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Full length article

The effects of activated biochar addition on remediation efficiency of co-composting with contaminated wetland soil

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ABSTRACT

Unreasonable disposal of organic wastes has caused great problems, including occupying land, affecting organoleptic feeling and causing environmental re-contamination. By incorporation of biochar as raw material into co-composting with agricultural organic matter, it not only can the wastes recycling be realized, but also can the remediation efficiency of multi-element contaminated soil be improved to reach land reclamation. Activated biochar was added for investigating the influences on physico-chemical properties of soil during composting. The concentrations of available metals and arsenic in soil were detected on great reduction in the treatment of biochar-blended composting. Based on the stronger adsorption and microbial activity induced by biochar, the initial content of available PAHs in wetland soil decreased to remain about 3.03% and 5.47% below the control (9.47%) in co-composting added with activated and fresh biochar, respectively. It was related to the observation with improved microbial number as well as optimized fungi/bacteria gene copy number ratio by activated biochar addition, which is conducive to vegetation replanting of contaminated land. The feasibility of applying biochar into co-composting with contaminated soil was confirmed by not only the high remediation efficiency but also the sustainable recycling of agricultural waste.

1. Introduction

Due to the increasingly rapid development of industrialization and frequent human activities, hazardous materials including metals (metalloid) and polycyclic aromatic hydrocarbons (Σ16PAHs: total concentrations of 16 PAHs which are served as priority pollutants and listed by USEPA) release into the wetland system with waste water, gas and residue (Chen et al., 2016; Lee et al., 2018; Zhang et al., 2011). Ultimately, these anthropogenic or natural pollutants may be accumulated in soil and further cause compound contamination, posing long-term complicated threats to human health and ecosystem (soil deterioration and desertification) because of their persistence and carcinogenic potential (Ke et al., 2010; Song et al., 2017; Zeng et al., 2013a,b). Moreover, the intricate interaction among co-existing pollutants may interfere with the remediation processes of contaminated soil (Huang et al., 2008; Ke et al., 2010; Wang et al., 2014; Ye et al., 2017b). Wang et al. (2010) reported that pollutants containing metals and PAHs are likely to change the humic content, water soluble organic carbon, and other essential properties in compost pile, and these changes make a complex case to influence the transformation of other coexisting pollutants. The development of suitable remediation technology is imperative, especially for the increasingly serious co-contaminated soil polluted by combination of metals (metalloid) and PAHs.

Biochar amendment and co-composting are not only identified as two attractive soil remediation technologies which have less negative impact on the environment, but also served as the attractive waste management options (Quina et al., 2017; Wu et al., 2016,a; Zeng et al., 2015). Biochar, produced by pyrolysis (under low oxygen conditions) of biomass, contains a large number of aromatic ring structures, and has been showed to achieve chemisorption and oxidation of pollutants as a consequence of surface microstructure (Alhashimi and Aktas, 2017; Xu et al., 2012; Ye et al., 2017a; Zhang et al., 2013b; Zeng et al., 2019). Composting is an aerobic bio-decomposition process of organic wastes, depending on widely distributed bacteria, actinomycetes, and fungi (Guo et al., 2018; Satari and Karimi, 2018; Wu et al., 2017a). Numerous studies showed that composting of contaminated soil (composting with waste raw materials and contaminated soil) is successful at improving nutrient content and decreasing the free metals by adsorption, redox

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Table 1
Chemical properties of co-composting materials. Data is presented as mean ± standard deviation (n = 3).

Properties	Tested soil	Fresh biochar	Activated biochar	Straw	Wheat bran
pH	7.56 ± 0.18	10.15 ± 0.16	9.86 ± 0.21	6.82 ± 0.13	5.23 ± 0.15
TOC[a] (g/kg)	30.25 ± 2.21	517.21 ± 32.15	473.28 ± 30.71	47.81 ± 2.62	40.23 ± 3.28
Total N (g/kg)	2.43 ± 0.30	6.86 ± 0.35	6.49 ± 0.41	0.95 ± 0.20	2.02 ± 0.31
C/N	12.45 ± 0.88	75.39 ± 2.51	72.92 ± 1.68	51.33 ± 1.62	20.06 ± 1.36
WEOC[b] (mg/kg)	81.01 ± 3.87	6.32 ± 0.73	4.85 ± 0.39	313.27 ± 26.85	352.32 ± 39.85
CEC[c] (cmol/kg)	13.56 ± 1.52	281.64 ± 7.67	365.87 ± 11.85	120.61 ± 6.95	136.17 ± 4.59

[a] Total organic carbon.
[b] Water-extractable organic carbon.
[c] Cation exchange capacity.

reaction, co-precipitation, (trans)methylation and complexation (Chen et al., 2015; Fan et al., 2008; Wu et al., 2017b). However, Beesley and Dickinson (2010) reported that adding highly organic wastes into soil contributes to a consequence of fast mobilization and vertical transportation of metals.

To maximize the potential benefits of biochar and co-composting, the interaction between them has been a research focus in recent years (Anyaoha et al., 2018; Wu et al., 2016; Ye et al., 2017a), and should also be the hotspot of further study (Karami et al., 2011; Zeng et al., 2015). Previous researches have proved that incorporation of biochar into composting materials shows improvement in physico-chemical properties of piles, acceleration in humification, and transformation in species and abundance of microorganism (Anyaoha et al., 2018; Jindo et al., 2012a; Otterpohl, 2012; Zeng et al., 2015; Zhang and Sun, 2014). Activated biochar, obtained surface microstructure modification by gas/acid/alkali etc., has captured the increasing attention because of its improved properties (Hadjittofi et al., 2014; Sizmur et al., 2017; Tan et al., 2015). Compared with fresh biochar, these improvements by activation can theoretically further promote composting efficiency: (1) activated biochar has higher porosity, interpenetrating pore structure is conducive to promote aeration conditions and transport oxygen molecules; (2) activated biochar has larger specific surface area, and the hydrophobic surface is the habitat for microbial attachment; (3) activated biochar has stronger carbon sequestration capacity, and the appropriate C/N ratio is beneficial to the growth and reproduction of microorganisms as energy source; (4) activated biochar has greater ion exchange capacity and more surface functional groups, and the improved surface is beneficial to enhance the adsorption capacity for pollutants. In the course of composting process, the surface of biochar is changed due to the adsorption of organic compounds and abiotic/biotic oxidation in composting (Wu et al., 2016). These changes on biochar surface may also affect their efficiency for soil remediation. Except for the changes in composting process, there are relatively few studies devoted to reduction of contaminants in composting assisted with biochar. The influences of activated biochar addition on the remediation efficiency of composting for co-polluted soil were meaningful and needed further research.

