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CONTENTS

Aquatic environment

Three-dimensional hydrodynamic and water quality model for TMDL development of Lake Fuxian, China
Lei Zhao, Xiaoling Zhang, Yong Liu, Bin He, Xiang Zhu, Rui Zou, Yuanguan Zhu 1355
Removal of dispersant-stabilized carbon nanotubes by regular coagulants
Ni Liu, Changli Liu, Jing Zhang, Daohui Lin 1364
Effect of environmental factors on the effectiveness of ammoniated bagasse in wicking oil from contaminated wetlands
Seungjoon Chung, Makram T. Suidan, Albert D. Venosa 1371
Cationic content effects of biodegradable amphoteric chitosan-based flocculants on the flocculation properties
Zhen Yang, Yabo Shang, Xin Huang, Yichun Chen, Yaobo Lu, Aimin Chen, Yuxiang Jiang, Wei Gu, Xiaozhi Qian, Hu Yang, Rongshi Cheng 1378
Biosorption of copper and zinc by immobilised and free algal biomass, and the effects of metals biosorption on the growth and cellular structure of *Chlorella* sp. and *Chlamydomonas* sp. isolated from rivers in Penang, Malaysia
W. O. Wan Maznah, A.T. Al-Fawwaz, Misni Surif 1386
Variation of cyanobacteria with different environmental conditions in Nansi Lake, China
Chang Tian, Haiyan Pei, Wenrong Hu, Jun Xie 1394
Enhancing sewage sludge dewaterability by bioleaching approach with comparison to other physical and chemical conditioning methods
Fenwu Liu, Jun Zhou, Dianzhan Wang, Lixiang Zhou 1403
Effect of chlorine content of chlorophenols on their adsorption by mesoporous SBA-15
Qingdong Qin, Ke Liu, Dafang Fu, Haiying Gao 1411
Surface clogging process modeling of suspended solids during urban stormwater aquifer recharge
Zijia Wang, Xinqiang Du, Yuesuo Yang, Xueyan Ye 1418
Adsorptive removal of iron and manganese ions from aqueous solutions with microporous chitosan/polyethylene glycol blend membrane
Neama A. Reiad, Omar E. Abdel Salam, Ehab F. Abadir, Farid A. Harraz 1425
Polyphenylene sulfide based anion exchange fiber: Synthesis, characterization and adsorption of Cr(VI)
Jiajia Huang, Xin Zhang, Lingling Bai, Siguo Yuan 1433

Atmospheric environment

Removal characteristics and kinetic analysis of an aerobic vapor-phase bioreactor for hydrophobic alpha-pinene
Yifeng Jiang, Shanshan Li, Zhuowei Cheng, Runye Zhu, Jianmeng Chen 1439
Characterization of polycyclic aromatic hydrocarbon emissions from diesel engine retrofitted with selective catalytic reduction and continuously regenerating trap
Asad Naeem Shah, Yunshan Ge, Jianwei Tan, Zhihua Liu, Chao He, Tao Zeng 1449
Size distributions of aerosol and water-soluble ions in Nanjing during a crop residual burning event
Honglei Wang, Bin Zhu, Lijuan Shen, Hanqing Kang 1457
Aerosol structure and vertical distribution in a multi-source dust region
Jie Zhang, Qiang Zhang, Congguo Tang, Yongxiang Han 1466

Terrestrial environment

Effect of organic wastes on the plant-microbe remediation for removal of aged PAHs in soils
Jing Zhang, Xiangui Lin, Weiwei Liu, Yiming Wang, Jun Zeng, Hong Chen 1476
Nitrogen deposition alters soil chemical properties and bacterial communities in the Inner Mongolia grassland
Ximei Zhang, Xingguo Han 1483

Environmental biology

Augmentation of tribenuron methyl removal from polluted soil with *Bacillus* sp. strain BS2 and indigenous earthworms
Qiang Tang, Zhiping Zhao, Yajun Liu, Nanxi Wang, Baojun Wang, Yanan Wang, Ningyi Zhou, Shuangjiang Liu 1492
Microbial community changes in aquifer sediment microcosm for anaerobic anthracene biodegradation under methanogenic condition
Rui Wan, Shuying Zhang, Shuguang Xie 1498

