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Drought and soil amendment effects on monoterpene emission in rosemary plants



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HIGHLIGHTS

GRAPHICAL ABSTRACT

- Rosemary isoprenoid emissions under drought and fertilisation were studied.
- Drought reduced photosynthetic rates, stomatal conductance and isoprenoid emissions.
- Non-oxygenated monoterpene emission was dependent on photosynthesis.
- Organic amendment seemed not to induce a significant effect on isoprenoid emission.



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ABSTRACT

The aim of this work was to study the changes during 15 days in the monoterpene emission rates of the Mediterranean shrub rosemary (*Rosmarinus officinalis* L.), in response to increasing drought stress and fertilisation using two different composts derived from livestock anaerobic digestates (cattle and pig slurry). Drought stress considerably reduced photosynthetic rates, stomatal conductance and isoprenoid emissions and also induced a change in blend composition. In the drought stressed rosemary plants, a positive relationship of non-oxygenated monoterpene emissions and a negative relationship of oxygenated monoterpene with photosynthesis were observed, indicating a different control mechanism over the emissions of the two types of isoprenoids. The emission of nonoxygenated monoterpenes seemed to depend more on photosynthesis and "de novo" synthesis, whereas emission of oxygenate monoterpenes was more dependent on volatilisation from storage, mainly driven by cumulative temperatures. In the short term, the addition of composted organic materials to the soil did not induce a significant effect on isoprenoid emission rates in the rosemary plants. However, the effect of the interaction between fertilisation and seasonality on isoprenoid emission factors throughout the experiment, probably indicating changes in the leaf developmental stage.

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Abbreviations: A, photosynthesis; g_s, stomatal conductance.

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1. Introduction

Volatile isoprenoids are the most abundant Biogenic Volatile Organic Compounds (BVOCs) synthesized and emitted by plants. These compounds play an important role in tropospheric photochemistry by affecting the ozone budget and by increasing the yield of secondary organic aerosols (Atkinson and Arey, 2003; Carlton et al., 2009). Isoprenoids emission is a main defensive line against abiotic (Loreto and Schnitzler, 2010) and biotic stress conditions (Niinemets et al., 2013) and mediate ecological interactions with the biotic environment (Gershenzon and Dudareva, 2007; Gouinguene and Turlings, 2002; Boege and Marquis, 2005; Niinemets, 2010; Loreto et al., 2014). These compounds also have a role in protecting leaves against oxidative and thermal stresses (Loreto et al., 2004; Grote and Niinemets, 2008) possibly through the enhancement of membrane stability and the scavenging of reactive oxygen species (Vickers et al., 2009).

The main driving variables for the emission of isoprenoids that are not stored in permanent pools are photosynthetically active radiation (PAR) and temperature, which form the basis of all emission models (Arneth et al., 2008; Guenther et al., 2006; Monson et al., 2012). However, other environmental factors, such as seasonality, CO₂ and ozone level, mechanical stresses, and drought, have also been reported to influence isoprenoid emissions (Staudt et al., 2000, 2002; Plaza et al., 2005; Blanch et al., 2007; Curci et al., 2009; Peñuelas and Staudt, 2010; McKinney et al., 2011; Holopainen and Gershenzon, 2010; Loreto and Schnitzler, 2010). In some plant species, monoterpenes are synthesized and stored in secretory organs. This is the case for the glandular trichomes of many Lamiaceae (Grote and Niinemets, 2008), e.g. Rosmarinus officinalis L. In these cases, emissions seem to be mainly produced by temperature driven diffusion processes of the stored monoterpenes (Schurgers et al., 2009), though there are increasing evidences that "de novo" synthesized monoterpenes also contribute to the total emission of monoterpenes. For instance, several authors have reported an absence of relationship between emitted and stored monoterpenes in R. officinalis, suggesting that a fraction of the overall monoterpenes produced by R. officinalis leaves could be emitted to the atmosphere directly after synthesis, instead of being stored in storage pools (Peñuelas and Llusià, 1997; Ormeño et al., 2007c, 2009).

Soil water availability represents a major environmental constraint under Mediterranean conditions, and models estimate a further decrease in precipitation in the Mediterranean basin (Gibelin and Deque, 2003). Drought stress caused by low soil water availability does not have a clear impact on isoprenoid emissions. Isoprenoids may decrease, due to restricted carbon acquisition (Hansen and Seufert, 1999; Staudt et al., 2002; Grote et al., 2009; Šimpraga et al., 2011; Burney and Jacobs, 2012), may increase, due to the build-up of intercellular concentration (Sharkey and Loreto, 1993), or may remain unaltered, especially when the stress is prolonged (Peñuelas and Llusià, 1997). Mediterranean soils are also characterised by nutrient deficiencies (Sardans et al., 2006), N and P being the most limiting elements for plant growth and nutrition. Intensive agriculture practices, together with adverse climatic conditions are among the main causes of soil degradation and the loss of soil organic carbon (Bustamante et al., 2011), with negative consequences for plant growth and yield (Turbé et al., 2010).