In this study, we initially investigated the effects of activated biochar addition on the remediation efficiency of co-composting for wetland soil contaminated with metals (including arsenic) and PAHs. The objectives of this research were to: (1) analyze the combined remediation efficiency of activated biochar addition and co-composting for soil contaminated with Zn, Cu, Cd, arsenic and PAHs; and (2) examine the changes of correlative fungi/bacterial gene copy number during composting with biochar addition, for microbes play a key role in composting processes and PAH degradation.

2. Materials and methods

2.1. Contaminated soil and amendment materials

Soil (clay: 24.19%, silt: 45.54% and sand: 30.27%) used in the study was obtained from 10 to 20 cm top-depth on beach of the Dongting Lake wetland (28°30′-29°38′ N, 112°18′-113°15′ E) which located in the middle reach of Yangtze River region in China. The lake beach is covered with phragmites and would be regarded as an important wintering habitat and pathway for East Asian migratory birds. However, unreasonable discharges of wastewater from mine field and metallurgical industries around the lake and upstream, besides the higher background value of pollutants, cause pollutant accumulation in water and soil (Zeng et al., 2015), as shown in Table S1. Due to the important role of Dongting Lake wetland in the global ecological protection (Wu et al., 2015), the remediation of the contaminated soil is essential. The collected soil was air-dried before sieving to a particle size of < 2 mm, and larger biological debris was removed in the meanwhile for the following tests. Biochar was manufactured by slow pyrolysis of corn cob in kilns operating at a continuous flow of N_2 gas and a temperature of 450 °C for a residence time of 2 h (Zeng et al., 2015). Dried biochar was crushed and sieved to 1 mm and thereby used as fresh biochar. Activated biochar was produced by pickling fresh biochar with 1 mol/L HCl for 30 min, and then the product was rinsed with deionized water and dried in oven at 65 °C for 12 h after filtration. The specific surface area and pore distributions of biochar were measured by N_2 adsorption-desorption conducted on a Micromeritics 2020 analyzer at 77 K. The detailed information is listed in Table 1.

2.2. Co-composting experiment

Original composting materials consisted of chopped straw, wheat bran, rotten vegetables and contaminated soil at a weight ratio of 11:2:3:8, called control treatment (Com). On the basis, the fresh and activated biochar were added to the composting materials at a weight ratio of 7% at the beginning of composting, which were called BCom and ABCom, respectively. Three kinds of aerobic compost pile were set up in a controlled environment chamber with 28% relative humidity, and the experiments were conducted in triplicate. To ensure proper aerobic composting, the piles were turned once everyday by automatic turned-pile system during the first 18 days, followed by flipping the piles once every 3 days. Deionized water was added twice a week to maintain the moisture content of 55 ~ 60% water filled pore space. The representative samples were taken on day 0, 7, 15, 30, and 45 during the composting process after turning piles. Samples were sealed in a self-sealed bag and stored in a refrigerator at −20 °C for the subsequent analysis and determination of indicators.

2.3. Analysis of the physico-chemical properties of composting samples

The temperature was measured daily by inserting the thermometer into the center of the pile. Sample pH was determined using digital pH meter by testing the filtrate after shaking with deionized water (1 g: 10 mL) at 200 r/min for 1 h. A total organic carbon analyzer (TOC-V $_{CPH}$, Shimadzu, Japan) was used to examine the water-extractable organic carbon (WEOC), for which the filtered supernatant was obtained from suspension (2.5 g sample: 25 mL water) undergone horizontal

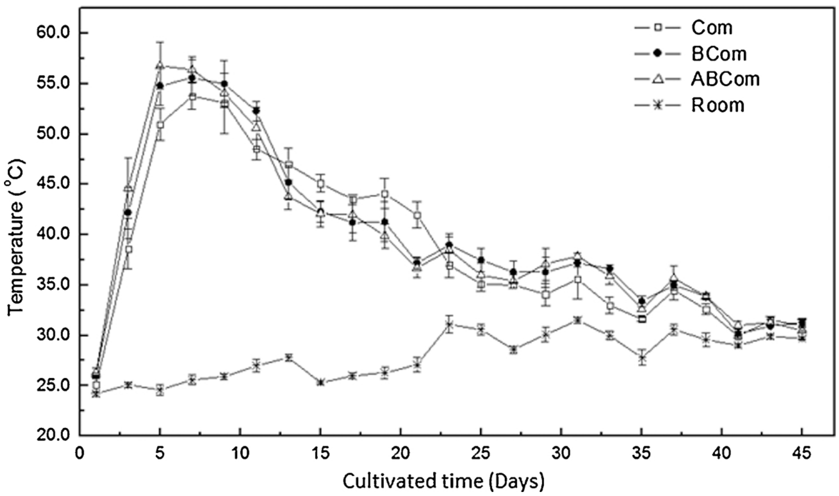

Fig. 1. Changes of temperature during composting (Com), composting added with fresh biochar (BCom) and composting added with activated biochar (ABCom), as well as room temperature. Error bars represent standard deviation (n = 3).

shake for 3 h and centrifugation at 3000 rpm for 10 min (Karami et al., 2011). According to Lu (1999), the determination of nitrate nitrogen (NO_3^--N) concentration depends on the phenol disulphonic acid spectrophotometric method, and the detection of ammonium nitrogen (NH_4^+-N) content relies on indophenol blue colorimetry method. Available P content of soil sample was measured according to previous study with $NaHCO_3$ extraction (Ngo et al., 2013).

2.4. Determination of pollutant concentrations of composting samples

The metal contents in contaminated soil amended with biochar-blended composting over exposure of 45 days were determined. Pseudototal concentrations of metal (contained metalloid) were extracted by aqua regia extractant ($HNO_3 + HCl$) and undergone microwave digestion process. As an indicator of pollution risk, available metals were extracted using 0.5 M $CaCl_2$ solution (5 g soil: 25 mL solution) by shaking for 3 h (250 r/min) and centrifuging for 10 min (3000 r/min). And the extracted metal contents in filtered supernatant were determined by atomic absorption spectrometry (Zeng et al., 2015). Content of available arsenic extracted by 0.5 M $CaCl_2$ solution was detected by atomic fluorescence spectrometry.

The concentrations of PAHs were examined by GC–MS analysis according to the method reported by Beesley et al. (2010). Three replicate samples of 5 g for each composting system were added into 10 mL mixture solution of acetone and hexane (volume ratio of 1:1) after thawing and drying. Extraction, acquired by shaking at 200 rpm for 2 h, was left to settle in a lasting time of 30 min. Subsequently, 2 mL resultant extract was loaded into a tube involving 0.1 g of dry sodium sulfate, preparing for the GC–MS determination. Bioavailable PAHs, a target chemical fraction which is available for bioconversion in soil, was measured based on cyclodextrin extractions. Composting sample (1.5 g) was placed in an Erlenmeyer flask, and 25 mL mixture of 60 mM hydroxypropyl-β-cyclodextrin (HPCD) in deionized water was added and homogenized in an orbital shaker for 20 h. The supernatant was discarded after centrifuging at 2500 rpm for 30 min. The remaining sample was shaken with 10 mL deionized water, and supernatant was also discarded after centrifuged again in order to remove any remaining HPCD. Resulting samples were exhaustively extracted by solution of acetone and hexane, and repeated above-mentioned GC–MS analysis for the measurement of remaining PAHs.