Environmental health and toxicology

Molecular toxicity of earthworms induced by cadmium contaminated soil and biomarkers screening
Xiaohui Mo, Yuhui Qiao, Zhenjun Sun, Xiaofei Sun, Yang Li 1504
Effect of cadmium on photosynthetic pigments, lipid peroxidation, antioxidants, and artemisinin in hydroponically grown *Artemisia annua*
Xuan Li, Manxi Zhao, Lanping Guo, Luqi Huang 1511

Environmental catalysis and materials

Influences of pH value in deposition-precipitation synthesis process on Pt-doped TiO_2 catalysts for photocatalytic oxidation of NO
Shuzhen Song, Zhongyi Sheng, Yue Liu, Haiqiang Wang, Zhongbiao Wu 1519
Adsorption of mixed cationic-nonionic surfactant and its effect on bentonite structure
Yaxin Zhang, Yan Zhao, Yong Zhu, Huayong Wu, Hongtao Wang, Wenjing Lu 1533

Municipal solid waste and green chemistry

Recovery of phosphorus as struvite from sewage sludge ash
Huacheng Xu, Pinjing He, Weimei Gu, Guanzhao Wang, Liming Shao 1525

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Effect of organic wastes on the plant-microbe remediation for removal of aged PAHs in soils

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Abstract

The effectiveness of *in-situ* bioremediation of polycyclic aromatic hydrocarbons (PAHs) may be inhibited by low nutrients and organic carbon. To evaluate the effect of organic wastes on the PAHs removal efficiency of a plant-microbe remediation system, contaminated agricultural soils were amended with different dosages of sewage sludge (SS) and cattle manure (CM) in the presence of alfalfa (*Medicago sativa* L.) and PAHs-degraders (*Bacillus* sp. and *Flavobacterium* sp.). The results indicated that the alfalfa mean biomasses varied from 0.56 to 2.23 g/pot in root dry weight and from 1.80 to 4.88 g/pot in shoot dry weight. Low dose amendments, with rates of SS at 0.1% and CM at 1%, had prominent effects on plant growth and soil PAHs degradation. After 60-day incubation, compared with about 5.6% in the control, 25.8% PAHs removal was observed for treatments in the presence of alfalfa and PAHs-degraders; furthermore, when amended with different dosages of SS and CM, the removed PAHs from soils increased by 35.5%–44.9% and 25.5%–42.3%, respectively. In particular, the degradation of high-molecular-weight PAHs was up to 42.4%. Dehydrogenase activities (DH) ranged between 0.41 and 1.83 μg triphenylformazan/(g dry soil·hr) and the numbers of PAHs-degrading microbes (PDM) ranged from 1.14×10^6 to 16.6×10^6 most-probable-number/g dry soil. Further investigation of the underlying microbial mechanism revealed that both DH and PDM were stimulated by the addition of organic wastes and significantly correlated with the removal ratio of PAHs. In conclusion, the effect of organic waste application on soil PAHs removal to a great extent is dependent on the interactional effect of nutrients and dissolved organic matter in organic waste and soil microorganisms.

Key words: polycyclic aromatic hydrocarbons (PAHs); soil contamination; bioremediation; organic wastes
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Introduction

Polycyclic aromatic hydrocarbons (PAHs) have been recognized as a heterogeneous group of persistent contaminants, due to their toxic, carcinogenic and mutagenic properties and high recalcitrance to different types of degradation (Ockenden et al., 2003; Mohan et al., 2006). They are widely distributed in environments such as soil, sediment, water and air as results of both natural and anthropogenic processes, and therefore have known harmful effects on humans and wildlife (Keith, 1979; Eibes et al., 2006).

Bioremediation is a low-cost and environmentally-friendly alternative for decontamination of PAHs-contaminated soils. Recently, numerous studies investigating the application of bioremediation to treat PAHs-contaminated soils using a variety of plant (Liste and Alexander, 2000; Fismes et al., 2002; Fan et al., 2008) or microbial species (Gomes et al., 2005; Wu et al., 2008) or plant-microbe combinations (Tam and Wong, 2008) have been carried out. However, the effectiveness of *in-situ* bioremediation of aged PAHs-contaminated soils is always limited, because the residual components of PAHs in aged soil have poorer water solubility and are more strongly adsorbed by soil particles, which leads to a lower biodegradation effectiveness compared with fresh PAHs-contaminated soils (Leonardi et al., 2007; Hwang and Cutright, 2002). Furthermore, PAHs-contaminated soils are often nutrient- and organic matter-deficient so that a low PAHs degradation capability is exhibited due to the low number and activity of indigenous microbial population (Kobayashi et al., 2008).

