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## **Adverse effects of human activities on the diversity of macrofungi in forest ecosystems**

### **Abstract**

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A rapid decrease in the size and composition of macromycetes populations has been observed in various regions of Europe. Such changes have been mainly attributed (directly or indirectly) to human interference, and most notably to the degradation of natural ecosystems and to air pollution. Especially as regards ectomycorrhizal fungi, it has been demonstrated that many of them are very suitable bio-indicators of the disturbance of forest ectotrophic stability. Air pollution affects negatively not only the number of species but also the number of basidiomata produced by this category of fungi; in contrast, fungal communities seem to get enriched in lignicolous species. Studies focusing on the determination of the deterioration of forest ecosystems are practically non-existent in the Mediterranean area. Hence, monitoring of the suitable groups of macrofungi in terms of both qualitative and quantitative assessments could provide valuable pertinent data.

### **Introduction**

The lack of knowledge on fungal biodiversity is evident by the fact that possibly only 5-10% of fungal species have been discovered and described (Hawksworth 1991). Only during the last two-three decades fungi started to receive attention as essential components of natural ecosystems. As regards the Mediterranean region in particular, pertinent data are rather scarce and fragmentary, while studies on the ecology of fungal species and the influence that environmental factors exert on their communities have been significantly delayed.

On the other hand, of significant importance to the ongoing fungal conservation initiatives in Europe is the publication of national and/or regional check-lists in many Mediterranean countries (e.g. Bernicchia & al. 2005; Dimou & al. 2002; Onofri & al. 2003; Ortega & Linares-Cuesta 2002; Venturella 1991; Zervakis & al. 1998, 1999), and the compilation of provisional-preliminary Red Lists (e.g. Ivančević 1998, Venturella & al. 1997, 2002).

### Environmental factors and the diversity of macrofungi: a review of literature data

During the recent past, the intensification of human activities led to high increase of air pollution and to the degradation of ecosystems, which in turn caused significant adverse effects in fungal communities as well. Air pollution may influence fungi through the greenhouse effect producing a climate change, as well as exerting an indirect effect through modification of vegetation. Such changes might threaten climate-sensitive species and/or favor the appearance/growth of more thermophilic taxa which could in turn act as alien competitors against native species. In addition, interactions between biotrophs and their hosts are also modified. Another global problem is the destruction of habitats and the decrease of forest areas. Some other environmental or pollution-related issues with a more local or specific effect on fungi include:

- deposition of various pollutants (incl. depositions from acid rain) leading to soil modifications or contamination of water resources,
- accumulation of metals on substrates interfering with fungi,
- occurrence of residues resulting from the widespread use of pesticides and fungicides mainly in adjacent agricultural lands,
- eutrophication, i.e. contamination of water resources with nutrients draining off from agro-industrial activities,
- desertification, caused by shifts in precipitation patterns resulting from climatic change,
- fragmentation of habitats resulting from forest cutting, urban extension, alterations in land uses and change of agricultural practices.

Experimental data on the effects which the above mentioned factors exert on fungal diversity are indicative of the state of relevant research in Europe.

The most indicative examples of the adverse effects of natural or anthropogenic disturbances are provided by studies on ectomycorrhizal-forming fungi. Decline in the populations of the edible mushroom *Cantharellus cibarius* is well documented in the Netherlands, and it is likely that the decline of this species is representative for many more ectomycorrhizal species (Dahlberg 1991). Forests which are particularly rich in ectomycorrhizal fungi are often situated on hilly sites with a nutrient poor soil with a thin or absent organic layer (soil-erosion problems) and a low coverage of vascular plants. These forest communities are threatened by eutrophication and/or acidification caused by air pollution. Clearly many of the ectomycorrhizal fungi dependent on living trees and other fungi characteristic of old, undisturbed soils will disappear. However, the inoculum potential of (some) ectomycorrhizal fungi remains present for several years in clearcuts (Dahlberg 1991).

