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Assessment of maturity during co-composting of penicillin mycelial dreg via fluorescence excitation-emission matrix spectra: Characteristics of chemical and fluorescent parameters of water-extractable organic matter

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HIGHLIGHTS

- Residual penicillin decayed with a degradation rate of 98.1% on the 6th day.
- Biochemical parameters indicated the co-composting endpoint with a stable level.
- $P_{V,n}/P_{III,n}$ based on FRI can reflect maturity for co-composting of PMD.
- EEM-FRI has the potential for characterizing the process of this co-composting.

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ABSTRACT

To investigate characteristics of water-extractable organic matter (WEOM) from different stages and evaluate the maturity for co-composting penicillin mycelial dreg (PMD) via fluorescence regional integration (FRI) of fluorescence excitation-emission matrix (EEM), a pilot-scale co-composting was carried out. The results showed that a classical temperature profile showed and a degradation rate of 98.1% for residual penicillin was obtained on the 6th day. DOC and DOC/DON ratio were in a low level of 4.0 g kg^{-1} and 3.7, respectively, after the 32nd day. In addition, respirometric rate (SOUR) decreased to 0.87 mg O_2 g^{-1} VS h^{-1} finally. The EEM showed that the specific Ex/Em peak related to microbial byproduct-like vanished on the 32nd day, while those related to fulvic-like and humic acid-like appearing on the 24th day. The fluorescence regional integration (FRI) results demonstrated that $P_{V,n}/P_{III,n}$ increased to 3.28 finally, suggesting a desirable maturity for co-composting PMD. The EEM-FRI consequently has the potential for characterizing the WEOM from the co-composting of PMD.

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1. Introduction

Penicillin mycelial dreg (PMD) that contains a high proportion of organic medium for *penicillium* growth and reproduction is a moisture-high (about 80%) bio-waste from the production of penicillin. Since the organic medium are highly unstable and also hold penicillin residue, improper disposal of these bio-waste materials will result in an environmentally negative impacts, such as water pollution, odors occurrence and resistant genes development (Hu et al., 2011; Wang et al., 2015). Composting has been considered as an economical and environment-friendly approach (Bernal et al., 2009; Grigatti et al., 2011; Hu et al., 2011). Composting is able to convert organic wastes into a product that is used as a nutrient source for plant growth or a soil amendment to improve its structure and the organic matter content (Hu et al., 2011). PMD containing a large amount of organic matters can be utilized as raw

Abbreviations: PMD, penicillin mycelial dreg; EEM, excitation-emission matrix; FRI, fluorescence regional integration; SFI, specific fluorescence intensity; OM, organic matter; VS, Volatile solid; WEOM, water extractable organic matter; DOC, dissolved organic carbon; DTN, dissolved total nitrogen; DON, dissolved organic nitrogen; SOUR, specific oxygen uptake rate; SS, sewage sludge; RS, rice straw; SD, sawdust.

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material for co-composting (Zhang et al., 2015). Therefore, its final compost can provide plenty of macro- and micronutrients for plant growth in agriculture or amendment of infertile soil (Grigatti et al., 2011).

The quality of compost mainly depends on the stability and maturity of organic matter (Bernal et al., 1998; Grigatti et al., 2011). Since biochemical transformation of organic matter via microbial metabolism always occurs in water soluble phase, water extractable organic matter (WEOM) may influence significantly the transformation of organic matter during composting (He et al., 2013; Said-Pullicino et al., 2007; Xing et al., 2012). WEOM that contains the most biologically and chemically active fraction in organic matter is defined as water soluble organic material passing through a 0.45 μm membrane filter. (Guo et al., 2012b; Traversa et al., 2010). It is considered as a good indicator of the transformation, stability and maturity of organic matter during composting or vermicomposting (He et al., 2013; Said-Pullicino et al., 2007; Xing et al., 2012; Yu et al., 2011). Thus, it is of significance to track change in WEOM for better understanding of composting process and effectively evaluating maturity of compost (Said-Pullicino et al., 2007).

Previous studies on composting mainly focused on process performance from different organic wastes (Grigatti et al., 2011), influential factors such as aeration rates, C/N ratio and moisture (Gao et al., 2010; Guo et al., 2012a), as well as heavy metal etc. (Singh and Kalamdhad, 2012). To date, although the characteristics of WEOM have been investigated during vermicomposting of sewage sludge and cow dung, during composting of cattle manure and during biostabilization of municipal solid waste (Guo et al., 2012b; Said-Pullicino et al., 2007; Shao et al., 2009; Xing et al., 2012), no report on characteristics of WEOM during co-composting of PMD has been published.