Organic wastes, such as animal manure, straw, compost and sewage sludge, have often been used to improve soil quality, by altering the physical properties (soil aggregation), ameliorating soil organic matter (SOM) quantity, increasing nutrient availability, and other soil functions (Debosz et al., 2002; Celik et al., 2004; Courtney and Mullen, 2008). Based on the introduction of microorgan-

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isms capable of degrading high molecular weight PAHs, organic wastes have been considered to be an effective solution to stimulate the indigenous and exogenous microbial population and enhance their PAHs metabolic capability. On one hand, the organic matter plays an important role in PAHs biodegradation through contribution of soil nutrients as a result of their mineralization and by stimulating microbial activity (Manilal and Alexander, 1991; Kästner and Mahro, 1996; Cheng et al., 2008). On the other hand, organic wastes always are rich in dissolved organic matter (DOM), and the presence of DOM in the aqueous phase could decrease the sorption of PAHs, and thus influence their mobility (Yu et al., 2011). Moreover, the influence of exotic DOM on PAHs sorption was related to DOM concentrations (Gao et al., 2007). Several studies have focused on improved organic pollutant (PAHs, PCB or PCP) biodegradation by waste organic material amendments in artificially contaminated soil (Álvarez-Bernal et al., 2006; Contreras-Ramos et al., 2008; Fernández-Luqueno et al., 2008; Simeon et al., 2008; Kobayashi et al., 2008; Tejada and Masciandaro, 2011). However, no details about the effect of organic wastes on the removal of aged PAHs from naturally contaminated soils in plant-microbe systems has been published so far.

The objective of this study was to evaluate the influence of cattle manure and sewage sludge on bioremediation of aged PAHs soil in a plant-microbe system. It was postulated that organic waste may enhance the biodegradation of PAHs in soil by stimulating and increasing the activity and number of exogenous and indigenous PAHs-degrading microorganisms and finally improving PAHs bioavailability.

1 Materials and methods

1.1 Characteristics of soil and organic wastes

The soil used in this experiment was collected from a field located in Wuxi, Jiangsu Province, eastern China (30°36′14″N, 120°28′33″E). The main soil characteristics are shown in Table 1. The organic wastes applied were sewage sludge and cattle manure, respectively. Sewage sludge was the dry powder of ammonium sulphite pulping wastewater from a papermaking plant and cattle manure was the fermentation product of cow dung from a cattle farm. General properties of the organic wastes are also listed in Table 1.

Table 1 Characteristics of experimental soil and organic waste

	Soil	Sewage sludge	Cattle manure
pH	6.4	5.74	6.89
Organic matter (g/kg)	19.2	83.5	169.3
Total N (g/kg)	1.0	29.64	15.10
Total P (g/kg)	0.5	0.05	3.71
Total K (g/kg)	14.2	8.73	4.92
CEC (cmol/kg)	21.5	ND	ND
Zn (mg/kg)	ND	297	126
Cu (mg/kg)	ND	278	231
Cd (mg/kg)	ND	2.56	1.25
Pb (mg/kg)	ND	80.1	29

ND: not detected.

1.2 Experimental design

The soil was carefully collected, homogenized, air-dried and passed through a 2 mm sieve. Nine treatments were designed with four replicates of each treatment as shown in Table 2.

Alfalfa (*Medicago sativa* L.) was selected for the experiment because its extensive, widely branched root system provides a large root surface for the growth of microbial populations. The PAHs-degrading bacterial inoculums, which mainly contained two strains, *Flavobacterium* sp. and *Bacillus* sp., were cultivated in tryptic soy broth (Burd et al., 2000) at 37°C for 2 days. About 10^7 cells were inoculated to one gram of soil on a dry basis. The doses of cattle manure and sewage sludge amended were 0.5%, 1%, 2% and 0.05%, 0.1%, 0.2% (m/m), respectively.