2.5. DNA extraction and qPCR amplification

Yang et al. (2007) provided a high-quality DNA extraction method of compost samples. According to their method, sample of 1.0 g was subjected to cell disruption and DNA extraction. The bacterial universal primer set, GC-338 F/518R, were chosen to amplify the variable V3 region of the 16S rDNA, and NS1 GC-Fung was used as primer set for that of fungal 18S rDNA. The qPCR assay was conducted by iCycler IQ5 Thermocycler (Bio-Rad, USA) with 20 μL volume mixture including 1 μL of template DNA, 0.2 μM of primers, 10 μL of 2 × SYBR real-time PCR premixture (Bioteke, Beijing, China) and sterile water. The operation conditions for amplification of bacterial primer were as follows: initial denaturation at 94 °C for 5 min, followed closely by 40 cycles of denaturing (45 s, 94 °C), annealing (40 s, 56 °C) and DNA elongation (40 s, 72 °C), along with a final extension for 10 min at 72 °C. Amplification of fungal primer was performed as below: denaturation at 94 °C for 5 min, followed by 40 cycles of denaturing (45 s, 94 °C), annealing (50 s, 55 °C) and DNA elongation (60 s, 72 °C), with a final 72 °C extension for 7 min. The standard curves of qPCR were generated using the ten-fold serial dilution of linearized plasmids which contains each gene (Wu et al., 2015). These standard curves were used to compare with the threshold cycle values of samples for the determination of initial gene copy number.

2.6. Statistical analysis

Data among different treatments were subjected to one way analysis of the variance (ANOVA) and the significant differences of their mean values were compared by Tukey's test at 5% level. The Pearson correlation analysis was applied to express the correlationship between related parameters. All statistical calculations were performed using SPSS Statistics (version 19, IBM Corporation, New York, USA).

3. Results and discussion

3.1. The effects of biochar addition on characteristics during composting

Temperature plays an important role in biological process, and it is considered as the main indicator for the performance of composting. As shown in Fig. 1, temperature in piles assisted with activated biochar increased rapidly and obtained highest peak temperature (Com: 52.23 ± 1.26a °C; BCom: 55.56 ± 1.73ab °C; ABCom: 56.87 ± 1.36b °C) in thermophilic phase, which reflects the enhancement of biological respiration intensity at this phase. The specific surface area (BET model) of activated biochar was detected by increasing from 76.8 m^2/g (fresh biochar) to 106.7 m^2/g through acid pickling for impurities removal, which provides more available sites for microbe habitat with

Table 2
Physico-chemical properties analyses of plies during composting (Com), composting added with fresh biochar (BCom) and composting added with activated biochar (ABCom). Data is presented as mean ± standard deviation (n = 3). Significant differences ($p < 0.05$) between each treatment in the same time are indicated by different letters.

	Materials	Days	pH	WEOC (mg/kg)	NH_4^+-N (mg/kg)	NO_3-N (mg/kg)	Available P (mg/g)
Com	chopped straw	0	7.88 ± 0.11a	332.51 ± 18.71a	343.40 ± 27.82a	149.31 ± 11.18a	1.22 ± 0.13a
	wheat bran	7	9.25 ± 0.15a	251.70 ± 23.80a	771.51 ± 33.38a	87.08 ± 7.92a	1.59 ± 0.32a
	rotten vegetables	15	8.31 ± 0.13a	198.39 ± 13.32a	468.27 ± 30.66a	198.23 ± 18.61a	4.25 ± 0.63a
	contaminated soil	30	8.03 ± 0.10a	151.73 ± 14.39a	330.38 ± 24.32a	311.72 ± 21.60a	4.12 ± 0.40a
		45	7.56 ± 0.06a	128.27 ± 8.64a	267.61 ± 17.02a	326.43 ± 14.67a	3.75 ± 0.51a
BCom	chopped straw	0	8.19 ± 0.07b	358.21 ± 16.34a	297.12 ± 18.47ab	130.63 ± 8.81a	0.73 ± 0.02a
	wheat bran	7	8.98 ± 0.21a	223.82 ± 28.61ab	716.24 ± 34.01a	108.72 ± 8.90ab	1.21 ± 0.12a
	rotten vegetables	15	7.88 ± 0.15b	151.18 ± 18.08b	349.29 ± 28.56b	244.01 ± 15.68b	3.25 ± 0.39ab
	contaminated soil	30	8.08 ± 0.12a	140.10 ± 15.36a	320.81 ± 31.53a	280.78 ± 17.83a	1.98 ± 0.16b
	7% fresh biochar	45	7.31 ± 0.08ab	104.73 ± 10.27b	215.40 ± 13.13b	294.49 ± 12.59b	1.37 ± 0.05b
ABCom	chopped straw	0	8.10 ± 0.08b	349.79 ± 15.52a	284.73 ± 22.72b	136.52 ± 10.30a	0.81 ± 0.05a
	wheat bran	7	9.10 ± 0.22a	208.30 ± 29.81b	726.58 ± 38.50a	117.82 ± 10.14b	1.18 ± 0.27a
	rotten vegetables	15	7.71 ± 0.19b	146.43 ± 15.09b	327.69 ± 27.14b	256.73 ± 20.32b	3.10 ± 0.51b
	contaminated soil	30	8.17 ± 0.16a	142.13 ± 16.55a	325.01 ± 37.59a	289.27 ± 16.15a	2.11 ± 0.06b
	7% activated biochar	45	7.17 ± 0.08b	83.92 ± 9.31c	203.32 ± 15.53b	305.03 ± 10.91ab	1.93 ± 0.03ab

WEOC: water-extractable organic carbon.

improving metabolic activity (Wei et al., 2014). Furthermore, owing to the higher porosity and nano-particle size, activated biochar could be used as an oxygen carrier to enhance aeration in piles (Zhang et al., 2014), strengthening the microbial respiration and resulting in largest peak of temperature. However, due to the limited number of the easily decomposable materials, the activities of microorganism were restricted at later thermophilic phase and caused slightly shorter duration of this phase in activated biochar treatment than that of fresh biochar treatment.

