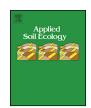
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## **Applied Soil Ecology**

journal homepage: www.elsevier.com/locate/apsoil



## Influence of buffalo manure, compost, vermicompost and biochar amendments on bacterial and viral communities in soil and adjacent aquatic systems



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#### ARTICLE INFO

#### Article history: Received 26 March 2013 Received in revised form 20 August 2013 Accepted 30 August 2013

Keywords: Organic fertilization Soil Water Mesocosms Bacteria

#### ABSTRACT

Vermicompost and biochar amendments are management practices which may contribute to sustainable agroecosystems by reducing dependence on inorganic fertilizers. However, little is known about their impacts on soil microorganisms and their transfer and evolution in connected aquatic systems. The aim of this study was to determine the influence of organic manure (buffalo manure, compost or vermicompost) and biochar amendments on bacterial and viral properties in soil and water. A three year experiment was carried out with terrestrial mesocosms which were used to test the effect of organic matter amendment on maize growth. In the last year of the experiment, runoff and infiltration waters from the terrestrial mesocosms were transferred to aquatic mesocosms. Organic fertilization improved soil properties (higher C, N content and  $pH_{H_2O}$ ) and as a consequence increased soil bacterial and viral abundance. Bacterial diversity (Shannon 'H' and richness 'S' indices calculated from DGGE fingerprint) was also enhanced after the continuous application of organic amendments. Compared with compost, vermicompost reduced viral abundance and S but similar H and bacterial abundance were observed. The pH<sub>H2O</sub>, C content and bacterial and viral abundance increased in the aquatic mesocosms following organic fertilization. As a consequence, bacterial and viral diversity also increased in the water, although no differences were found between compost and vermicompost. Biochar increased soil bacterial abundance for the mineral fertilizer treatment but did not influence bacterial and viral abundance in water. However, the combination of biochar and vermicompost led to an increase of viruses in soil and a reduction of bacteria in water. Similarity dendrograms from the DGGE banding patterns showed that the structure of bacterial communities was mainly influenced by the fertilizer treatments in soil but by the presence of biochar in water. In conclusion, this study demonstrated that the nature of the organic amendment has important consequences on both soil and water microbial abundance and diversity.

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#### 1. Introduction

Land use planning and management practices appear to be important factors influencing soil organic matter (OM) turnover, soil erosion and the transfer of nutrients into aquatic systems

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(Lal, 2005). These practices are also important factors controlling the abundance and diversity of microorganisms in soil (Dequiedt et al., 2009; Ranjard et al., 2010). Although their influence on the transfer of soil microorganisms to aquatic systems remains poorly documented, recent research showed that when rainfall generates runoff and leaching from agricultural land, high concentrations of bacteria can be transferred from the soil to the water system (Oliver et al., 2007). In both soil and water, bacteria play a key role in the decomposition of OM and in the functioning of trophic food webs (Azam and Worden, 2004). Bacteria are accompanied by a cortege of highly abundant viral parasites (Weinbauer and Rassoulzadegan,

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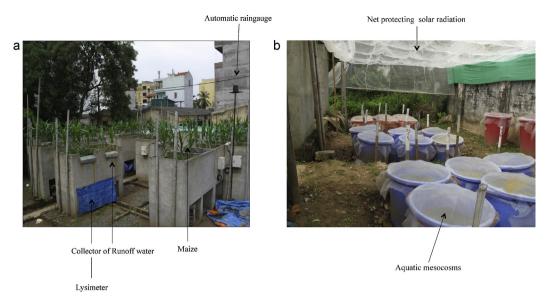


Fig. 1. Photos showing the set-up of the terrestrial (a) and aquatic (b) mesocosms.

2004; Suttle, 2005; Williamson et al., 2005; Bouvier and Del Giorgio, 2007). Although viruses are assumed to be important agents of microbial mortality in aquatic systems and to play a key role in the microbial loop (Weinbauer, 2004) much less is known about their ecology in the soil system (Kimura et al., 2008). The soil environment is a more diverse habitat for microorganisms than aquatic environments and the activity, diversity and abundance of microorganisms differ in each system (Kimura et al., 2008). How terrestrial microorganisms evolve in aquatic systems remains totally unexplored. It is likely that their transfer, as well as the interstitial water, influence the abundance and diversity of local indigenous populations, and as a consequence largely modify ecosystem functioning (Bardgett et al., 2001).

