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Review

# Arbuscular mycorrhizal fungi act as biostimulants in horticultural crops

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ABSTRACT

In the coming years, more sustainable horticultural practices should be developed to guarantee greater yield and yield stability, in order to meet the increasing food global demand. An environmentally-friendly way to achieve the former objectives is represented by the biostimulant functions displayed by arbuscular mycorrhizal fungi (AMF). AMF support plant nutrition by absorbing and translocating mineral nutrients beyond the depletion zones of plant rhizosphere (biofertilisers) and induce changes in secondary metabolism leading to improved nutraceutical compounds. In addition, AMF interfere with the phytohormone balance of host plants, thereby influencing plant development (bioregulators) and inducing tolerance to soil and environmental stresses (bioprotector). Maximum benefits from AMF activity will be achieved by adopting beneficial farming practices (e.g. reduction of chemical fertilisers and biocides), by inoculating efficient AMF strains and also by the appropriate selection of plant host/fungus combinations. This review gives an up to date overview of the recent advances in the production of quality AMF inocula and in the biostimulant properties of AMF on plant health, nutrition and quality of horticultural crops (fruit trees, vegetables, flower crops and ornamentals). The agronomical, physiological and biochemical processes conferring tolerance to drought, salinity, nutrient deficiency, heavy metal contaminations and adverse soil pH in mycorrhizal plants are encompassed. In addition, the influence of bacterial interactions and farm management on AMF is discussed. Finally, the review identifies several future research areas relevant to AMF to exploit and improve the biostimulant effects of AMF in horticulture.



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## 1. Introduction

A primary issue for modern horticulture is facing two contradictory objectives, such as the need to produce food for the increasing world population and to minimise damage to the environment, which can in turn negatively impact horticulture (Duhamel and Vandenkoornhuyse, 2013). Meeting the former two goals represents a major sustainability challenge to the horticultural industry and scientists (Owen et al., 2015). In the last decade, several technological innovations were proposed in order to enhance the sustainability of production systems through a significant reduction of chemicals. A promising and effective tool would be the use of 'biostimulants'. The term biostimulants, often used in the plural form (Hamza and Suggars, 2001), refers to a group of compounds that act neither as fertilisers nor as pesticides, but have a positive impact on plant performance when applied in small quantities (Du Jardin, 2012; Calvo et al., 2014). However, plant biostimulant is still a 'moving target' in the European Union, and its use in the scientific community is still nebulous (Du Jardin, 2012). According to a general definition introduced by the European Biostimulants Industry Council (EBIC) in 2012, 'Plant biostimulants contain substance(s) and/or microorganisms whose function when applied to plants or to rhizosphere is to stimulate natural processes to enhance nutrient uptake, efficiency, tolerance to abiotic stress, and crop quality, with no direct action on pests' (www.biostimulants.eu). Among beneficial microorganisms, arbuscular mycorrhizal fungi (AMF) play a key role in plant performance and nutrition due to their capacity to improve plant mineral uptake (Smith and Read, 2008). AMF can only be grown in the presence of host plants (i.e. obligate symbionts; Owen et al., 2015), and are widely used in horticulture, in particular *Rhizophagus* (formerly known as *Glomus*) *intraradices* and *Funneliformis* (formerly known as *Glomus*) *mosseae* (Krüger et al., 2012). In fact, while the majority of inoculants presented on the market were mostly nitrogen-fixing bacteria products, it is expected that phosphorus-mobilising products including AMF will see an increase in demand (Transparancy Market Research, 2014).

AMF symbiosis is particularly important for enhancing the uptake of the relatively immobile and insoluble phosphate ions in soil, due to interactions with soil bi- and trivalent cations, principally $Ca^{2+}$, $Fe^{3+}$, and $Al^{3+}$ (Tinker and Nye, 2000; Fitter et al., 2011). The basis of this symbiosis is the capacity of AMF to develop a network of external hyphae capable of extending the surface area (up to 40 times) and also the explorable soil volume for nutrient uptake (Giovannetti et al., 2001), throughout the production of enzymes and/or excretions of organic substances (Marschner, 1998). AMF can secrete phosphatases to hydrolyse phosphate from organic P compounds (Koide and Kabir, 2000; Marschner, 2012), and thus improving crop productivity under low input conditions (i.e. phosphorus deficiency, Smith et al., 2011). The extraradical hyphae are also important to increase the uptake of ammonium, immobile micronutrients such as Cu and Zn and other soil-derived mineral cations ($K^+$, $Ca^{2+}$, $Mg^{2+}$, and Fe $^{3+}$) (Clark and Zeto, 2000; Smith and Read, 2008). AMF have been shown not only to improve plant nutrition (biofertilisers), but they also interfere with the phytohormone balance of the plant, thereby influencing plant development (bioregulators) and alleviating the effects of environmental stresses (bioprotector). This leads not only to increases in biomass and yield, but also to changes in various quality parameters (Antunes et al., 2012). The production of horticultural crops with high contents of phytochemicals (i.e. carotenoids, flavonoids and polyphenols) is a primary target that meets the demands of consumers and researchers due to their health-benefit effects (Rouphael et al., 2010a). In a recent review, Sbrana et al. (2014) reported that AMF symbiosis could induce changes in plant secondary metabolism leading to the enhanced biosynthesis of phytochemicals with health promoting properties. The same authors suggested that further research should investigate the mechanism(s) responsible for the increase in plant secondary metabolism through the selection of promising AMF taxa that are able to improve the nutraceutical value of horticultural products (Giovannetti et al., 2013).

In addition to the advantages mentioned above, AMF impart other important benefits such as tolerance to drought (Augé, 2001; Jayne and Quigley, 2014) and adverse soil chemical conditions in particular salinity (Evelin et al., 2009; Porcel et al., 2012), nutrient deficiency, heavy metal contamination (Garg and Chandel, 2010) and adverse soil pH conditions (Seguel et al., 2013; Rouphael et al., 2015).

Another promising tool and a meaningful approach for sustainable horticulture would be the co-inoculation with AMF and other microorganisms such as bacteria (i.e. PGPR) and beneficial fungi (i.e. *Trichoderma* spp.) (Xiang et al., 2012; Nadeem et al., 2014; Colla et al., 2015). The combined use of bacteria and AMF has been investigated in several studies but with contrasting results (Nadeem et al., 2014; Baum et al., 2015; Owen et al., 2015; Colla et al., 2015 and references cited therein). The synergetic/antagonistic effects of microbial inoculants were attributed to the nature and compatibility of the microbial strains used, as well as the interactions that take place between bacteria/fungi and plant species. Therefore, understanding which factors limit the performance of these bio inoculants will be very useful for improving the efficiency of this inoculum pool (Xiang et al., 2012; Nadeem et al., 2014).

Crop management involves a number of practices, which can influence AM symbiosis positively or negatively (see chapter 4; Gosling et al., 2006 and references cited therein). For instance, ploughing and high fertiliser application (i.e. P) can decrease AMF abundance and colonisation (Daniell et al., 2001; Avio et al., 2013; Lehmann et al., 2014). Other factors that may have detrimental effects on AMF symbiosis include the use of specific biocides and cropping with non-host plants (i.e. Brassicaceae, Chenopodiaceae) (Njeru et al., 2015). The later factor can be more deleterious to a highly mycorrhizal plants than phosphorus application or tillage (Gavito and Miller, 1998).

Another important factor is the genotype of a crop. Different cultivars of tomato, for instance, can respond to mycorrhization either with positive growth responses or with an increase in shoot phosphate concentrations (Boldt et al., 2011). Also, the fungal strain