The pots (12 cm in diameter, 18 cm in height) with 3 kg dry soil each were placed into a greenhouse. Alfalfa seeds were sterilized in 10% (V/V) H_2O_2 for 20 min and washed three times with distilled water. Fifteen seeds were sown in four replicates and the seedlings were thinned to 10 seedlings after germination. The growth conditions were: 25°C during a 12 hr day and at 20°C during a 12 hr night. The light intensity was 4500–7300 lux and the soils were watered daily and adjusted to approximately 50% of the water holding capacity during plant growth.

1.3 Sampling and analysis

The soils were collected by soil auger from the surface (0–20 cm) in the vicinity of the root 60 days after germination. Soils were manually crushed and homogenized, then passed through a sieve (2 mm). The subsamples were stored at 4°C for assessing soil enzymes and enumeration of soil microorganisms. Shoots and roots were harvested, respectively. Root fragments were collected by sieving the soil and adding them to the root samples. Roots were carefully washed with tap water to remove any adhering soil particles. Then shoots and roots were freeze-dried and weighed. Soil dehydrogenase activity (DH) was determined by the reduction of triphenyltetrazolium chloride (TTC) to triphenylformazan (TPF) as described by Tabatabai (1994). PAHs degraders were enumerated using the most-probable-number (MPN) method with five replicates per dilution (Wrenn and Venosa, 1996). A mixture of phenanthrene (10 g/L), anthracene (1 g/L), fluorene (1 g/L) and fluoranthene (1 g/L) was supplied as the sole

Table 2 Treatments of experimental design

Treatment	Plant	PAHs-degrading bacteria	SS	CM
CK1	–	–	–	–
CK2	+	–	–	–
CK3	+	+	–	–
SS1	+	+	0.05%	–
SS2	+	+	0.1%	–
SS3	+	+	0.2%	–
CM1	+	+	–	0.5%
CM2	+	+	–	1%
CM3	+	+	–	2%

CK: control; SS: sewage sludge; CM: cattle manure; +: with corresponding treatments; –: without corresponding treatments.

carbon source to a mineral medium (Wrenn and Venosa, 1996). Serially diluted soil solutions, ranging from 10^{-3} to 10^{-6}, were performed, inoculated into the medium and incubated at 28°C in the darkness. After 3 weeks, samples of the medium that turned yellow or brown were treated as positive.

Another subunit of soil samples was stored at –20°C for PAHs analysis. Five gram freeze-dried samples were extracted with 60 mL dichloromethane in a Soxhlet apparatus for 24 hr. Extracts were then concentrated using a rotary evaporator and purified with a chromatography column filled with activated silica gel. Purified extracts (10 μL) were analyzed using HPLC (Waters, USA), which was fitted with a special PAHs column (particle size 5 μm, C18 covered, 250 mm×4.6 mm ID) (Waters, USA) and a guard column packed with the same material (Waters, USA). A mobile phase acetonitrile/water gradient was used. The gradient started at the ratio of acetonitrile/water 6:4 (*V*/*V*) at 0–12 min and 1:0 (*V*/*V*) at 12–25 min, then 6:4 (*V*/*V*) at 25–45 min. Separation was performed at 30°C and flow rate of 1.0 mL/min. A fluorescence detector with variable wavelengths (Waters 2475, USA) was used for PAHs analysis. The excitation/emission wavelengths were 215–330 nm from 0.0 to 8.5 min, 290–335 from 8.5 to 11.4 min, 240–375 nm from 11.4 to 16.7 min, and 235–420 nm from 16.7 to 22.3 min. Individual PAHs were identified by their retention time according to PAHs standards.

1.4 Statistical analysis

Analysis of variance (ANOVA) was performed with SPSS version 13.0 software. Comparisons of means were made by calculation of least significant difference (LSD) at the 5% level.