Fellner (1993) described the decline of ectomycorrhizal fungi in Central Europe in terms of the disturbance of ectotrophic forest stability as a consequence of air pollution. This process includes latent, acute and lethal stages corresponding with both specific phases of the impoverishment of ectomycorrhizal and the enrichment of lignicolous mycocoenoses. Ohtonen & Markkola (1989) estimated the effect of local air pollution on the basidiomata production by mycorrhizal fungi and on microbial activity in Scots pine forests. The species composition of the mycorrhizal fungi varied in the differently polluted areas. The biomass and number of basidiomata and biological activity in the humus seemed to decrease towards the most polluted areas. Reasons for this may lie in the pH, the amount of ammonium nitrogen, total nitrogen and total sul-

phur present, all of which were at a higher level in the most polluted areas. Basidiomata production and biological activity showed negative correlations with these soil parameters. The mycorrhizal fungi were in the poorest condition in the most polluted area.

Poor soil quality hinders propagation of fungi and plants, and greater distances for inoculum dispersal have to be overcome to the nearest undisturbed vegetation. Erosion, in particular, is an issue of concern in rangelands and has been documented as a cause of inoculum depletion (Hall 1979). Many less drastic disturbances occur, that also reduce inoculum, including tillage, fertilization, irrigation, pesticides and heavy grazing. Grazing has variable effects on mycorrhizal fungi depending upon the degree of grazing and the yearly variability in precipitation.

Baar & Kuyper (1993) observed that removal of litter and humus layers by management practices or by wind had a positive effect on ectomycorrhizal fungi. Field experiments have been set up in stands of *Pinus sylvestris* to study the effects of removing the ectorganic layer in a more detailed way. Sod-cutting had a positive effect on the number of ectomycorrhizal species and on the number of sporomata. Adding sods had a negative effect on the number of ectomycorrhizal species and on the number of sporomata.

Modern agriculture increasingly uses chemical treatments that can give rise to various kinds of environmental modifications. This problem is especially well documented in Europe for grassland fungi, both macromycetes such as *Hygrocybe* spp. but also soil fungi. The nitrogen content of the basidiomata of *Lactarius rufus* and *Suillus variegatus* was measured on unfertilized and fertilized plots in four forest stands of different types in Finland in 1978-1984 (Ohtonen 1986). The effects on the fungal diversity of fertilizing forests has been reviewed by Kuyper (1989). In experiments N-fertilizer was applied with the purpose to stimulate the growth of trees or to compensate for (harmful) influences of air pollution. Nitrogen fertilization has a strong inhibitory effect on ectomycorrhizal symbiosis in laboratory as well as in field experiments. Only a few species increase after fertilizing e.g. *Paxillus involutus*, *Laccaria bicolor* or are indifferent (*Lactarius rufus*). Many species decrease or even disappear completely, e.g. many *Cortinarius* and *Suillus* species. It is not yet apparent whether the mycorrhizas themselves are affected as well, yet the fructification process seems to be most sensitive. NPK fertilizer application also has a strong negative effect on most ectomycorrhizal fungi, comparable with N fertilizer alone (Hall 1978). Fertilizing causes a decline in some of the saprotrophic species and a shift in species composition towards eutraphent species. In a fertilization experiment in a Scots pine forest in the Netherlands *Mycena sanguinolenta*, *Clitocybe vibecina*, *Entoloma cetratum* decreased whereas *Mycena galopus*, *Clitocybe ditopus* and *C. metachroa* increased after application of fertilizer (Kuyper 1989).

Furthermore, according to the investigation of Newsham & al. (1992) the occurrence and the ecophysiology of fungi are affected by sulphur dioxide (SO<sub>2</sub>), a common pollutant in the atmosphere over continental Europe and North America.