The pH values in three treatments on the course of composting are displayed on Table 2, and all pH values in the process of composting were in the range of suitable for microbial growth. The variation in pH value is determined by the NH_3 releasing, organic acid formation and ammonification processes as rising one after another during composting. It should be pointed out that the lower resultant pH was observed by activated biochar treatment, which is attributed to more humic acids isolated from degradation of organic materials by higher microbial metabolism. Another explanation is that the stronger carbon and nitrogen immobilization capacity of activated biochar than that of fresh one can decrease the available N and retain larger amount of ammonia gas and nitrate ions, which is consistent with the observation from the lowest inorganic nitrogen containing ammonium-nitrogen (NH_4^+-N) and nitrate nitrogen (NO_3^--N) in the activated biochar treatment (Table 2). The highest concentrations of NH_4^+-N was observed at thermophilic phase yet low concentration of NO_3^--N occurred at that phase, because the activity of nitrifying bacteria is restricted by the high temperature and excessive amounts of ammonia (Zhang and Sun, 2014). However, the decrease of bacteria activity was mitigated in the present of biochar by both functions of the biochar protection and higher microbial quantity.

All amendments with highly organic material resulted in high content of water-extractable organic carbon (WEOC) at initial phase. Our previous work had proved that the growth of microorganisms is closely correlated with WEOC but little with TOC (Zeng et al., 2015). Due to the recalcitrant aromatic ring structure of biochar (Wei et al., 2014), the carbon pool of biochar is comparatively stable and insoluble, and there were insignificant differences of WEOC between all treatments at the beginning. In assistant with activated biochar, the composting treatment finally obtained lowest content of WEOC, and the most rapid decrease of WEOC was presented on ABCom at thermophilic phase. Overall, intensive microbial metabolism for making use of WEOC as energy source and excellent adsorption of activated biochar for carbon immobilization are responsible for the larger extent decrease of WEOC and lowest resultant WEOC content in ABCom. It is worth to mention that the reduction of WEOC was observed to be stagnated in biochar additions at day 30, because the stronger microbial activities (especially the fungi) and higher temperature could convert the refractory organics into easily mineralized form to make up for the shortage of resources at cooling stage where fungi dominated.

3.2. The effects of biochar addition on contaminants during composting

Soil used in the test is polluted by metals, and Cd, Zn, Cu and arsenic contained therein were chosen as the representative of the examination due to higher pollution level (Table S1). They are all higher than the tree-level standard value (GB 15618-1995) of soil metal pollution (critical value for ensuring agricultural and forestry production and normal growth of plants). Co-composting of contaminated soil, either individually or in combination with biochar had little effect on pseudototal metal content as shown in Fig. 2, because these metals (metalloid) could not be break down by microbial metabolism. It was noteworthy that the comparatively low contents of pseudototal metal than untreated soil was associated with a dilution effect of raw materials of composting, which was larger in mass compared with pure soil (Beesley et al., 2010; Zeng et al., 2015). The available metal, although only a small fraction of metal, is always used to characterize the bioavailability and eco-toxicity of metal in soil (Zeng et al., 2015). As shown in Fig. 2, concentrations of available metal extracted from piles present different amendment responses on various treatments.

In terms of various remediation efficiency from metal types, the superiority of ABCom was significant in reducing available concentrations of Zn and Cd than the performance of BCom ($p < 0.05$), devoting to a 7 fold decrease of available Zn and a 2.5 fold decrease of available Cd than corresponding indicators in Com. It is associated with the increasing reactive sites on activated biochar with larger specific surface area, more surface functional groups, and stronger ion exchange capacity. In addition to the chelating effect by organic matter in composting to form metal-organic bound (Clemente and Bernal, 2006), biochar possess superior capability for immobilizing metals through metal exchange, chemical binding, inner-sphere surface complexation and precipitation (Zhang et al., 2013b). Moreover, organic matter could be adsorbed on biochar surface, and it is acted as a bridge between biochar and metals for pollutant immobilization. As shown in Fig. 2, higher levels of available Cu and arsenic were detected in Com than untreated soil, which could be attributed in part to the formation of soluble organometallic complexes with the elevating dissolved organic carbon (DOC) in composting process (Beesley and Dickinson, 2010; Karami et al., 2011; Moreno-Jimenez et al., 2013), as well as the enhanced competition between increasing DOC and metals for binding sites of soil particle surface (Fitz and Wenzel, 2002). Since the

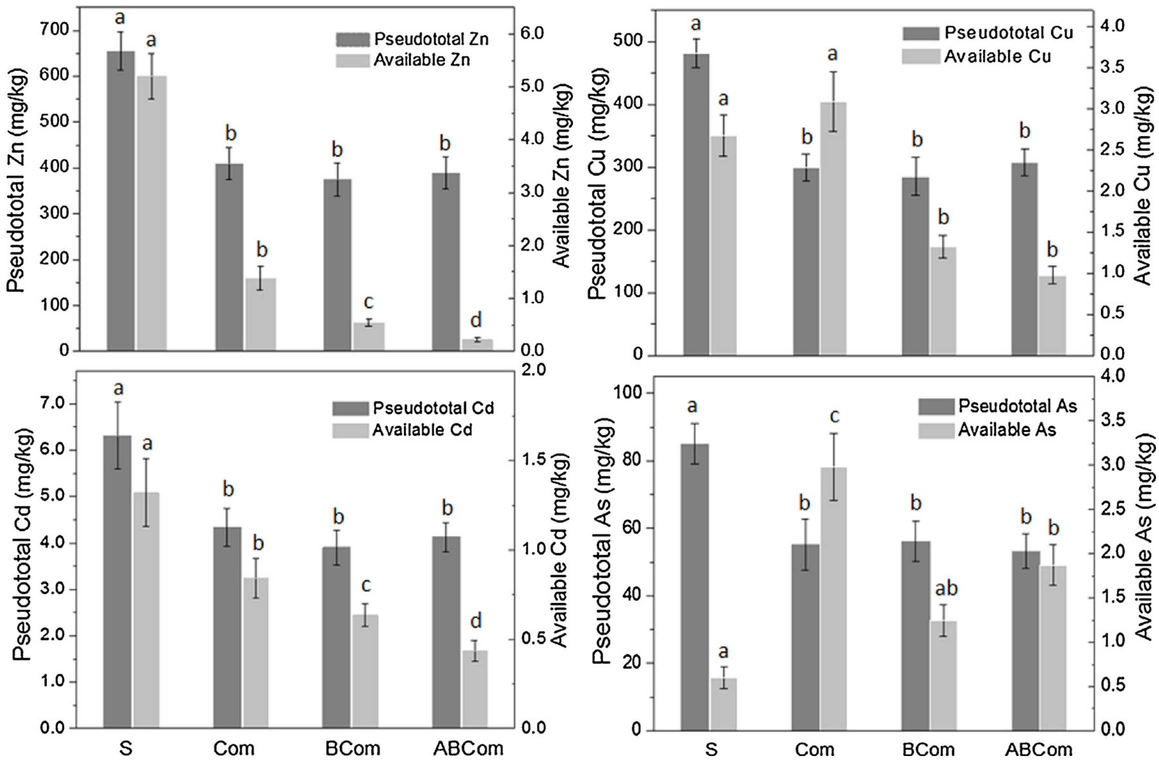

Fig. 2. Concentrations of pseudototal and available metals (contained metalloid) following 45 days exposure. Error bars represent standard error of the mean (n = 3).