2 Results

2.1 Plant biomass

The alfalfa mean biomasses in different treatments varied from 0.56 to 2.23 g/pot in root dry weight and from 1.80 to 4.88 g/pot in shoot dry weight (Table 3). When PAHs-degrading bacteria were inoculated into the soil, there was a large difference in whole plant biomasses systematically observed between inoculated and uninoculated soils; for example, root and shoot dry weight increased significantly by 297% and 136% ($p < 0.05$). Against our expectation, the biomass of alfalfa decreased to a greater extent with increasing doses of sewage sludge (SS) and cattle manure (CM) compared with that of CK3. However, there was no significant difference between SS1 and SS3 in plant biomass, with the exception of SS1 in root biomass. In contrast, the dosage effect of CM was more apparent; the shoot was stimulated by low and medium dose (CM1 and CM2) and significantly inhibited by high dose (CM3). The total biomass in the treatment with low dose CM, CM1 accumulated to 6.67 g/pot, which was 2.8 times higher than that of CK2, but decreased sharply to 3.79 g/pot at the 2% addition level.

Table 3 Biomass of alfalfa grown in different treatments

Treatment	Root (g/pot)	Shoot (g/pot)	Total (g/pot)	Root/Shoot
CK2	0.56 a	1.80 a	2.37 a	0.31 a
CK3	2.23 c	4.25 c	6.48 d	0.52 d
SS1	2.03 bc	4.14 c	6.16 cd	0.49 d
SS2	1.77 b	4.19 c	5.96 c	0.42 bc
SS3	1.62 b	4.02 c	5.64 c	0.40 bc
CM1	1.79 b	4.88 d	6.67 d	0.37 b
CM2	1.77 b	4.57 cd	6.34 cd	0.39 bc
CM3	0.92 a	2.87 b	3.79 b	0.32 a

Different letters following means in the column indicate significant differences by Fisher's LSD ($p < 0.05$).

2.2 Soil enzyme activities and soil microorganisms

Soil dehydrogenase activities (DH) ranged between 0.41 and 1.83 μg TPF/(g dry soil·hr) in all the treatments (Fig. 1). In the case of CK3, a higher value was observed due to the inoculation of PAHs-degrading bacteria. At the same time, DH activities were significantly stimulated by organic waste addition in the plant-microbe system. However, with increasing amounts of amendments, changing patterns were observed between SS and CM treatments; DH activities were relative stable in treatments with SS, while a decline of DH activity was observed in treatments with CM.

As shown in Fig. 2, the number of PAHs-degrading microorganisms in soil ranged from 1.14×10^6 to 16.6×10^6 MPN/g dry soil, respectively. As we can see, the general trends of changes in the number of microbes were similar to that of DH activity. When inoculated with PAH-degrading microorganisms in the present of alfalfa, the number of PAHs degrading microorganisms in CK3 was about 1.28 times higher than that in CK1. Application of SS and CM significantly increased the number of PAH-degrading microorganisms by about one order of magnitude higher than that of CK3.

2.3 Effects of organic wastes on PAHs degradation

Compared with the background concentration of the soil (8.34 mg/kg, Table 4), after 60 days, the PAHs removal ratio of treatment CK1 was only 5.6%, suggesting that soil indigenous microbes also had the capability of degrading PAHs during the natural dissipation process, though the process was relatively slow. The degradation in phytore-

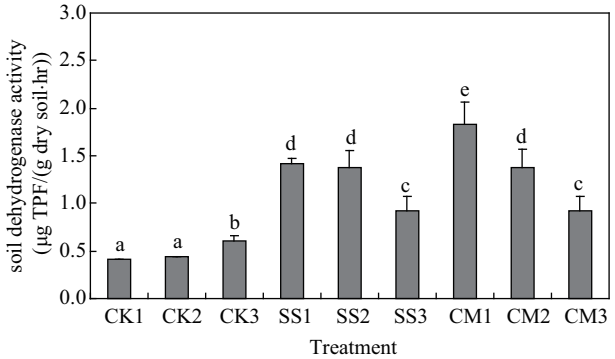

Fig. 1 Soil dehydrogenase activity in different treatments. Different letters above the columns indicate significant difference ($p < 0.05$).