### Mushrooms as bioaccumulators of heavy metals and radionuclides

Extensive research has been carried out since the 1970's on trace elements (mainly heavy metals) occurrence in mushrooms, which was mainly focusing at screening several mushrooms species as bio-indicators of environmental pollution and detecting those edible species accumulating high levels of heavy metals. The ability of macromycetes to adsorb and accumulate certain heavy metals (e.g. cadmium, mercury, lead and copper) in quantities higher than those detected in soil and plants has been demonstrated by several authors (Klan 1984; Ohtonen 1982, etc.). This is explained by the intensive contact attained between the mycelium network and the growth substrate (soil, litter, wood), and by the osmotrophic abilities of these organisms. In contrast, the proportion of metal concentration in sporomata deriving directly from atmospheric depositions seems to be of less importance due to the short lifetime of a fruiting body.

Several reviews of heavy metal concentrations in mushrooms have been published (Kalac & Svoboda 2000; Seeger 1982; Vetter 1994). Results demonstrate, among others, that some species accumulate high levels of cadmium and mercury even in unpolluted and mildly polluted areas, while the concentrations of both metals (and also of lead) increase considerably in heavily polluted sites. For example, the genus *Agaricus* seems to be a particularly effective accumulator for both for cadmium and mercury (Schmitt & Meisch 1985; Seeger 1982), while heavily accumulating mercury and lead species are also *Calocybe gambosa*, *Lepista nuda*, *Lycoperdon perlatum*, *Macrolepiota* and *Boletus* spp. (Kalac & Svoboda 2000; Sameva & al. 1999). The content of several other metals (e.g. chromium, copper, zinc, cobalt) has been measured in mushrooms, often in quantities exceeding common levels; comparison of metal concentrations in the sporomata demonstrate that many species may accumulate selectively (Kalac & Svoboda 2000).

In regions where the anthropogenic impact is more pronounced, dry deposition of particles containing heavy metals on the relatively large surface of macromycetes also plays an important role. This issue is of special interest since consumption of edible mushrooms growing in urban and industrial regions has caused several cases of human poisoning, which was attributed to their high (toxic) accumulation of content in heavy metals.

Dimitrova & al. (1999) investigated the arsenic content in basidiomata of different fungal species. They showed that the presence of arsenic content exceeded two times the MAA (Sanitary Standards of Maximum Admissible Amount) in *Agaricus silvicola*, *A. arvensis*, *Macrolepiota procera*, *M. rhacodes*, *Lycoperdon pyriforme*, *L. perlatum*, *Clitocybe gibba* and *Calvatia utriformis*. Stijve & al. (1990) analyzed the arsenic content from seven *Laccaria* species. The arsenic accumulating ability of *L. amethystina* was amply confirmed. *L. laccata* var. *pallidifolia* and *L. purpureobadia* might also possess the ability to concentrate arsenic, but they will only do so under certain (yet unknown) conditions.

Turnau (1989) investigated the effect of different types of industrial dust on the mycorrhizal status of a *Pino-Quercetum* plant community in a mixed forest near Krakow (Poland). A marked decrease in mycorrhizal plant population was observed in all plots. Turnau & Kozłowska (1991) analyzed the influence of industrial dust on the heavy metal content in different fungal species. *Armillaria lutea*, *Auriscalpium vulgare* and *Mycena ammoniaca* demonstrated their ability to accumulate heavy metals.

It is also known that mushrooms are efficient accumulators of radionuclides, as it was particularly evidenced after nuclear-plant accidents (e.g. Chernobyl, April 1986). By this accident a wide range of radioactive nuclides were released into the environment, the most important isotope of which was  $^{137}\text{Cs}$  because of its long half-life (30 years). Since then, the radiocesium levels detected in fruits and vegetables have decreased, while in fungi the activity of radiocesium continues to be very high. Wasser & Grodzinskaya (1996) studied radionuclide accumulation in the basidiomata of macromycetes during the vegetation season of 1990-1991 in 44 locations of the Ukraine. Content of  $^{137}\text{Cs}$  in basidiomata was 1-2 orders of magnitude higher than in the substrata on which they were growing. Species belonging to the families *Amanitaceae*, *Boletaceae* and *Russulaceae* were mainly characterized by high contents of radiocesium and may be considered as bioindicators of the radioactive contamination of the area. Increased radiocesium accumulation in mushrooms was as follows: lignotrophs > saprotrophs > mycosymbiotrophs.

