, *Epilohmannoides* sp., *Haplochthonius* sp., *Malaconothrus* sp.2, *Palaeacaroides* sp. and *Sellnicknochthonius* sp.) were the type of small (< 0.4 mm) and soft and 5 species (*Hypochthonius rufulus*, *Cythermannia parallela*, *Heminothrus yamasakii*, *Lohmannia* sp. and *Pseudocryptacarus* sp.) were large (> 0.4 mm) and sclerotic. Differently, all 5 species of Gymnionota were sclerotic and all 10 species of Poronata were large and sclerotic. That the Oribatida parameters were highest in Pol2 and positively correlated with the Cu concentrations (34.3-53.7 mg/kg⁻¹) in the present study might be also due to the stimulation of Cu in low concentration to the mites^[42]. Skubala and Zaleski (2012) concluded that small concentrations of heavy metals are positively correlated with the development of saprophagous oribatid mite communities, since the stimulation of low concentration of heavy metals on the rate of reproduction is recognized and is called hormesis^[21].

3.3 Life history classes and sex ratio of Mesostigmata

All species of Mesostigmata are gamasides, i.e. predatory mites in the present study. Density and species richness of 1 r -selected species, density and species richness of male-dependant species and male to female sex ratio of Mesostigmata were significantly higher in the reference site than in the two polluted sites, and the 1 r -species density, male density and sex ratio were negatively correlated with the concentrations of PAHs, suggesting that 1 r -species and male individuals and their related parameters could be sensitive to the PAHs. There are two possible explanations

for the effects of PAHs on mite community: Firstly, PAHs are fungicides that contaminate and eliminate the fungal and detrital food of microarthropods, and secondly, increased bulk density from PAH contamination reduces habitat space for microarthropods^[9]. The results of higher fine and lower coarse content of soil texture in the PAH-contaminated soils in the present study support the second explanation. Studies on PCP showed that total numbers of predatory mites were extremely low at high PCP concentrations^[43] and studies on land use showed that there were marked differences in Gamasina fauna among four different land use types and indicated that Gamasina community structure changed significantly with material and energy inputs and mechanical perturbations^[16], which are consistent with the present study, although the perturbation factors and mite community parameters are different. So, the results in the present study support the idea that predatory soil mite fauna seem to be a good indicator when their life history traits are taken into account and can be considered as one of the most sensitive groups in a hierarchical multi-taxon classification system for the ecological quality of soils^[14-16, 44]. By the way, although the parameters in the whole Mesostigmata community were significantly different between the reference and the polluted sites, those of each species were not. As suggested by^[45], specific assemblages but not the same species dominating the community were associated with the specific environmental conditions.

In conclusion, results of the present study show that PAH-contaminated soils in the field affect soil mite community structure, but different parameters of the mite community structure have differing sensitivity. Species richness of Oribatida and density and species richness of Macropylina could be sensitive to the PAH-contaminated soils, but might not be directly influenced by PAHs in the current concentrations, instead, by other soil environmental factors mediated by PAHs such as soil texture, soil nutrients and heavy metals. For Mesostigmata parameters, density of 1 r -species, male density and male to female sex ratio seem more sensitive to the PAH-contaminated soils and may directly relate to PAHs as well as the PAH-mediated effects of the other soil environmental factors. Therefore, these parameters may be sensitive to PAH-contaminated soil and can be further devel-

oped to be indicators for soil environmental quality. Because this is a field study, which may need laboratory or microcosm studies to prove the results later, we cautiously draw the conclusions.

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华东多环芳烃污染农田土壤螨群落结构研究

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摘要: 土壤螨目是土壤质量评价的较好指示生物, 但还很少用于多环芳烃(PAHs)污染的研究。已有的研究主要采用群落组成参数作为指标特征, 且大多在类群水平上, 如, 密度、类群多度、多样性指数和均匀性指数等。由于这些指标是建立在较粗的分类基础上, 因此, 评价结果通常变异性较大, 不够稳定。本研究是在物种水平上, 除对群落组成参数外, 还着重对生活史和雌雄性比例等生物学特征进行了研究, 以筛选更为敏感的指标特征。研究采用污染田与对照田对比的方法, 于2008年对华东区一蔬菜田的PAHs污染及其对土壤螨群落结构的影响进行了调查研究。除土壤螨群落结构参数和16种优先监控PAH化合物外, 还对土壤理化和重金属进行了测定。PAHs在污染田中的总浓度为18.7 to 26.8 mg kg⁻¹ dw soil, 比对照田高5.3到7.6倍。污染田甲螨亚目低等类的密度和种丰富度比对照田分别高3.2到8.5和1.0到2.2倍。相反的, 中气门亚目的1r策略选择种密度、雄性个体密度和雌/雄性比例在对照田比在污染田分别高0.9到1.1、2.2到3.3和1.0到2.6倍, 且与总PAH浓度呈显著负相关。这一研究结果表明: 甲螨亚目低等类和中气门亚目的1r种、雄性个体以及雌/雄性比例等相关特征对土壤PAH污染敏感, 因而, 可进一步筛选作为土壤污染的评价指标。

关键词: 螨亚纲; 土壤污染; 生物指标; 物种多样性; 雌雄性比; 生活史