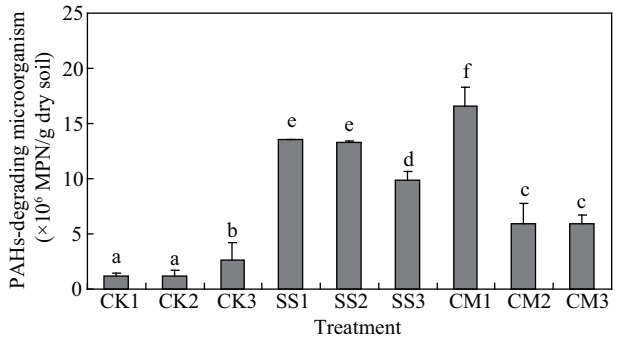

Fig. 2 Soil PAHs-degrading microorganisms in different treatments. Different letters above the columns indicate significant difference ($p < 0.05$).

mediation with alfalfa (CK2) was 11.49%, which was 2 times higher than that of CK1. Furthermore, when inoculated with PAHs-degrading bacteria that were isolated from the culture enriched with high molecular weight PAHs (HMW-PAHs), 25.78% PAHs was removed from the plant-microbe system compared to CK2. Addition of organic material accelerated the removal rate of PAHs with larger percentages observed (32.7%–36.2%). In addition, we found sewage sludge addition had no significant dosage effect on PAHs dissipation. However, the result was a little different with addition of cattle manure, where 42.3% of PAHs was removed by a low dose of CM (CM1), and 26.1% and 25.5% removal ratio were achieved in treatments with medium and high doses of CM (CM2, CM3), respectively.

HMW-PAHs, referring to the PAHs with more than three rings (4, 5 and 6 ring PAHs), dominated with up to 84.27% of the total PAHs, while the low molecular weight PAHs (LMW-PAHs), referring to the PAHs with 2 and 3 rings, only accounted for 15.73%. The removal rate of LMW-PAHs was above 26.89% in the phytoremediation using alfalfa (CK2), and was increased by SS and CM addition at different degrees in the corresponding treatments (Fig. 3). In terms of HMW-PAHs, their dissipation was only up to 11.61% and was little influenced by phytoremediation alone within the 60-day experiment. Furthermore, after innoculation with PAHs-degrading bacteria, the removal rate was 25.6%, which was 120% higher than that of CK2. It is worth mentioning that the addition of waste organic materials caused a great improvement in the dissipation

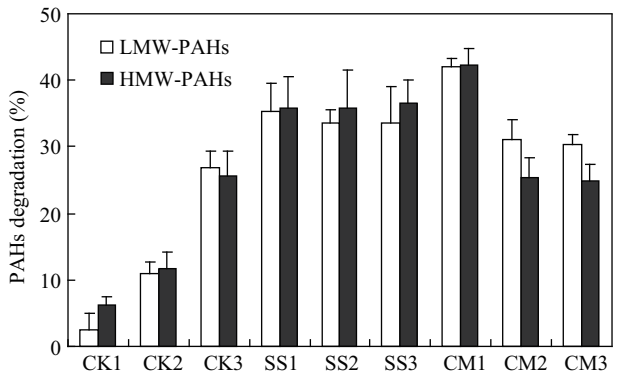

Fig. 3 Degradation of LMW-PAHs and HMW-PAHs in different treatments.

of HMW-PAHs. By contrast with CK3, the PAHs concentration decreased by 40% with addition of SS, although the dose effect was not significant between the treatments. The highest removal ratio of HMW-PAHs was observed in CM1 at 42.4%, which was 2.27 times higher than that of CK1. With increasing dose of CM, the removal ratio decreased to about 40% compared to CM1. The highest degradation of individual PAHs (benzo[a]anthracene, benzo[b]fluoranthene, benzo[k]fluoranthene) dissipated in soil was 55.7%, 50.4% and 54.8% in CM1, respectively.

3 Discussion

3.1 Plant biomass

Application of organic wastes significantly increased total soil organic carbon content and also provided excessive nutrients, such as total N, P and K, which favors plant growth (Debosz et al., 2002; Soumaré et al., 2003; Mbarki et al., 2008). Higher biomass accumulation helped the plants tolerate the toxicity of PAHs in soil, and increased plant uptake and extraction (Gawronski and Gawronska, 2007; Kobayashi et al., 2008). Additionally, root biomass has a close relation with exudates, which contribute to the stimulation of growth and activity of the degrading microbes (Fletcher and Hegde, 1995). However, parallel increases of heavy metals and salt from organic wastes also had a toxic or stress effect on plant growth and yield (Courtney et al., 2008; Cherif et al., 2009). Therefore, the plant biomass accumulation generally depended on the balance of nutrients and toxic effects under the appropriate amendment dose.

