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Soil enzyme activities, microbial community composition and function after 47 years of continuous green manuring

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Abstract

Green manuring practices can influence soil microbial community composition and function and there is a need to investigate the influence compared with other types of organic amendment. This study reports long-term effects of green manure amendments on soil microbial properties, based on a field experiment started in 1956. In the experiment, various organic amendments, including green manure, have been applied at a rate of 4 t C ha^{-1} every second year. Phospholipid fatty acid analysis (PLFA) indicated that the biomass of bacteria, fungi and total microbial biomass, but not arbuscular mycorrhizal (AM) fungi, generally increased due to green manuring compared with soils receiving no organic amendments. Some differences in abundance of different microbial groups were also found compared with other organic amendments (farmyard manure and sawdust) such as a higher fungal biomass and consequently a higher fungal/bacterial ratio compared with amendment with farmyard manure. The microbial community composition (PLFA profile) in the green manure treatment differed from the other treatments, but there was no effect on microbial substrate-utilization potential, determined using the Biolog EcoPlate. Protease and arylsulphatase activities in the green manure treatment were comparable to a mineral fertilized treatment receiving no additional C, whereas acid phosphatase activity increased. It can be concluded that green manuring had a beneficial impact on soil microbial properties, but differed in some aspects to other organic amendments which might be attributed to differences in quality of the amendments.

Keywords: Biolog; Farmyard manure; Long-term field experiment; Organic amendment; Phospholipid fatty acid; Soil C

1. Introduction

Green manuring is considered an important management practice with potential to reduce the dependence on mineral fertilizers and to maintain soil organic matter content. As other organic amendments, green manure provides nutrients for plant growth and the organic carbon serves as an energy supply for heterotrophic soil microorganisms. Green manure commonly replaces farmyard manure amendments on stockless farms and a question of great concern is whether green manure can be considered to be equal to farmyard manure as a carbon source for improving and sustaining soil biological properties and fertility. Inputs of green manure or crop residues can increase the size and activity of soil microbial communities (Bolton et al., 1985; Martens et al., 1992; Kirchner et al., 1993; Fauci and Dick, 1994; Kautz et al., 2004; Manici et al., 2004), but the impact on soil microbial community composition and function is less well understood. Green manuring or crop residue amendments have caused shifts in microbial community composition (Buyer and Drinkwater, 1997; Schutter and Dick, 2001), increased soil bacterial diversity (Sessitsch et al., 2001) and influenced microbial carbon source

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utilization profiles (Buyer and Drinkwater, 1997; Lupwayi et al., 1998; Schutter and Dick, 2001). But other studies have failed to detect any community changes (Wander et al., 1995). The quality of the amendments, such as C/N ratio, can influence the decomposer community composition. Fungi have been shown to be the most important degraders of high C/N ratio straw in arable soil (Cheshire et al., 1999) and bacteria the main contributors to decomposition of low C/N ratio rye shoots (Lundquist et al., 1999). In contrast to this, Marschner et al. (2003) found increased bacterial/fungal ratios due to long-term straw amendment. Contrasting results have thus been obtained, but it appears that the use of green manure might alter microbial community composition and function. However, it is not clear how the impact of green manure differs compared with other organic fertilizers. In addition, most studies have reported on short-term responses, which may differ considerably from long-term responses to different fertilizer regimes.

In order to evaluate how green manure influences soil biological properties and if it can replace farmyard manure, we used the Ultuna Long-term Soil Organic Matter Experiment at the Swedish University of Agricultural Sciences. This experiment was established in 1956 in order to study the effect of different nitrogen sources on crop yield and soil organic matter development (Kirchmann et al., 1994) and provides an excellent opportunity to study the long-term influence of different fertilizer regimes on soil microbial communities. Previous research on soil from this experiment has shown increasing microbial biomass with increasing amounts of C input (Schnüer et al., 1985; Witter et al., 1993; Witter and Kanal, 1998). One study has shown increased bacterial diversity in manured treatments (green manure and farmyard manure) compared with treatments without organic amendments, and a particularly high abundance of some Gram-positive bacterial species in the green manure treatment (Sessitsch et al., 2001). In the present study, the analysis of the effects of organic amendments was extended by investigating the impact on (1) microbial biomass and community composition through analysis of phospholipid fatty acid profiles (PLFA); (2) activities of the enzymes protease, acid phosphatase and arylsulphatase and (3) microbial substrate-utilization potential as determined with the Biolog EcoPlate. The methods were chosen to characterize both structural (PLFA) and functional (Biolog) aspects of soil microbial communities. The enzymes were chosen due to their importance in the cycling of N (protease), P (phosphatase) and S (arylsulphatase). The specific question posed was: How does almost 50 years of green manure amendment affect soil microbial community composition, function and soil enzyme activities compared with other types of carbon amendment?

2. Materials and methods

2.1. Site description and experimental design

The Ultuna Long-Term Soil Organic Matter Experiment is located in central Sweden (59°48′N, 17°39′E). The soil consists of 36.5% clay, 41.0% silt and 22.5% sand and has been classified as a Typic Eutrochrept or Eutric Cambisol (Kirchmann et al., 1994). Since the start of the experiment in 1956, cereals have dominated the crop rotation. Maize (*Zea mays* L. cv. Loft), which was grown in the year of sampling, has been grown continuously since 2000.

Fresh grass, harvested on a clover-free ley and incorporated into the soil by hand, was initially used as a green manure amendment, starting in 1956. The same procedure is followed today except that dried grass (hay) is used. Five complementary treatments were selected to represent soils receiving either a different quantity of C compared with the green manure treatment or an equal amount of C but of different quality than the green manure. The treatments were; unfertilized, inorganic N, farmyard manure (FYM) and sawdust (coniferous) + inorganic N (Table 1). The

Table 1
Total C, N, P and S added with inorganic and organic fertilizers in the respective treatments of the Ultuna Long-term Soil Organic Matter Experiment (average for the years 1989–1999). Cellulose, hemicellulose and lignin added with the organic fertilizers in 2001 and C/N ratio (average 1989–1999)

Treatment	C	N	P	S	Hemicellulose (kg ha^{-1} yr^{-1})	Cellulose	Lignin	C/N
Green manure	2052	65	32	33	1341	1307	173	26
Unfertilized	ND	ND	ND	ND	ND	ND	ND	ND
$Ca(NO_3)_2$	ND	80	20	27	ND	ND	ND	ND
Farmyard manure	2212	104	56	49	1180	871	595	18
Sawdust + $Ca(NO_3)_2$	2054	81	20	28	697	2446	774	1205

ND: not determined.