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# Relationships between soil physico-chemical, chemical and biological properties in a soil amended with spent mushroom substrate

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#### ABSTRACT

The effects of two different spent mushroom substrates on soil physico-chemical, chemical and biological properties were studied in a plot experiment with a horticultural crop (Lactuca sativa L. var. linus). Two organic treatments were applied: spent mushroom substrate from Agaricus bisporus crop (T1) and a mixture of spent mushroom substrate from A. bisporus crop and spent mushroom substrate from Pleurotus crop (50% (v/v)) (T2), both treatments providing 100 kg ha<sup>-1</sup> of nitrogen. The unamended soil was used as control treatment (C). pH, electrical conductivity (EC), oxidisable organic C (oxidisable OC), available P, organic N, NH<sub>4</sub><sup>+</sup>-N, NO<sub>3</sub><sup>-</sup>-N, N loss, soil respiration and enzymatic activities (catalase, urease and phosphatase) were measured during 126 days after spent mushroom substrate addition to the soil. The organic amendment, particularly T1 treatment, increased the oxidisable OC, organic N and available P content of the soil. The application of spent mushroom substrates did not produce great changes on soil physico-chemical properties (pH and EC) with respect to the control soil. Organic N content in the amended soils increased at the beginning of the experiment, coinciding with the period in which N losses were positive (N gain). In all soils, NH<sub>4</sub><sup>+</sup>-N concentration increased throughout the first two weeks of experiment and then these parameters decreased to the end of the experiment. However, the evolution of the NO<sub>3</sub><sup>-</sup>-N content tended to decrease in all soils throughout the experiment, in the amended soil observing an initial nitrification inhibition. Finally, the addition of spent mushroom substrates increased soil respiration rate and phosphatase activity, not producing great differences in catalase and urease activities with the incorporation of these wastes into soil.

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#### 1. Introduction

The mushroom industry, especially in Spain where mushroom production represents about 12% of the entire production in the European Union (EU) (Food and Agriculture Organisation, 2010), produces increasing quantities of spent mushroom substrates. About 5 kg of these waste substrates is produced for each kilogram of mushrooms (Williams et al., 2001), so in the last year the spent mushroom substrate production in Spain was approximately 660,000 t (Food and Agriculture Organisation, 2010). The mushroom industry generates two main types of spent mushroom substrate, one for *Agaricus bisporus* (spent mushroom substrate-AB) and other for *Pleurotus* (spent mushroom substrate-P). Spent mushroom substrate-AB is composed of a composted mixture of cereal straw and manure (poultry and/or horse manure and/or pig slurry), calcium sulphate, soil and residues of inorganic nutrients and pesticides, whereas spent mushroom substrate-P contains fermented cereal straw and residues of inorganic nutrients and pesticides (Paredes et al., 2006).

Soil application is a promising strategy for the sustainable recycling of the spent mushroom substrates. In fact, the spent mushroom substrate-AB has similar organic matter content, similar or even higher macronutrient concentrations, especially in the case of Ca, and lower micronutrient contents in comparison with other organic fertilisers, such as urban wastes (Pascual et al., 1997) and animal manure (Moreno-Caselles et al., 2002). Also, spent mushroom substrate-P contains higher organic matter content and lower macro and micronutrient concentrations compared to those of manures and urban wastes used as organic fertilisers (Paredes et al., 2006). The effectiveness of spent mushroom substrate as an organic amendment has been positively evaluated by Maher (1994), Jordan et al. (2008), Morlat and Chaussod (2008), Ribas et al. (2009) and Courtney et al. (2009). However, the high contents of mineral salts of the spent mushroom substrate-AB could limit its use for growing of salt sensitive plants (Medina et al., 2009) and its application to land in sensitive areas (Paredes et al., 2006). For this reason, passive leaching by rainfall and snowmelt is a method to treat spent mushroom substrates before its use (Guo and Chorover, 2006). Also, different authors have found an initial net immobilisation of nitrogen (Stewart et al., 1998a) or a nitrification inhibition (Maher, 1994; Stewart et al., 1998b), when these waste substrates were used as organic amendments. So, the mineralisation dynamics of spent mushroom





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substrates in soil needs to be thoroughly evaluated since it is the main process regulating nutrient availability. However, very few studies are reported in the literature dealing with this topic (Ribas et al., 2009; Stewart et al., 1998a, 1998b).

On the other hand, soil microbiological parameters can provide valuable information about the changes in the soil properties and these parameters are simple, rapid and effective indicators of modification in soil status due to pollution and variations in soil management (Brookes, 1995). However, the use of biological indicators has the problem of not knowing which indicator responds to a specific soil treatment or contaminant. So, the use of multiple biological and biochemical properties is often suggested (Ros et al., 2006). For instance, soil respiration rate and enzyme activities related to the cycle of main nutritive elements could give indications on the rates of substrate turnover, soil metabolic potential, as well as to the resilience of the soil when subjected to various natural and antropogenic impacts (Insam et al., 1991; Shaw and Burns, 2006).

Therefore, the main aim of this work was to study, under field conditions and during crop growth, the effects of applying spent mushroom substrates with respect to an unamended soil used as a reference, on several soil physico-chemical, biological and biochemical properties in order to evaluate the soil fertility improvement and C and N mineralisation dynamics.

#### 2. Materials and methods

#### 2.1. Characteristics of the spent mushroom substrates

The spent mushroom substrates used in the present study were obtained from a composting facility located in the city of Quintanar del Rey (Cuenca), Spain, which manages the organic wastes produced from the mushroom industry of Castilla-La Mancha community. This community is one of the main mushroom producing regions in Spain. The main characteristics of the spent mushroom substrate used are shown in Table 1. In this study, the spent mushroom substrate-P was mixed with the spent mushroom substrate-AB in the proportion 50% (v/v) to improve its nitrogen content, so a similar dose of amendment in both organic treatments was applied.