On the other hand, the intensification of the livestock production implies a potential environmental risk, associated to an inadequate management of the great amounts of organic wastes produced. Anaerobic digestion is an efficient biological method for the energetic valorisation of livestock and agroindustrial wastes, which transforms organic wastes into biogas and the digested material (digestate), the latest being usually composted to improve its properties as organic fertiliser in agriculture (Bustamante et al., 2012, 2013). The application of compost to degraded soils has become a suitable environmental strategy for improving soil physical structure and increasing the amounts of soil organic carbon and other major nutrient such as N and P (Tejada et al., 2006; Bustamante et al., 2012). Compost could affect plant isoprenoid emissions since N and P, which are supplied via compost amendment (Larchevêque et al., 2010), are required for isoprenoid synthesis (Lerdau et al., 1995; Niinemets et al., 2002). However, it has been previously reported that after P and/or N fertilisation isoprenoid emission rates can not only increase (Blanch et al., 2012), but also decrease (Blanch et al., 2007; Fares et al., 2008). Three studies by Ormeño et al. (2009), Olivier et al. (2011a) and Olivier et al. (2011b) on the effects of sewage sludge compost onto isoprenoid emission show variable results, depending on the dose and timing of the treatments (Ormeño et al., 2009). Other previous experiments of fertilisation have also been reported to increase (Lerdau et al., 1995; Possell et al., 2004), decrease (Fares et al., 2008), or not to change (Rosenstiel et al., 2004; Blanch et al., 2007) foliar isoprenoids emissions, depending on plant species, leaf developmental status, type and dose of nutrients, and experimental conditions.

In this work, we aimed to study the changes in monoterpene emission rates of the Mediterranean shrub *R. officinalis* L., in response to increasing drought stress and fertilisation with two different composts derived from livestock anaerobic digestates.

2. Materials and methods

2.1. Characteristics of the soil and of the organic amendments

The soil used in this study was collected from the surface layer (0-20 cm) of a semiarid agricultural area abandoned for ten years in Montelibretti (Rome, Italy, 42° 8′ 7″ N, 12° 44′ 17″ E, 232 m a.s.l). After removal of vegetation, bigger roots and stones, the soil was airdried and then passed through a 2 mm sieve. The soil at the site was a highly calcareous loam soil, slightly alkaline (pH = 7.6), with low salinity (0.10 dS/m), low concentrations of total N (0.067%) and poor total organic C contents (0.75% C).

The composts were elaborated mainly using the solid fraction of anaerobic digestates of cattle and pig slurry (hereafter named CS and PS, respectively), mixed with vine shoot pruning. On a dry mass basis we used: cattle/pig slurry (75%, CS/PS) + vine shoot pruning (25%). A detailed description of the composting process has been reported elsewhere (Bustamante et al., 2012, 2013). These composts showed high contents of total N (29.0 g kg⁻¹ for CS and 30.3 g kg⁻¹ for PS) and a suitable degree of maturity to be used as soil amendments (Bustamante et al., 2012, 2013).

2.2. Experimental procedure

The experiment was carried out in a polycarbonate greenhouse placed at the experimental fields of the Istituto di Biologia Agroambientale e Forestale (IBAF-CNR) (42° 06′ 12″ N 12° 38′ 53″ E, elevation 227 m a.s.l., Montelibretti, Rome, Italy), to avoid potential water incomings from rain and to maintain homogeneous Mediterranean-like environmental conditions.

In this study, two factors were applied simultaneously: fertilisation and drought. For the fertilisation treatment, polyethylene pots were filled with 1 kg of soil thoroughly mixed with PS or CS anaerobic digestate based compost at a dose of 60 t compost (fresh weight basis)/ha. These organic amended soils were compared with unamended soil samples (Control). Each treatment was replicated six times, obtaining a total of 18 experimental units. Cuttings of rosemary were planted a week after establishing the fertilisation treatment, in each one of the pots with the corresponding treated soil. To assure genetic identity, the plants used were exclusively propagated by rooted cuttings. Pots were distributed in a randomised complete block design inside a greenhouse. All the pots were well watered and maintained under natural environmental conditions of light and temperature until the beginning of the drought treatment, four months after planting rosemary. Then, the irrigation strategy was diversified to apply the drought factor in two variables: control plants (well-watered, WW),

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