Due to the non-destructive, efficient and sensitive characteristics, fluorescence spectra of excitation-emission matrix (EEM) has received much attention on composting or vermicomposting (He et al., 2013; Wang et al., 2013; Xing et al., 2012; Yu et al., 2011). The fluorescence regional integration (FRI) of EEM spectra (EEM-FRI), a quantitative technique that integrates volumes beneath different excitation–emission regions within EEM spectra, has been used to qualify and quantify fluorescent organic compounds such as protein, humic and fulvic acids (Lv et al., 2013; Tian et al., 2012; Wang et al., 2013; Yu et al., 2011). Lv et al. (2013) used this method to investigate composition and transformation of the WEOM at different stages during vermicomposting of cattle dung. Tian et al. (2012) investigated spectral characteristics of WEOM during composting of dairy manure and rice chaff via the fluorescence EEM and drew a conclusion that EEM-FRI technique is suitable to characterize maturity of compost.

However, WEOM during co-composting of PMD via EEM-FRI analysis has not been previously characterized. Therefore, the objective of this study is to investigate the characteristics of WEOM extracted from different stages and to evaluate the maturity during co-composting of PMD with EEM-FRI.

2. Materials and methods

2.1. Source materials, co-composting process and sampling

PMD was from a biologically pharmaceutical industry (Harbin, China), and dewatered sewage sludge (SS), consisting of primary and secondary biosolids, was collected from a locally municipal wastewater treatment plant (Harbin, China). Sawdust (SD) and rice straw (RS) as bulking agents were obtained from a local wood processing industry and farmland in suburbs of Harbin. SD was characterized by powdery particles of sawn wood, the average diameter of about 0.50 mm. RS was manually cut into a length of 1.0–2.0 cm. Characteristics of these materials are shown in Table S1.

PMD, SS, RS and SD were mixed in a ratio of 2.0: 2.0: 1.0: 1.0 (w/w, wet weigh), and then were loaded into a composting reactor (100 cm height, 65 cm length and 60 cm width, about 390 L). Each mixture weighing about 72 kg was homogenously blended before co-composting. The process of co-composting lasted 51 d during which the mixture was turned and watered at about 60% on the 15th day, and this process was repeated in triplicate under aerobic conditions. The characteristics of the initial mixture are presented in Table S2.

Reactors were covered with styrene cystosepiment (foam board) of 2-cm thickness on their all sides and their top for thermal insulation. Three sampling ports (5-cm inter diameter) on the side of the reactor were set at equal intervals (30 cm). The central positions of three sampling ports were located at the heights of 17 cm, 47 cm and 77 cm from the bottom of the reactor, respectively (Fig. S1). The ports were sealed by rubber stoppers during co-composting. To ensure a uniform gas distribution, the matrix was supported by a stainless steel grid, with holes of diameter in 3 mm, installed at the bottom of the reactor and the aeration rate of about 0.52 L min^{-1} kg^{-1} OM was used.

A temperature Pt100 sensor (WRPX-12, Changjiang Temperature Meter Factory, Shanghai city, China) located at the 50-cm height from the bottom was connected to a digital thermometer (XMT, Changjiang Temperature Meter Factory, Yuyao city, Zhejiang in China) to automatically record the data.

One representative sample was obtained by mixing three subsamples corresponding to three sampling ports on the 1st, 2nd, 3rd, 4th, 5th, 6th, 8th, 12th, 18th, 24th, 32nd, 40th and 51st day. The sample was divided into two parts, one of which was immediately analyzed and the other air-dried, ground to pass through a 1-mm sieve and stored in a desiccator for further analysis.

2.2. Extraction and analysis of WEOM

The sample was shaken with deionized water (solid to water ratio of 1:20 w/v) for 24 h in a horizontal shaker at 25 °C, and then the WEOM was obtained by centrifugation at 8000 rpm for 10 min and filtration through a 0.45 mm membrane filter. The moisture was determined by drying fresh samples at 105 °C in an oven for 24 h. After the measurement of moisture, organic matter (OM) and ash of the dried samples were determined by burning them at 550 °C for 4 h in a muffle furnace (Hu et al., 2011). The concentration of DOC and the DTN in aqueous extract was determined by the TOC analyzer (SSM-5000A, Shimadzu, Japan). The concentration of dissolved organic nitrogen (DON) was calculated as the formula of ($DTN-NH_4^+-N-NO_3-N$). The concentrations of water-soluble NH_4^+–N and NO_3–N were determined according to the standard procedure (SEPA, 2002). The measurement of SOUR was performed as Lasaridi and Stentiford (1998).

2.3. Analytical method of penicillin

Solid phase extraction (SPE) was applied to the purification and pre-concentration of the extract liquid using a solid phase extraction column (Waters Oasis HLB, 3 cc/60mg) which was previously conditioned with 5 mL methanol and 5 mL ultrapure water. The extract liquid went through the SPE cartridge (Supelco VisiprepTM, USA) with a flow rate of 1.0 mL min^{-1}, and then the cartridge was rinsed with 5 mL of 5% methanol/water solution, followed by being eluted with the extraction buffer solution of 5 mL. The eluent collected was concentrated to near dryness under a gentle stream of N_2 gas at 25 °C, and then reconstituted with the injection solution of 1 mL (50% acetonitrile in ultrapure water) followed by

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