#### 2.2. Study site and soil sampling

The experiment was conducted from October 2006 to February 2007 in the Segura river valley, in the Southeast of Spain, at the Research Station of the Miguel Hernandez University (Orihuela-Alicante) ( $38^{\circ}4'0''N$ ,  $0^{\circ}58'0''W$  and elevation 24 m above sea level). The climate of this

#### Table 1

Physico-chemical properties and chemical composition of the spent mushroom substrates.

Parameter <sup>a</sup>	T1 <sup>b</sup>	T2 <sup>c</sup>
рН	7.98	8.25
Electrical conductivity (dS m <sup>-1</sup> )	7.47	5.88
Total organic C (g kg <sup>-1</sup> )	273	351
Total N (g kg $^{-1}$ )	22.2	17.9
$NH_4^+$ -N (mg kg <sup>-1</sup> )	327	186
$NO_{3}^{-}-N (mg kg^{-1})$	81	53
P (g kg <sup>-1</sup> )	6.80	3.74
$K (g kg^{-1})$	26.2	20.1
Ca (g kg <sup>-1</sup> )	89.3	51.6
Mg (g kg <sup><math>-1</math></sup> )	4.81	3.45
Na (g kg <sup>-1</sup> )	2.97	2.11
Fe (mg kg <sup><math>-1</math></sup> )	4527	2586
Cu (mg kg <sup><math>-1</math></sup> )	38	22
$Mn (mg kg^{-1})$	320	185
$Zn (mg kg^{-1})$	170	91

<sup>a</sup> Values on a dry matter basis.

<sup>b</sup> T1: spent mushroom substrate-Agaricus bisporus.

<sup>c</sup> T2: mixture of spent mushroom substrate-*Agaricus bisporus* and spent mushroom substrate-*Pleurotus* 50% (v/v).

region is semi-arid subtropical Mediterranean, with an average cumulative annual precipitation of 271 mm and an average annual temperature of 17.9 °C. The mean fortnightly temperature and rainfall during the experimental period are given in Fig. 1 (MARM, 2011a). The soil of this area is classified as a Xerofluvent (Soil Survey Staff, 2006) with a clayey-loam texture, an alkaline nature and low salinity and organic C content. The main characteristics of the soil are shown in Table 2.

Three treatments, in a completely-randomised design with three replicates per treatment, were set up in experimental plots of 6 m<sup>2</sup> each. The treatments were: control without amendment (C), spent mushroom substrate-AB (T1) (77 t  $ha^{-1}$ ) and the mixture of spent mushroom substrate-AB and spent mushroom substrate-P 50% (v/v) (T2) (85 t  $ha^{-1}$ ), both organic treatments providing 100 kg  $ha^{-1}$  of nitrogen. This nitrogen level was adequate for the nitrogen requirement of the crop selected (lettuce). The treatments were uniformly applied and immediately incorporated to a soil depth of 30 cm by light rototilling. The organic wastes were applied to the soil one month prior to the planting. Lettuce (Lactuca sativa L. var. linus) seedlings of uniform size were selected and thirty-six seedlings were planted in each plot (60,000 plant ha<sup>-1</sup>). Three irrigations with tap water were used during growing season (ninety-eight days), on days 28, 63 and 84. Treatments of weed-killer, insecticide and fungicide were not applied throughout experimental period. Soil samples were collected after 0, 7, 14, 21, 42, 63, 84, 112 and 126 days of the experiment. All soil samples were taken by mixing six sub-samples from six sites of each plot at 0-20 cm depth. Each soil sample was sieved to 2 mm, after removal of vegetation, bigger roots and stones, and the granulometric fraction was divided into two sub-samples: one was stored at 4 °C for enzymatic activity and soil respiration determinations, while the other was air-dried and used for the rest of the measurements.

#### 2.3. Analytical methods

The pH and electrical conductivity (EC) of the soil samples were measured on a 1:2.5 and 1:5 soil:water (w/v) ratio, respectively (Allison and Moodie, 1965). Active calcium carbonate in soil was measured by titration of 0.2 N ammonium oxalate extract (1:100 w/v)with 0.1 N KMnO<sub>4</sub> (Allison and Moodie, 1965). Soil texture was measured by the Bouyoucos densimeter method and oxidisable organic C (oxidisable OC) by the Walkley and Black modified method (Yeomans and Bremner, 1989). Soil respiration rate was determined by titration with 0.1 M HCl after trapping CO<sub>2</sub> in 0.1 M NaOH and adding BaCl<sub>2</sub> (Stotzky, 1965). Carbon mineralisation of added spent mushroom substrates was evaluated by calculating the total amount of extra cumulative CO<sub>2</sub>-C evolved in the different treatments (cumulative CO<sub>2</sub>-C evolved from amended soil minus cumulative CO2-C evolved from control) (Bustamante et al., 2007). NH<sub>4</sub><sup>+</sup>-N was measured in the 2 M KCl extract (1:10 w/v) by the indophenol blue method (Dorich and Nelson, 1983; Keeney and Nelson, 1982). NO<sub>3</sub>-N was determined in the

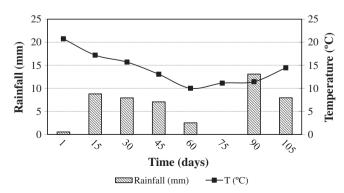


Fig. 1. Mean fortnightly temperature and rainfall of the experimental site during the experimental period (October 2006–February 2007).

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