complexes formed between WEOC and metal ions normally become mobilized, lower WEOC (Table 2) and additional strong adsorption capacity caused by biochar assistance led to the Cu availability drop below that of untreated soil at the end of composting. Fresh biochar amendment visibly reduced arsenic mobility below Com owing to the combined effect of adsorption capacity, lower WEOC as well as pH effect. Lower pH value in biochar addition that discussed above was responsible a part for the lower available arsenic, since arsenic solubility might decrease with more acid pH (Beesley et al., 2010, 2014). However, because the evaluated available P might compete with arsenic for retention sites of biochar or soil particles, the combination of composting and activated biochar showed increase in arsenic leaching than BCom. The transformation of valence and speciation of arsenic on the activated biochar surface in co-composting also influence the arsenic mobility

Compared with untreated soil, organic amendments containing co-composting of contaminated soil alone or accompanied by biochar were effective at decreasing the concentrations of both total and available PAHs (Fig. 3). Applying highly organic materials by co-composting of contaminated soil introduces a great number of microorganisms and nutrients. Microbial activities containing enzymatic secretion and metabolism are conducive to reducing the PAHs concentrations by serving PAHs as substrate and energy sources (Chen et al., 2015). Furthermore, a portion of organic matter produced from compost materials could increase the solubility of PAHs, which improves the accessibility of PAHs to the degrading microbes (Kobayashi et al., 2009), but sometimes the sorption by organic matter might contribute to the organic-binding of PAHs (Plaza et al., 2009). As shown on Fig. 3, the addition of activated biochar received great degradation rate of PAHs in early stage, which is associated with the increase in microbial number induced by activated biochar (Fig. 4), thereby enhancing the enzymatic activity involved in recalcitrant compound degradation (Jindo et al., 2012b). But the gaps of residual content of total PAHs (TPAHs) between composting alone and biochar combination were narrowed with cultivated time, and the statistical difference disappeared after 45 days

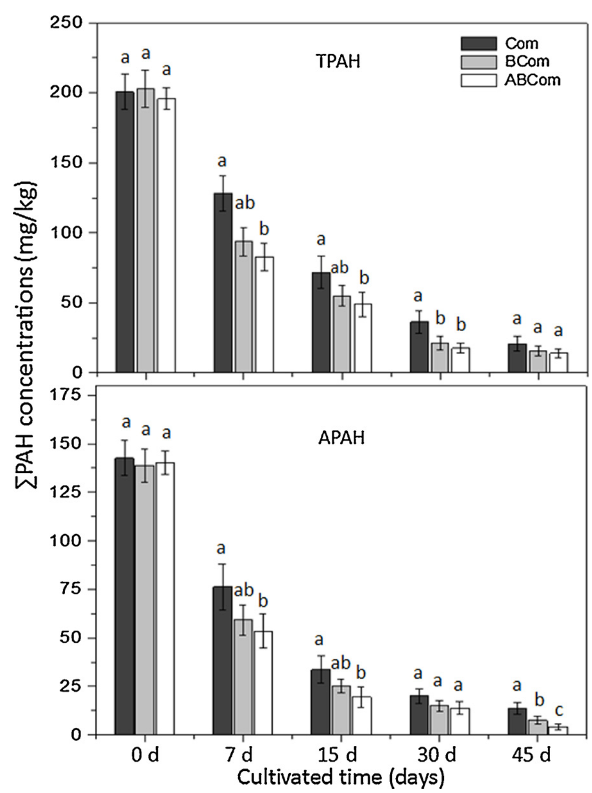

Fig. 3. Residual concentration of total and available PAH during the incubation time. Error bars represent standard deviation (n = 3). Significant differences ($p < 0.05$) between each treatment in the same time are indicated by different letters.

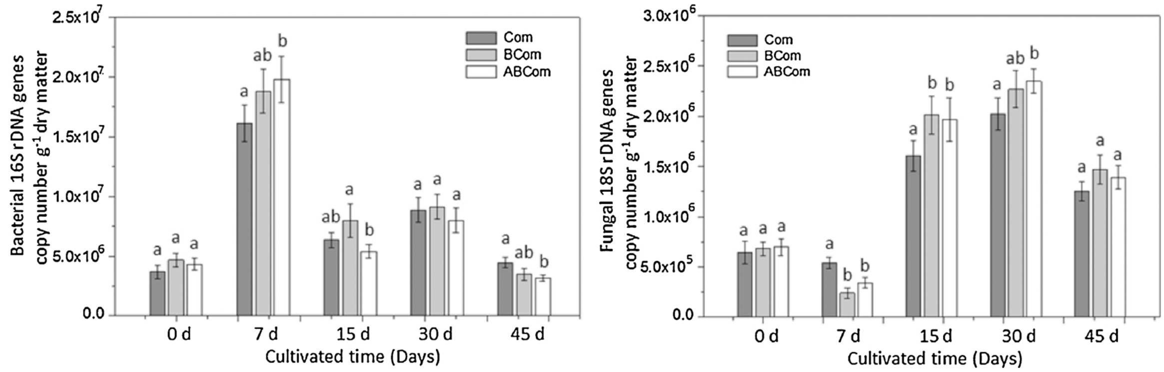

Fig. 4. Changes in gene copy numbers per gram of dry matter for bacterial 16S rDNA and fungal 18S rDNA at sampling time across composting. Error bars represent standard error of the mean (n = 3). Different letters indicate significant difference ($p < 0.05$) between each treatment.

incubation ($p > 0.05$), indicating no statistical difference of TPAHs content remaining in treatment groups. It was similar with Beesley et al. (2010), who found antagonistic effect on PAH degradation when compost and biochar amended in combination. On one hand, available nutrients for microbial metabolism were assembled and adsorbed by biochar, restricting the microbial growth and activity. On the other hand, the decrease in bioaccessibility of PAHs for microbes caused by biochar adsorption also limits the matter biodegradation. This result contradicts Marchal et al. (2013), whose observation indicated that there was no inhibitory effect of sorption by biochar on phenanthrene biodegradation, with the premise that bacterial number maintained adequate enough.