3.2 Soil enzyme activities and soil microorganisms

Dehydrogenase has been proposed as a measure of overall microbial activity (Masciandaro et al., 2004), since it is an intracellular enzyme related to oxidative phosphorylation processes (Trevors, 1986). According to Liang et al. (2005), the incorporation of organic amendments to soil stimulates DH activity because the input material may contain intracellular and extracellular enzymes and may also stimulate soil microbial activity. Although a large number of bacterial and fungal strains, such as *Mycobacterium* sp. (Zeng et al., 2010), *Pseudomonas* sp. (Ma et al., 2006), and *Bacillus* sp. (Zhuang et al., 2002) have been reported to be capable of degrading PAHs in soil, microbial degradation can be limited by sub-optimal nutrient levels, temperatures and pH. Organic waste addition improves the nutrients and dissolved organic matter (DOM) in soil, and further has a direct and/or indirect effect on the proliferation of soil microorganisms (Bhattacharyya et al., 2005; Roca-Pérez et al., 2009). DOM from manure compost is considered a rich carbon source for microbial growth as well as for mobilizing soil-bound PAHs. Moreover, it has been confirmed to enhance the biodegradation of PAHs by bacterial cells (Kobayashi et al., 2009; Scelza et al., 2007). However, accumulation of heavy metals and salt from organic wastes may contribute to a possible inhibition of the activity of indigenous and exogenous microbial populations, thereby

Table 4 Changes of 15 PAHs contents in different treatments (unit: ×10 mg/kg)

PAHs	Initial concentration	Residual concentration								
		CK1	CK2	CK3	SS1	SS2	SS3	CM1	CM2	CM3
NAP	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
ACN	0.77 a	0.74 ab	0.68 ab	0.51 bc	0.47 bc	0.47 bc	0.48 bc	0.43 d	0.49 bc	0.49 bc
FLE	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
PHE	0.42 a	0.42 a	0.38 a	0.36 ab	0.30 ab	0.32 ab	0.31 ab	0.26 b	0.33 ab	0.34 ab
ANT	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
FLA	1.30 a	1.29 a	1.18 ab	1.09 bc	0.87 cd	0.89 cd	0.87 cd	0.77 d	1.07 bc	1.08 bc
PYR	1.05 a	1.04 a	0.99 ab	0.89 ab	0. 77 bc	0.79 bc	0.73 bc	0.66 c	0.9 ab	0.89 ab
BAA	0.79 a	0.72 a	0.66 ab	0.54 bc	0.47 c	0.47 c	0.46 c	0.35 c	0.49 bc	0.56 bc
CHR	0.50 a	0.49 a	0.48 a	0.41 abc	0.34 c	0.32 c	0.34 c	0.31 c	0.40 abc	0.43 ab
BBF	1.17 a	1.03 ab	0.95 ab	0.78 bc	0.67 c	0.68 c	0.68 c	0.58 c	0.66 bc	0.69 bc
BKF	0.42 a	0.38 ab	0.36 ab	0.28 bc	0.26 c	0.25 c	0.27 c	0.19 c	0.31 bc	0.29 bc
BAP	0.27 a	0.27 a	0.25 a	0.24 a	0.19 b	0.18 b	0.18 b	0.17 b	0.23 ab	0.24 a
DBP	0.16 a	0.14 ab	0.14 ab	0.11 bc	0.12 bc	0.10 c	0.10 c	0.12 bc	0.12 bc	0.13 abc
IPY	1.25 a	1.13 ab	1.10 ab	0.81 bc	0.75 c	0.77 c	0.76 c	0.86 bc	0.97 bc	0.86 bc
BGP	0.24 a	0.22 a	0.21 a	0.17 b	0.14 b	0.14 b	0.15 b	0.11 b	0.19 ab	0.21 a

NAP: naphthalene; ACN: acenaphthene; FLE: fluorene; PHE: phenanthrene; ANT: anthracene; FLA: fluoranthene; PYR: pyrene; CHR: chrysene; BAA: benzo[a]anthracene; BBF: benzo[b]fluoranthene; BKF: benzo[k]fluoranthene; BAP: benzo[a]pyrene; BGP: benzo[g,h,i]perylene; IPY: indeno[1,2,3-cd]pyrene; DBP: dibenzo[a,h]pyrene.
ND: not detectable. Different letters above the columns indicate significant difference ($p < 0.05$).

counteracting the beneficial effects of the organic substrate supply (García-Gil et al., 2000; Franco et al., 2006; Mbarki et al., 2008; Roca-Pérez et al., 2009).

