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Interactions between phosphorus availability and an AM fungus (*Glomus intraradices*) and their effects on soil microbial respiration, biomass and enzyme activities in a calcareous soil

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Summary

The interactions between soil P availability and mycorrhizal fungi could potentially impact the activity of soil microorganisms and enzymes involved in nutrient turnover and cycling, and subsequent plant growth. However, much remains to be known of the possible interactions among phosphorus availability and mycorrhizal fungi in the rhizosphere of berseem clover (*Trifolium alexandrinum* L.) grown in calcareous soils deficient in available P. The primary purpose of this study was to look at the interaction between P availability and an arbuscular mycorrhizal (AM) fungus (*Glomus intraradices*) on the growth of berseem clover and on soil microbial activity associated with plant growth. Berseem clover was grown in P unfertilized soil (−P) and P fertilized soil (+P), inoculated (+M) and non-inoculated (−M) with the mycorrhizal fungus for 70 days under greenhouse conditions. We found an increased biomass production of shoot and root for AM fungus-inoculated berseem relative to uninoculated berseem grown at low P levels. AM fungus inoculation led to an improvement of P and N uptake. Soil respiration (SR) responded positively to P addition, but negatively to AM fungus inoculation, suggesting that P limitation may be responsible for stimulating effects on microbial activity by P fertilization. Results showed decreases in microbial respiration and biomass C in mycorrhizal treatments, implying that reduced availability of C may account for the suppressive effects of AM fungus inoculation on microbial activity. However, both AM fungus inoculation and P fertilization affected neither substrate-induced respiration (SIR) nor microbial metabolic quotients (qCO_2). So, both P and C availability may concurrently limit the microbial activity in these calcareous P-fixing soils. On the contrary, the activities of alkaline phosphatase (ALP) and acid phosphatase (ACP) enzymes responded negatively to P addition, but positively to AM fungus inoculation, indicating that

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AM fungus may only contribute to plant P nutrition without a significant contribution from the total microbial activity in the rhizosphere. Therefore, the contrasting effects of P and AM fungus on the soil microbial activity and biomass C and enzymes may have a positive or negative feedback to C dynamics and decomposition, and subsequently to nutrient cycling in these calcareous soils. In conclusion, soil microbial activity depended on the addition of P and/or the presence of AM fungus, which could affect either P or C availability.

Introduction

Soil microbial activity including soil respiration (SR) and enzyme activities, and the size of microbial biomass, have been shown to depend on P fertilization and the presence of AM fungi in the soil–plant system (Amador and Jones, 1993; Wright and Reddy, 2001; Wamberg et al., 2003; López-Gutiérrez et al., 2004; Baligar et al., 2005; Marschner and Timonen, 2006). Phosphorus fertilization may affect soil microbial respiration and biomass, especially soil enzymes, with variable results depending on the soil P status. Phosphorus additions resulted in increased microbial respiration in soils with low P contents, but not in soils with high P contents (Amador and Jones, 1993; Smith, 2005). In contrast, P fertilization had an inhibitory effect on microbial respiration and substrate-induced respiration (SIR) in a pine forest floor, while no effects on microbial metabolic quotients (qCO_2) were detected (Thirukkumaran and Parkinson, 2000). Increasing levels of soil applied P significantly reduced acid phosphatase (ACP) activities and resulted in lower arylsulfatase and urease activities in acidic infertile upland soils under white clover cover (Baligar et al., 2005). Similarly, application of phosphate decreased activities of phosphatase, sulfatase, and urease (Haynes and Swift, 1988). Higher P concentrations may also depress the activity of some soil enzymes under natural conditions. In a wetland soil, P loading negatively influenced only the activity of alkaline phosphatase (ALP) while other soil enzymes remained unaffected (Wright and Reddy, 2001). The association of plants with mycorrhizal fungi can have strong influences on the responses of microbial activities to P fertilization, due to the fact that these fungi are able to enhance P availability to and uptake by plants (Smith and Read, 1997). Following inoculation, AM fungi may further influence microbial population and activity, and consequently nutrient dynamics in the soil through the release of organic compounds. There are many positive or negative interactions between AM fungus and soil microorganisms (see reviews by Bonkowski et al., 2000; Hodge, 2000; Johansson et al., 2004; Jones et al., 2004). Various studies indicated that AM fungus may alter the population composition (Bansal and Mukerji, 1994; Andrade et al., 1997; Vázquez et al., 2000) and activity of soil microorganism (Wamberg et al., 2003; Langley et al., 2005), likely due to quantitative and qualitative changes in root exudation of colonized plants occurring in the rhizosphere (Hodge, 2000; Barea et al., 2002). For example, root colonization by AM fungi reduced the exudation of sugars, amino acids and other organic compounds from the roots (Graham et al., 1981; Schwab et al., 1984; Bansal and Mukerji, 1994).

SR is one of the least studied microbial processes associated with AM fungi. Yet, the influence of mycorrhizal inoculation on SR is variable, and depends on experimental conditions and the methodology involved. In the rhizosphere of pea plants (*Pisum sativum*) inoculated with *Glomus intraradices*, SR was negatively or positively affected by fungi, depending on the growth stage of the plant (Wamberg et al., 2003). The negative effect was apparently due to the change in carbon flow from plant to fungal hyphae and therefore higher anabolism occurred by fungus (Wamberg et al., 2003). Mycorrhizal inoculation increased microbial SR in sunflower rhizosphere before plant maturity, due to the mycorrhizal stimulation of plant growth (Langley et al., 2005). However, when shoots were removed, mycorrihzal sunflower had lower SR than the corresponding non-mycorrhizal plants. AM fungi may also influence other microbial properties of the soil. Van Aarle et al. (2003) reported an increased microbial biomass and bacterial activity in the presence of AM fungal hyphae in a limestone soil. In contrast, Kim et al. (1998) showed that inoculation of tomato with *Glomus etunicatum* had no effects on total soil microbial biomass C (MBC). In the Entisols and Vertisols under savannas, lower P mineralization and higher microbial immobilization were associated with higher AM fungus colonization (López-Gutiérrez et al., 2004). Therefore, AM fungi may directly (Zhu and Miller, 2003) or indirectly (Langley