



Rhizosphere effect of three plant species of environment under periglacial conditions (Majella Massif, central Italy)



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ABSTRACT

The chemical, physical and biological processes occurring in the rhizosphere can influence plant growth by modifying root associated microorganisms and nutrient cycles. Although rhizosphere has been widely investigated, little is known about the rhizosphere effect of pioneer plants in soils of periglacial environments. The knowledge of the processes controlling soil–plant relationships in these severe environments may help understanding the ecological evolution of newly deglaciated surfaces. We selected three plants [*Helianthemum nummularium* (L.) Mill. subsp. *grandiflorum* (Scop.), *Dryas octopetala* (L.), and *Silene acaulis* (L.) Jacq. subsp. *cenisia* (Vierh.) P. Fourn.] that sparsely occupy deglaciated areas of central Apennines (Italy), with the aim to assess changes between rhizosphere and bulk soil in terms of physical, chemical, and biological properties. The three plants considered showed to have different rhizosphere effect. *Helianthemum* induced a strong rhizosphere effect through a synergistic effect between root activity and a well adapted rhizosphere microbial community. *Dryas* did not foster a microbial community structure specifically designed for its rhizosphere, but consumes most of the energetic resources supplied by the plant to make nutrients available. Conversely to the other two species, *Silene* produced slight soil changes in the rhizosphere, where the microbial community had a structure, abundance and activity similar to those of the bulk soil. The ability to colonize harsh environments of *Silene* is probably linked to the shape and functions of its canopy rather than to a functional rhizosphere effect.

This study showed that the rhizosphere effect differed by species also under high environmental pressure (periglacial conditions, poorly developed soil), and the activity of roots and associated microbial community is decisive in modifying the soil properties, so to create a suitable environment where plants are able to grow.

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1. Introduction

In the rhizosphere, the soil in proximity to the root, processes like rhizodeposition, intense microbial activity, root nutrient uptake, redox reactions, and CO₂ production induce modifications of soil components and properties (Hinsinger et al., 2003). The chemical, physical and biological differentiation of the rhizospheric soil with respect to the rest of the soil is called “rhizosphere effect”, which has been investigated in many ecosystems, including those with environmental constraints and nutrient-poor soils (e.g.,

Hinsinger et al., 2005; Teixeira et al., 2010). However, little is known about the rhizosphere effect of pioneer plants in young and poorly developed soils from periglacial environments (Wookey et al., 2009).

Periglacial environments are those affected by severe frost action that dominates geomorphic processes, and amount to about 25% of the Earth's land surface. The knowledge of the rhizosphere effect of pioneer plants in these environments is the basis in understanding how soil–plant relationships respond to environmental constraints. In general, arctic and alpine plants have a higher proportion of their biomass below-ground than trees and bushes from other ecosystems (Jackson et al., 1996; Körner, 2003), and this relatively high below-ground biomass increases the proportion of rhizosphere soil (Hinsinger et al., 2005; Finzi et al., 2015). Indeed, in poorly developed soils of cold areas, the presence of

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vascular plants strongly modifies the soil properties and the structure and function of the soil microbial community (Yergeau et al., 2007). In this areas, rhizospheric processes resulting from soil–plant–microbes interactions may improve the ability of plants to overcome abiotic disturbances such as freezing, high soil daily and seasonal temperature excursions, freeze–thaw and wet–dry cycles, excessive drainage, and strongly oligotrophic conditions (e.g., Tscherko et al., 2004, 2005; Edwards et al., 2006; Ciccazzo et al., 2014). The amount of energy supplied by the plants in form of exudates to rhizosphere heterotrophic microbial community is key for stimulating rhizospheric processes (Kuzakov, 2002; Wookey et al., 2009; Ciccazzo et al., 2014; Jorquera et al., 2014). In fact, most arctic and alpine vascular plants allocate 10–30% of net carbon fixation to establish mycorrhizal associations (Read et al., 2004; Cripps and Eddington, 2005), although allocation patterns of these energetic resources depend on plant species and soil nutrient availability (Högberg et al., 2003; Wardle et al., 2004; Wookey et al., 2009). Hence, different plants colonizing the same soil might differently shape a specific rhizosphere microbial community depending on the quantity and quality of their root exudates (Haichar et al., 2008; Huang et al., 2014).

Our research focuses on the rhizosphere effect of three plant species [*Helianthemum nummularium* (L.) Mill. subsp. *grandiflorum* (Scop.) Sch. and Th., *Dryas octopetala* (L.), and *Silene acaulis* (L.) Jacq. subsp. *cenisia* (Vierh.) P. Fourn.] that sparsely occupy soils of deglaciated areas actually submitted to periglacial conditions (central Apennines, Italy). These soils are characterized by environmental constraints such as harsh climatic and nutritional conditions. Specifically, we tested the following hypotheses:

- (i) the physical and chemical soil properties differ in rhizosphere versus bulk soil for the three plant species;
- (ii) the microbial community structure and abundance, and microbial respiration differ in rhizosphere versus bulk soil within and among the three plant species.