3.3 PAHs biodegradation

The benefits of animal manure and sewage sludge application to soil organic matter quantity, nutrient availability, soil aggregation, and other soil functions are well known (Mao et al., 2008; Roca-Perez et al., 2009). As seen, the organic wastes were rich in nutrients, such as N, P and K. This indicated that some manure-bound nutrients were gradually released from the organic component of the manure, which favored the growth of microorganisms and subsequently induced shifts in the metabolism of PAHs-degrading microorganisms, hence resulting in an improvement in PAHs biodegradation (Debosz et al., 2002; Xu and Obbard, 2003). As for DOM or dissolved organic carbon (DOC) originating from organic waste, this highlighted the possible role of DOM or DOC not only as nutrient source and PAHs-carrier but also as PAHs degradation enhancer (Kobayashi et al., 2009). Previous studies had shown that the DOM fraction of soil organic matter could positively contribute to PAHs bioavailability by enhancing the mobility of hydrophobic organic contaminants in aquifers and soils (Maxin et al., 1995; Haftka et al., 2008; Kobayashi et al., 2008).

In our experiment, the response of PAHs biodegradation was greatly different between CM and SS samples: apparent dosage effects on stimulation of PAHs dissipation were observed with CM but not SS. There are several possible explanations for this phenomenon. First, the partition of PAHs depended on the origin and properties of DOM. As reported by Raber and Kögel-Knabner (1997), the DOM from compost and sewage sludge can influence the transport of non-ionic organic contaminants because of the large concentrations of DOC released from these materials. The DOM from compost contained a large percentage of organic molecules > 14000 Da (32%–46%), whereas DOM from waste disposal leachates contained only 7%–10%, and so bonded less PAHs (Bengtsson and Zerhouni, 2003). Although the cattle manure and sewage sludge used were not characterized for their DOM component, the DOM concentrations were indeed different between them. Secondly, within an optimal concentration range of organic matter (or humic acid), biodegradation of PAHs in the plant-microbe system could be faster and more effective. As an important component of organic matter, there are contradictory conclusions on the interactions between humic acid (HA) and PAHs; some studies showed HA could increase the aqueous solubility of PAHs (Johnson and Amy, 1995; Haderlein et al., 2001), but other studies indicated that HA had no effect or even had a negative effect (Shimp and Pfaender, 1985). Liang et al. (2007) showed that the addition of Elliott soil humic acid solution into spiked soil enhanced pyrene mineralization at the range of 20–200 μg ESHA/g soil, but inhibition and neutral effects occurred beyond this concentration range. It was suggested that inhibition could be caused by the formation of micelles, which interfered with substrate transportation to the cell under high HA concentrations (Maxin et al., 1995). Finally, the absence of PAHs-degrader adaptation to degrade PAHs sorbed to HA could feasibly be considered. Taking into account that sorption of PAHs to HA affects bioavailability, a specific group of competent bacteria needs to be isolated to breach this barrier. However, the majority of PAHs degraders isolated by conventional enrichments with PAHs supplied as crystals in aqueous solutions probably lack characteristics that enable interactions with sorbents (Vaccas et al., 2005). In this case, the role of PAHs-degrading bacteria inoculation may be hindered by interaction between HA and PAHs in soil.

4 Conclusions

The present study demonstrated that organic waste amendments helped to significantly decrease soil PAHs in a plant-microbe bioremediation system. The possible underlying mechanism could partly be due to the fact that

amendments of organic wastes increased soil nutrients and soil organic matter, and then stimulated proliferation of soil microorganisms and promoted plant growth, and finally improved soil physical properties, fertility, and biological activity. In addition, the inoculated PAHs-degrading bacteria could also enhance the degradation rate and in turn reduce the toxic effect of PAHs to the plants.

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