Notably, available PAHs (APAHs), considered as a bioavailable fraction, was found to finally remain significant lower in biochar treatment than that of control, due to the functions of high microbial metabolism, as well as strong adsorption of biochar through electron donor-acceptor interactions, pore-filling and hydrophobic effect on biochar surface. Since high ash content might obstruct available sites on the surface (Zhang et al., 2013a), activated biochar shows more binding sites for sequestration of PAHs. Assisting with other advantages of intense microbial activity, and low metal toxicity in pile, ABCom was observed by the lowest residue content of APAHs. There were no statistically significant differences among all treatments on the APAH concentrations at day 30, indicating that the decrease rate of APAH content was slowed down in treatments added biochar. It may be explained by the phenomenon that fungi dominated in this stage (Fig. 5) which contributed to the accumulation of APAHs, since the high-molecular-weight PAHs could be converted preliminary to the light and available form by fungus metabolism. Besides, the decomposition of humic acid by intense fungal activities can release the organic-bound PAHs to available form (Plaza et al., 2009). Following 45 days exposure, the reduction in average of APAH concentration with the treatments of Com, BCom and ABCom was 90.53%, 94.45% and 96.90%, respectively. Compared with control, the residual content of APAHs was reduced with greater than 65% and 40% decrease in ABCom and BCom, respectively.

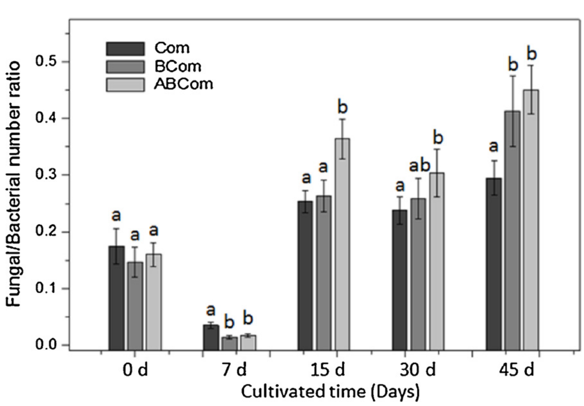

Fig. 5. The variation of gene copy numbers ratio values of fungal to bacterial during composting at three treatments. Error bars represent standard deviation (n = 3). Different letters indicate significant difference (p < 0.05) between each treatment in the same time.

3.3. The effects of biochar amendment on microorganisms during composting

Gene copy numbers of bacterial 16S rDNA and fungal 18S rDNA, shown in Fig. 4, reflect the quantity changes of microorganism during co-composting with contaminated soil. Microbial quantity is largely influenced by temperature and WEOC over the course of composting. ABCom maximized the quantity of bacteria in the early thermophilic phase, while BCom had little larger bacterial quantity in other composting phase. When biochar was used as the co-material in composting, larger number of bacteria was observed (Fig. 4), because biochar can reduce bulk density and improve soil structure, thereby providing more appropriate conditions including substantial nutrients, aeration environment and colonization chances for microbial growth and reproduction (Ngo et al., 2013; Zhang et al., 2014; Zhang and Sun, 2014). Moreover, the bio-toxicity might be decreased as the contents of available contaminants reducing when biochar incorporated into the co-composting. It is beneficial to the growth of sensitive bacteria (Marchal et al., 2014). The activated biochar has a pore volume of 0.06017 cm^3/g, about 3 times larger than the fresh biochar (0.02657 cm^3/g), indicating a better growth condition for microbes. Particularly, the micro-pores of activated biochar might protect microbes against grazers or other competitors. Integrated considering the limited concentration of available pollutants, the number of bacteria rises rapidly in the early stage and reaches a maximum in the thermophilic phase with the aid of activated biochar. However, rapid content decrease in easily decomposed organic matter by microbial decomposition and biochar adsorption contributed to the lowest final bacterial number in ABCom, coinciding with WEOC content in the end. Besides, fresh biochar is partly beneficial to the microbial growth associated with mineral contents (e.g. K, Ca and Na) (Hua et al., 2012), which might lead to slightly larger bacterial number in BCom than ABCom.

In contrast, the opposite trends were detected in gene copy number of 18S rDNA, with the smallest number of fungi observed at day 7, owing to the relatively weak position compared with abundant bacteria for carbon and energy source at that stage. Upon the basis of consumption of easily degradable carbon source, fungi obtain competitive advantage than bacteria thanks to their amazing catabolic abilities to use more recalcitrant carbon compounds contained large-ringed PAHs (Covino et al., 2016; Langarica-Fuentes et al., 2014), correlated well with the fugal number followed by increasing progressively and reached top value during mesophilic and cooling phase. Insignificant difference in gene copy number of 18S rDNA was counted at the end (Fig. 4), although obvious difference of WEOC (Table 2) existed, which also indicated the ability of fungi to transform recalcitrant organic matter. Hua et al. (2012) found that nutrients could be retained to higher level by biochar for preventing the nutrient loss. The nutrients, reserved on biochar, could be released slowly and became available for microbial metabolism, contributing to greater number of fungi in treatments with biochar. In general, there was less influence of biochar addition on fungal number than that on bacterial number based on the corresponding Pearson coefficient (Table S3).

The ratio of fungal to bacterial gene copy numbers could show the microbial community shift in composting processes (Covino et al., 2016). It must also be mentioned that the addition of biochar could further optimize the ratio of fungi to bacteria, which can be observed from Fig. 5. Incorporation of the fresh and activated biochar into composting materials helped the gene copy numbers ratio of fungal to bacterial reach up to 41.3% and 45.1%, which revealed an 11.8% and 15.6% increase than control at maturation phase, respectively. Some bacteria such as archaebacteria could also withstand and grow after extreme conditions (Antizar-Ladislao et al., 2008), it is responsible for the stagnant increase of the ratio of fugal to bacterial number at day 30. According to the Pearson coefficient of the relevant parameters presented on Table S3, the analysis showed that the degradation amount of APAH is positively correlated with the number of bacteria. More notable is that the correlation coefficient masked with asterisk is below the significance level of 0.05, indicating that the degraded content of APAHs had a significant correlation with the ratio of bacterial/fungi. Since biochar is conducive to slowly releasing the retained nutrients, its function sustainability might influence the quantity of microorganisms for long-terms.

4. Conclusions

The present work initially demonstrated that co-composting of contaminated soil with activated biochar addition showed efficient performance for decontamination and detoxification of soil polluted with metals and PAHs in tidal wetlands. Activated biochar with more excellent surface characteristics, served as co-substrate can receive significantly positive effect on composting with soil structure modification and nutrient retention, providing better condition for microbial growth and activity. Biochar-blended co-composting combines the prominent positive effects of biochar and composting, for not only decreasing the available concentrations of pollutants in contaminated soil but also promoting sustainable agriculture. Further researches are required for investigating the transformation of metal speciation and the shift of detail microbial species for PAH degradation during co-composting with activated biochar addition. And the long-term effects of biochar on composting efficiency also require deeper study.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.resconrec.2018.10.004.