To this aim, we investigated physical, chemical and microbiological properties of both rhizosphere and bulk soil, and the results were compared with those of the adjacent bare soil.

2. Materials and methods

2.1. Site description

The study site is located in one of the highest mountains of central Apennines (Italy), the Majella massif (Fig. 1) and, in particular, in the Cannella Valley, whose altitude ranges from 1900 to 2750 m, and has a southeast orientation. The mean annual precipitation is about 2100 mm (mostly snow) and the mean annual air temperature is 2.3 °C. January is the coldest month, with an average temperature of −4.3 °C, whereas August is the warmest month, with an average temperature of 11.4 °C (Corti et al., 2012). The area, that experienced a relatively recent glacier recession initiated about 12,700 and ended about 11,000 years before present (Giraudi, 2004), is mantled by thick morainic deposits (till) mostly made of limestone, from which the present soils developed. The area is covered by sparse vegetation mostly made of *H. nummularium* (L.) Mill. subsp. *grandiflorum* (Scop.) Sch. and Th., *D. octopetala* L., *S. acaulis* (L.) Jacq. subsp. *cenisia* (Vierh.) P. Fourn., *Carex kitaibeliana* Degen ex Bech., *Anthyllis vulneraria* L. subsp. *maura* (Beck) Maire, *Campanula scheuchzeri* Vill., *Minuartia verna* (L.) Hiern subsp. *verna*, *Trifolium pratense* L. subsp. *semipurpureum* (Strobl) Pign., with spots covered by *Salix retusa* L. and rare dwarf mountain pines (*Pinus mugo* Turra). Where the vegetation forms a rather continuous mat, the soils are loamy-skeletal, mixed, frigid Oxyaquic Haplocryolls

(SSS, 2010), while in the bare areas the soils are loamy-skeletal, mixed, frigid Oxyaquic Cryorthents (SSS, 2010). In both cases, the soil is frozen for meters from December to February/March.

The plant species chosen for this study differ for their above-ground and belowground traits. *H. nummularium* subsp. *grandiflorum* is an evergreen trailing plant with loose terminal clusters of bright yellow, saucer-shaped flowers that is rather common in dry and base-rich soils. *D. octopetala* forms dense mats with trailing branches bearing adventitious roots that inhabits particularly well-drained mineral soils (Blaschke, 1991), and that colonizes young soils developed on moraines, especially where nitrogen is scarce (Schwintzer and Tjepkema, 1990). Finally, *S. acaulis* subsp. *cenisia* is a cushion-forming gynodioecious plant with a taproot system that generally grows on wind-exposed ridges, rocky slopes and open alpine grasslands, and can survive extreme temperature from −80 to 60 °C (Larcher et al., 2010).

2.2. Soil sampling and sample preparation

During July 2011, at about 2455 m above sea level, within an area of about 1600 m² (40 × 40 m) we selected three plots with a mean diameter ranging from 5 to 10 m; in each plot all the three plants were present at once. As a control, for each plot a bare soil was also located at least at 1.5 m from each plant. The three individual plants for each species were chosen among those showing the maximum development. For *Helianthemum* we considered plants forming patches of 1.5–1.8 m of diameter, with an estimated age of at least 30–32 years (obtained by the annual ring counting of basal stems). For *Dryas*, we took into consideration plant mats with a diameter of about 1 m, with an estimated age of at least 18–22 years (obtained by the annual ring counting of basal stems). In the case of *Silene*, we selected fully healthy cushions with a diameter comprised between 35 and 40 cm. According to Benedict (1989) and McCarthy (1992), cushions of *S. acaulis* have a growth rate ranging from 0.06 to 3 cm yr^{−1}, even though the maximum rate of 2–3 cm yr^{−1} is reached in the intermediate part of their life, which can attain 350 years (Beschel, 1958). Because of this, we estimated the age of the selected cushions to be more than 50 years.

Within each plot, a soil profile was opened under each plant and in the bare soil. The soil morphological descriptions (Schoeneberger et al., 1998) are reported in the Appendix. From the A horizon (A1 plus A2 in the case of the profiles under the plants) of each profile, a large amount of sample (at least 2 kg) was collected and stored at the field moist conditions in a portable refrigerator. Once in the laboratory, the rhizosphere was isolated according to the method of Corti et al. (2005) from each soil samples by picking up the roots together with the adhering soil. Coarse and medium roots (diameter size larger than 2 mm) were discarded. The soil particles loosely adhering to the roots were detached by gentle shaking and added to the bulk soil. The soil material strictly adhering to the roots, considered as rhizosphere, was recovered by shaking and gentle brushing of the roots. During this operation, the root fragments were removed by using tweezers under a magnifying lens. Aliquots of rhizosphere, bulk and bare soil at field moist conditions were sieved through a 4-mm mesh and stored (for a period not exceeding four weeks) at 2 °C for the biological analyses: microbial biomass C content, basal respiration, and microbial community structure. The remaining soil samples were air-dried and sieved through a 2-mm mesh.

2.3. Physical and chemical analysis

The available water content (AWC) was calculated by difference between the amount of water retained by the soil at 33 kPa and at 1500 kPa, which was determined by pressure plate extractor

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