Chapter 8
Biological Significance of Truffle Secondary Metabolites

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8.1 Introduction

Fungal primary and secondary metabolites have an important impact on our society. Best known as mycotoxins, phytotoxins, antibiotics and natural aromas; they represent industries worth billions of dollars. Fungi are also of major importance in terms of biomass: they rank first with an estimated dry weight of 450 kg/ha, which represents 91% of the total soil biomass (microflora and microfauna) (Müller and Loeffler 1976). Yet our knowledge of the ecological significance of fungal metabolites is limited. Despite the pioneer work of Dick and Hutchinson (1966) and Hutchinson (1973) on the effect of volatile fungal metabolites on fungi and plants, this argument seems to have raised little interest in the scientific community. Since then, most studies have focused on parasitic interactions with plants (phytopathogens), while much less attention has been given to the ecological role of the metabolites of symbiotic fungi. An important group of the latter is represented by mycorrhizal fungi. Mycorrhizas are one of the oldest associations between plants and fungi. Dating back to the early colonization of the terrestrial environment (Brundrett 2002), they are classified as endomycorrhizas (arbuscular, ericoid, orchid mycorrhizas) or ectomycorrhizas depending on their ability to penetrate the host-plant root. Truffles fall in the last category of the ectomycorrhizal fungi. Best known for the complex aroma of their hypogeous fruitbodies, truffles were already known to the Greeks and the Romans, but only reached their luxury standing in the last 20 years owing to decreasing production (Fauconnet and Delher 1998; Hall and Yun 2001) and an ever-increasing demand. Despite their high commercial value, very little is known about their biology. Indeed, the unique features of mycorrhizal fungi, from their formation to signal exchange with the surrounding environment (the rhizosphere), are still poorly understood. In addition to the compounds involved in nutritional exchanges between the host plant and the fungus, various micromolecules and macromolecules are secreted into the rhizosphere. These exudates and volatile
organic compounds (VOCs) play an active role in the regulation of symbiosis and interactions with other organisms, including nonhost plants.

More than 200 VOCs and many nonvolatile compounds have been identified from various truffle species. The aim of this chapter is to discuss the ecological significance of these metabolites (VOCs and/or exudates) associated with three levels of differentiation: fruitbody, free-living mycelium and mycorrhizas. Furthermore, the possible role of these metabolites in the interaction with the host plants and nonhost plants (the so-called burnt, a zone with scarce herbaceous cover) shall be discussed.

8.2 Truffles: Life Cycle and Distribution

Ectomycorrhizal symbiosis has evolved repeatedly over the last 130 million to 180 million years (LePage et al. 1997). In boreal and temperate forests, 95% of the short roots of plants form ectomycorrhizae (Martin et al. 2001), with 5,000–6,000 species of basidiomycetes or ascomycetes—including truffles (Buscot et al. 2000; Martin et al. 2001). Ectomycorrhizae positively impact plant growth in nature (Read 1991) owing to improved nutrient uptake and protection against pathogens (Borowicz 2001; Buscot et al. 2000).

Truffles are hypogeous ascomycete fungi belonging to the genus *Tuber*, the family *Tuberaceae* and the order *Pezizales* (O’Donnell et al. 1997; Trappe 1979). Their mycorrhizal status was established worldwide in the 1960s (Harley and Smith 1983; Trappe 1962). Truffles live in symbiosis with plant roots, generally forming ectomycorrhizas. In contrast to the high degree of promiscuity exhibited by arbuscular mycorrhizal (AM) fungi towards their hosts, ectomycorrhizal fungi are rather host-specific. Indeed, truffles tend to associate with angiosperms and gymnosperms, predominantly with oaks, hazels, some species of pines, but also some species of shrubs like *Cistus*. For a complete list of the host plants of European truffle species, refer to Ceruti et al. (2003). Recently truffle mycelium has also been identified within orchid roots—even though it does not form ectomycorrhizas (Selosse et al. 2004).

The present information about truffle’s life cycle is very patchy. On the basis of observations both in nature and in the laboratory, as well as possible similarities with the life cycle of other ascomycetes fungi, Lanfranco et al. (1995) proposed a model for the life cycle of truffles which can be divided into three phases: (1) a reproductive phase (fruitbody), (2) a vegetative phase (free-living mycelium—saprotrophic phase actually only observed in the laboratory) and (3) a symbiotic phase (mycorrhizas) (Fig. 8.1). Indeed difficulties arise from the impossibility to follow the full life cycle in the laboratory. Even though Fassi and Fontana (1969) reported production in pots of fruitbodies of *Tuber maculatum* in association with *Pinus strobus* (Fig. 8.2), this achievement has not been repeated since then for any truffle species! Nevertheless more insight has recently been gained into the life cycle of truffles by Paolocci et al. (2006). In an elegant experiment the authors