References

Alhashimi, H.A., Aktas, C.B., 2017. Life cycle environmental and economic performance of biochar compared with activated carbon: a meta-analysis. Resour. Conserv. Recyl. 118, 13–26.
Antizar-Ladislao, B., Spanova, K., Beck, A.J., Russell, N.J., 2008. Microbial community structure changes during bioremediation of PAHs in an aged coal-tar contaminated soil by in-vessel composting. Int. Biodeter. Biodegr. 61, 357–364.
Anyaoha, K.E., Sakrabani, R., Patchigolla, K., Mouazen, A.M., 2018. Critical evaluation of oil palm fresh fruit bunch solid wastes as soil amendments: prospects and challenges. Resour. Conserv. Recyl. 136, 399–409.
Beesley, L., Dickinson, N., 2010. Carbon and trace element mobility in an urban soil amended with green waste compost. Int. J. Soil Sediment. Water 10, 215–222.
Beesley, L., Moreno-Jimenez, E., Gomez-Eyles, J.L., 2010. Effects of biochar and green-waste compost amendments on mobility, bioavailability and toxicity of inorganic and organic contaminants in a multi-element polluted soil. Environ. Pollut. 158, 2282–2287.
Beesley, L., Inneh, O.S., Norton, G.J., Moreno-Jimenez, E., Pardo, T., Clemente, R., Dawson, J.J., 2014. Assessing the influence of compost and biochar amendments on the mobility and toxicity of metals and arsenic in a naturally contaminated mine soil. Environ. Pollut. 186, 195–202.
Chen, M., Xu, P., Zeng, G., Yang, C., Huang, D., Zhang, J., 2015. Bioremediation of soils contaminated with polycyclic aromatic hydrocarbons, petroleum, pesticides, chlorophenols and metals by composting: Applications, microbes and future research needs. Biotechnol. Adv. 33, 745–755.
Chen, F., Tan, M., Ma, J., Zhang, S., Li, G., Qu, J., 2016. Efficient remediation of PAH-metal co-contaminated soil using microbial-plant combination: a greenhouse study. J. Hazard. Mater. 302, 250–261.
Clemente, R., Bernal, M.P., 2006. Fractionation of metals and distribution of organic carbon in two contaminated soils amended with humic acids. Chemosphere 64, 1264–1273.
Covino, S., Fabianova, T., Kresinova, Z., Cvancarova, M., Burianova, E., Filipova, A., et al., 2016. Polycyclic aromatic hydrocarbons degradation and microbial community shifts during co-composting of creosote-treated wood. J. Hazard. Mater. 301, 17–26.
Fan, T., Liu, Y., Feng, B., Zeng, G., Yang, C., Zhou, M., et al., 2008. Biosorption of cadmium(II), zinc(II) and lead(II) by Penicillium simplicissimum: Isotherms, kinetics and thermodynamics. J. Hazard. Mater. 160, 655–661.
Fitz, W.J., Wenzel, W.W., 2002. Arsenic transformations in the soil-rhizosphere-plant system: fundamentals and potential application to phytoremediation. J. Biotechnol. 99, 259–278.
Guo, W., Zhou, Y., Zhu, N., Hu, H., Shen, W., Huang, X., Zhang, T., Wu, P., Li, Z., 2018. On site composting of food waste: a pilot scale case study in China. Resour. Conserv. Recyl. 132, 130–138.
Hadjittofi, L., Prodromou, M., Pashalidis, I., 2014. Activated biochar derived from cactus fibres - preparation, characterization and application on Cu(II) removal from aqueous solutions. Bioresour. Technol. 159, 460–464.
Hua, L., Chen, Y.X., Wu, W.X., 2012. Impacts upon soil quality and plant growth of bamboo charcoal addition to composted sludge. Environ. Technol. 33, 61–68.
Huang, D.L., Zeng, G.M., Feng, C.L., Hu, S., Jiang, X.Y., Tang, L., et al., 2008. Degradation of lead-contaminated lignocellulosic waste by Phanerochaete chrysosporium and the reduction of lead toxicity. Environ. Sci. Technol. 42, 4946–4951.
Jindo, K., Sanchez-Monedero, M.A., Hernandez, T., Garcia, C., Furukawa, T., Matsumoto, K., et al., 2012a. Biochar influences the microbial community structure during manure composting with agricultural wastes. Sci. Total Environ. 416, 476–481.
Jindo, K., Suto, K., Matsumoto, K., Garcia, C., Sonoki, T., Sanchez-Monedero, M.A., 2012b. Chemical and biochemical characterisation of biochar-blended composts prepared from poultry manure. Bioresour. Technol. 110, 396–404.
Karami, N., Clemente, R., Moreno-Jimenez, E., Lepp, N.W., Beesley, L., 2011. Efficiency of green waste compost and biochar soil amendments for reducing lead and copper mobility and uptake to ryegrass. J. Hazard. Mater. 191, 41–48.
Ke, L., Luo, L., Wang, P., Luan, T., Tam, N.F., 2010. Effects of metals on biosorption and biodegradation of mixed polycyclic aromatic hydrocarbons by a freshwater green alga Selenastrum capricornutum. Bioresour. Technol. 101, 6961–6972.
Kobayashi, T., Murai, Y., Tatsumi, K., Iimura, Y., 2009. Biodegradation of polycyclic aromatic hydrocarbons by Sphingomonas sp. enhanced by water-extractable organic matter from manure compost. Sci. Total Environ. 407, 5805–5810.
Langarica-Fuentes, A., Zafar, U., Heyworth, A., Brown, T., Fox, G., Robson, G.D., 2014. Fungal succession in an in-vessel composting system characterized using 454 pyrosequencing. FEMS Microbiol. Ecol. 88, 296–308.
Lee, G., Cui, M., Yoon, Y., Khim, J., Jang, M., 2018. Passive treatment of arsenic and metals contaminated circumneutral mine drainage using granular polyurethane impregnated by coal mine drainage sludge. J. Clean. Prod. 186, 282–292.
Lu, R., 1999. Soil Agricultural Chemical Analysis Method. China's agricultural science and technology press, Beijing.
Marchal, G., Smith, K.E., Rein, A., Winding, A., Trapp, S., Karlson, U.G., 2013. Comparing the desorption and biodegradation of low concentrations of phenanthrene sorbed to activated carbon, biochar and compost. Chemosphere 90, 1767–1778.
Marchal, G., Smith, K.E., Mayer, P., Wollesen de Jonge, L., Jonge, L., Karlson, U.G., 2014. Impact of soil amendments and the plant rhizosphere on PAH behaviour in soil. Environ. Pollut. 188, 124–131.
Moreno-Jimenez, E., Clemente, R., Mestrot, A., Meharg, A.A., 2013. Arsenic and selenium mobilisation from organic matter treated mine spoil with and without inorganic fertilisation. Environ. Pollut. 173, 238–244.
Ngo, P., Rumpel, C., Ngo, Q., Alexis, M., Velásquez Vargas, G., Mora Gil Mde, L., et al., 2013. Biological and chemical reactivity and phosphorus forms of buffalo manure