Among promising agricultural approaches, vermicompost and the incorporation of biochar into soils are currently being promoted to improve soil quality, reduce water and fertilizer needs and therefore increase the sustainability of agricultural practices in tropical countries. Vermicomposting is a process which stabilizes OM under aerobic and mesophilic conditions through the joint action of earthworms and microorganisms. The products of vermicomposting have been successfully used to suppress plant pests and disease as well as increase crop productivity (Edwards et al., 2004; Arancon et al., 2005a,b; Gutiérrez-Miceli et al., 2007). Vermicompost is also characterized by specific microbial communities compared with the microbial communities in initial and composted substrates (e.g., Fracchia et al., 2006; Yasir et al., 2009; Fernández-Gómez et al., 2011, 2012). In recent years, numerous studies have also investigated the utilization of biochar for the management of soil fertility. Biochar is a highly condensed material generated from the incomplete pyrolysis of OM (Rovira et al., 2009; Lee et al., 2010). The incorporation of biochar into soil influences soil structure, increases nutrient retention, decreases N2O and CH4 emissions, reduces leaching of inorganic N and enhances plant growth (Steiner et al., 2007; reviewed by Lehmann et al., 2003b; Blackwell et al., 2009). The molecular structure of biochar shows a high degree of chemical and microbial stability (Atkinson et al., 2010). As a consequence, biochar incorporation usually leads to a modification in the abundance and diversity of bacteria in soil (Kim and Jung, 2007; Atkinson et al., 2010; Anderson et al., 2011; Khodadad et al., 2011).

To date, there are no reports on the potential combined effects of vermicompost and biochar on the transfer of soil microbial communities into aquatic systems and the development of these soil communities in water. Because nutrient cycling is mainly under the regulation of bacteria and viruses in marine and freshwater environments (Suttle, 2005), this study examined the influence of vermicompost and biochar on the diversity of these two biotic compartments in soil and water.

#### 2. Material and methodology

#### 2.1. Experimental design

The experiment was carried out outdoors at the Soil and Fertilizer Research Institute (SFRI), in Hanoi, Vietnam. The experimental design consisted of terrestrial mesocosms where maize was cultivated for three years and aquatic mesocosms which received runoff and leachate collected from the bottom of the terrestrial mesocosms during the final year of the experiment (Fig. 1). The aim of this simplified system was to represent the link between uplands where land use takes place (the terrestrial mesocosms) and reservoirs in the lowlands which receive runoff and infiltration water from the uplands.

The terrestrial mesocosms consisted of tanks  $(1 \text{ m} \times 1 \text{ m})$  and 2 m high) filled with 1 m<sup>3</sup> of soil. These tanks were equipped with runoff water collectors. The bottom of the tank was on a gentle slope (5°) ending with a lysimeter to allow infiltration water to be recovered. A thin layer of stones, previously cleaned with deionised water, was used to cover the bottom to retain and stop soil from being exported along with the infiltration water. The mesocosms were then filled with soil sampled from the 0-30 cm surface layer at the MSEC experimental catchment (Dong Cao village, Tien Xuan Commune, Hanoi). The soil was first sieved at 2 cm to discard stones and litter residue. It is an Acrisol (World Reference Base for Soil Resources, 1998) or Ultisols (Soil Survey Staff, 1999) with low organic matter (OC = 0.31%) and nitrogen content (N=0.02%). The soil pH is acid (p $H_{H_2O}=5.3$ ) and contains ~43% sand, 34.28% silt, and 22.3% clay. Four maize seedlings (variety NK4300) were grown per terrestrial mesocosm without irrigation for three growing seasons but with a minimum of chemical fertilizers to allow plant yield. Plants were treated with four different types of fertilization regimes: only chemical nutrients (mineral) or the same amount of chemical nutrients plus one of the three types of organic fertilizer: 2 kg of buffalo manure, compost or vermicompost mixed with the soil (n = 5 for all treatments). The influence of the incorporation of biochar (700 g) was also tested for the mineral (i.e., only chemical nutrients with or without biochar) and

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