### Edible mushroom harvesting

Air pollution and intensive forest management (leading to declines in forest health and forest ecosystems), together with climate change, pollution from growing urban areas, ozone depletion, introduced pathogens, and intensive timber harvesting, are thought to be major contributors to decreased mushroom diversity and productivity in Europe (Arnolds 1991). Initial studies of the impacts of edible mushroom picking have concluded that rational harvesting does not diminish subsequent fruiting (Egli & al. 1990, Norvell & al. 1995), but these small-scale studies have not adequately addressed the impacts of large-scale commercial mushroom harvesting or forest management activities over long periods of time. Ivančević (1998) stated that in the early nineties, warnings were issued about the diminishing quantities of the wild mushrooms harvested year per year, and that certain species of macromycetes were endangered. Nowadays, ECCF (European Council for the Conservation of Fungi) members from eastern and from southern Europe complain about the damage caused to their forest ecosystems by commercial harvesting of edible mushrooms that are exported to western Europe.

Pilz & Molina (1996) proposed a regional approach to edible forest mushroom monitoring and research in the USA's Pacific Northwest. Its objectives included:

Low-intensity, long-term monitoring of areas with heavy commercial harvesting to ensure harvest sustainability and evaluate reasons for potential trends.

Low-intensity, long-term monitoring of natural areas (where neither timber nor mushroom harvesting occurs) to provide control sites for interpreting trends in commercially harvested areas and to detect trends related to regional changes in the environment or forest health.

Intensive, short-term research on correlations between mushroom productivity, habitat, and stand management activities to provide forest managers with the information needed to ensure future habitat availability and mushroom collection opportunities.

Use development of this research and monitoring program as a prototype for cost-effectiveness ensuring the sustainable harvest of an array of other non-timber forest products by engaging interested public in research and monitoring activities.

The program design of this initiative involving (among others) integrated research and monitoring activities, voluntary participation by interested agencies, organizations or individuals, common core sampling procedures, site selection criteria and meta-data evaluation, is a good example of the type of work needed to rationalize managing of forest ecosystems and edible mushroom harvesting.

### **Influence of other factors on fungal diversity**

Besides the adverse effect that most human activities exert on fungal diversity, appearance and abundance of mushrooms are influenced by climatic conditions. The temperature and humidity of the air and soil are among the principal factors that regulate fungal growth and reproduction. The influence of environmental factors, both climatic and edaphic, could be evidenced by the major variations observed in species distribution and yields, and also as a function of time, i.e. in variations between years and growth seasons. The evidence reported from mycodiversity studies in Greece and Sicily (Venturella & Zervakis 2000; Zervakis & al. 2002, 2002a) confirmed that fungi require a certain level of moisture; rain-falls, air humidity and soil moisture, which are all significant factors for the achievement of a good crop of mushrooms; the rainfall during the last 3-5 months preceding mushroom appearance has a marked effect on their productivity; rare species occasionally appear as a consequence of a particularly hot summer; the number of mycorrhizal species do not increase in unusually warm summers; peak temperatures exercise their influence predominantly via the soil, affecting its moisture content; particularly low temperatures have a direct (detrimental) effect on the growth of fungi. Finally, the fact that high total annual precipitation is predictive of a poor yield of ectomycorrhizal mushrooms in the autumn of the following year, may be attributed to the good yield almost certainly obtained during the autumn of the previous year, especially when the preceding summer was rainy (since this high yield would have undoubtedly reduced the potential of the mycorrhizal fungi to produce another good yield immediately after).

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