compost, vermicompost and their mixture with biochar. Bioresour. Technol. 148, 401–407.
Otterpohl, R., 2012. Boosting compost with biochar and bacteria. Nature 486, 187–188.
Plaza, C., Xing, B., Fernandez, J.M., Senesi, N., Polo, A., 2009. Binding of polycyclic aromatic hydrocarbons by humic acids formed during composting. Environ. Pollut. 157, 257–263.
Quina, M.J., Soares, M.A.R., Quinta-Ferreira, R., 2017. Applications of industrial eggshell as a valuable anthropogenic resource. Resour. Conserv. Recyl. 123, 176–186.
Satari, B., Karimi, K., 2018. Citrus processing wastes: environmental impacts, recent advances, and future perspectives in total valorization. Resour. Conserv. Recyl. 129, 153–167.
Sizmur, T., Fresno, T., Akgul, G., Frost, H., Moreno-Jimenez, E., 2017. Biochar modification to enhance sorption of inorganics from water. Bioresour. Technol. 246, 34–47.
Song, B., Zeng, G., Gong, J., Zhang, P., Deng, J., Deng, C., et al., 2017. Effect of multi-walled carbon nanotubes on phytotoxicity of sediments contaminated by phenanthrene and cadmium. Chemosphere 172, 449–458.
Tan, X., Liu, Y., Zeng, G., Wang, X., Hu, X., Gu, Y., Yang, Z., 2015. Application of biochar for the removal of pollutants from aqueous solutions. Chemosphere 125, 70–85.
Wang, C., Wang, F., Wang, T., Bian, Y., Yang, X., Jiang, X., 2010. PAHs biodegradation potential of indigenous consortia from agricultural soil and contaminated soil in two-liquid-phase bioreactor (TLPB). J. Hazard. Mater. 176, 41–47.
Wang, W., Zhang, X., Huang, J., Yan, C., Zhang, Q., Lu, H., et al., 2014. Interactive effects of cadmium and pyrene on contaminant removal from co-contaminated sediment planted with mangrove Kandelia obovata (S., L.) Yong seedlings. Mar. Pollut. Bull. 84, 306–313.
Wei, L., Shutao, W., Jin, Z., Tong, X., 2014. Biochar influences the microbial community structure during tomato stalk composting with chicken manure. Bioresour. Technol. 154, 148–154.
Wu, H.P., Zeng, G.M., Liang, J., Guo, S.L., Dai, J., Lu, L.H., et al., 2015. Effect of early dry season induced by the Three Gorges Dam on the soil microbial biomass and bacterial community structure in the Dongting Lake wetland. Ecol. Indic. 53, 129–136.
Wu, H., Zeng, G., Liang, J., Chen, J., Xu, J., Dai, J., et al., 2016. Responses of soil bacterial community and functional marker genes of nitrogen cycling to biochar and compost (or composting) combined amendment in soil. Appl. Microbiol. Biotechnol. 100, 8583–8591.
Wu, H., Lai, C., Zeng, G., Liang, J., Chen, J., Xu, J., Dai, J., Li, X., Liu, J., Chen, M., Lu, L., Hu, L., Wan, J., 2017a. The interactions of composting and biochar and their implications for soil amendment and pollution remediation: a review. Crit. Rev. Biotechnol. 37 (6), 754–764.
Wu, Y., Yang, J., Tang, J., Kerr, P., Wong, P., 2017b. The remediation of extremely acidic and moderate pH soil leachates containing Cu (II) and Cd (II) by native periphytic biofilm. J. Clean. Prod. 162, 846–855.
Xu, P., Zeng, G.M., Huang, D.L., Feng, C.L., Hu, S., Zhao, M.H., et al., 2012. Use of iron oxide nanomaterials in wastewater treatment: a review. Sci. Total Environ. 424, 1–10.
Yang, Z.H., Xiao, Y., Zeng, G.M., Xu Zh, Y., Liu, Y., 2007. Comparison of methods for total community DNA extraction and purification from compost. Appl. Microbiol. Biotechnol. 74, 918–925.
Ye, S., Zeng, G., Wu, H., Zhang, C., Dai, J., Liang, J., et al., 2017a. Biological technologies for the remediation of co-contaminated soil. Crit. Rev. Biotechnol. 37 (8), 1062–1076.
Ye, S., Zeng, G., Wu, H., Zhang, C., Liang, J., Dai, J., Liu, Z., Xiong, W., Wan, J., Xu, P., Cheng, M., 2017b. Co-occurrence and interactions of pollutants, and their impacts on soil remediation-a review. Crit. Rev. Environ. Sci. Technol. 47, 1528–1553.
Zeng, G., Chen, M., Zeng, Z., 2013a. Risks of neonicotinoid pesticides. Science 340, 1403.
Zeng, G.M., Chen, M., Zeng, Z.T., 2013b. Shale gas: surface water also at risk. Nature 499 154–154.
Zeng, G., Wu, H., Liang, J., Guo, S., Hu, L., Xu, P., et al., 2015. Efficiency of biochar and compost (or composting) combined amendments for reducing Cd, Cu, Zn and Pb bioavailability, mobility and ecological risk in wetland soil. RSC Adv. 5, 34541–34548.
Zeng, Z., Ye, S., Wu, H., Xiao, R., Zeng, G., Liang, J., Zhang, C., Yu, J., Fang, Y., Song, B., 2019. Research on the sustainable efficacy of g-MoS2 decorated biochar nanocomposites for removing tetracycline hydrochloride from antibiotic-polluted aqueous solution. Sci. Total Environ. 648, 206–217.
Zhang, L., Sun, X., 2014. Changes in physical, chemical, and microbiological properties during the two-stage co-composting of green waste with spent mushroom compost and biochar. Bioresour. Technol. 171, 274–284.
Zhang, Z., Rengel, Z., Meney, K., Pantelic, L., Tomanovic, R., 2011. Polynuclear aromatic hydrocarbons (PAHs) mediate cadmium toxicity to an emergent wetland species. J. Hazard. Mater. 189, 119–126.
Zhang, P., Sun, H., Yu, L., Sun, T., 2013a. Adsorption and catalytic hydrolysis of carbaryl and atrazine on pig manure-derived biochars: impact of structural properties of biochars. J. Hazard. Mater. 244–245, 217–224.
Zhang, X.K., Wang, H.L., He, L.Z., Lu, K.P., Sarmah, A., Li, J.W., et al., 2013b. Using biochar for remediation of soils contaminated with metals and organic pollutants. Environ. Sci. Pollut. Res. 20, 8472–8483.
Zhang, J.N., Lu, F., Shao, L.M., He, P.J., 2014. The use of biochar-amended composting to improve the humification and degradation of sewage sludge. Bioresour. Technol. 168, 252–258.