Hosts and distribution of *Armillaria* species in Serbia

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Abstract

Twenty-five tree species were recorded as hosts for five European *Armillaria* species in studies on forest ecosystems in Serbia. *Armillaria* was most frequently isolated from the conifers *Picea abies* and *Abies alba* and from the deciduous trees *Fagus moesiaca* and *Quercus petraea*. *A. mellea* and *A. gallica* coexisted in hardwood forests in northern and central parts of Serbia, while *A. ostoyae* and *A. cepistipes* were mostly present in coniferous forests in the southern mountain region of Serbia. The distribution depended on the *Armillaria* species, altitude, and the forest type.

Introduction

The genus *Armillaria* has a worldwide distribution from tundra in the north to the tropical forests around equator and the forests of Australia and Patagonia in the south. The genus includes at least 36 species (Watling *et al*. 1991; Volk & Burdsall 1995), with seven morphological species present in Europe (Guillaumin *et al*. 1985; Termorshuizen & Arnolds 1987). Six of the European *Armillaria* species have a wide distribution in forest ecosystems, while *A. ectypa* is growing only on peat bogs (Korhonen 2004). The European species differ in geographical distribution, ecological behaviour, host range, and pathogenicity (Guillaumin *et al*. 1993).

The economic significance of *Armillaria* derives from its role as a parasite of woody plants. *Armillaria* species can behave as primary and secondary pathogens causing root and butt rot on numerous coniferous and broadleaved trees species both in natural regenerated forests and in plantations (Guillaumin *et al*. 1993; Morrison *et al*. 2000). As parasites, *Armillaria* spp. can cause significant economic loss and influence the tree species composition of forests (Kile *et al*. 1991).

This study was performed to increase the knowledge about hosts and distribution of *Armillaria* species in forest ecosystems in Serbia.

Materials and methods

The study was conducted on 34 sites in Serbia and on one site in Montenegro (Fig. 1). The sites were chosen so, that they were distributed evenly throughout the country. The Site Durmitor in Montenegro was chosen because of its importance as a National Park under protection of UNESCO and because of its conserved forests.

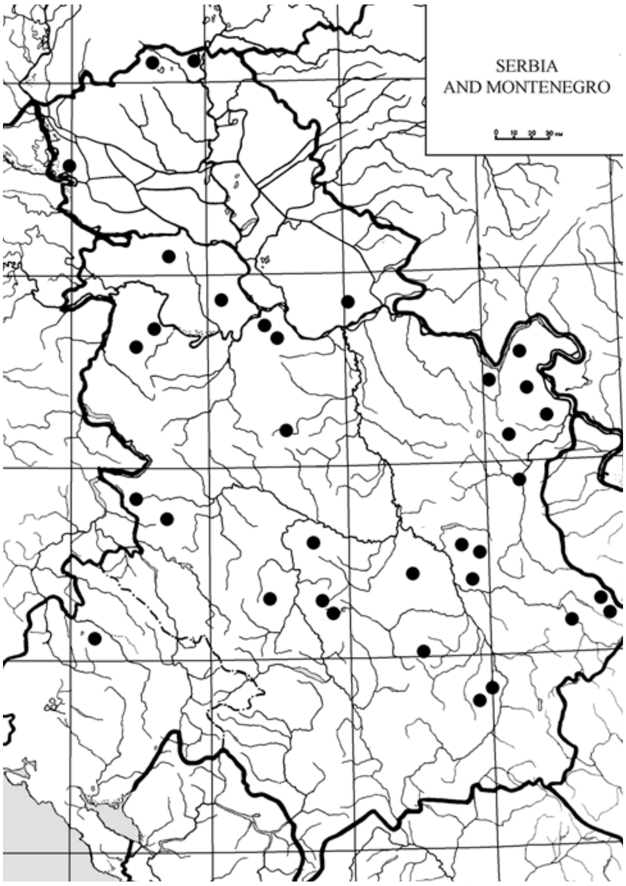

Fig. 1. Distribution of sites in Serbia from which Armillaria *species were found*

The sites studied included all dominant forest ecosystems. Different oak associations in the plain and beech associations in mountain regions were studied. Mixed forests of broadleaved and coniferous species (beech–fir, beech–spruce, beech–fir–spruce associations) were of special interest for this study, because of complex host – *Armillaria* spp. interactions.

Sampling

The sampling was done in 2002, 2003 and 2004. Sampling within the plots was systematic and focused on dominating tree species but if symptoms of *Armillaria* attack were present on other tree species samples were collected for those species as well. Sampling followed descending order of priority. Trees were examined for symptoms of decline such as crown dieback, early discolouration of needles or leaves, or presence of small leaves. If *Armillaria* species were suspected to be present, the root collar of major roots was excavated. When potential signs or symptoms of cambial infection were observed on the living trees (resin flow, discoloration or sunken areas of bark), small areas of bark were removed to check for the presence of mycelial mats in cambial zone. Following examination of living trees, recently died trees, snags, stumps, wind-thrown and broken trees were also examined and sampled. Rhizomorphs, wood samples, mycelial mats and basidiomata were collected from 59 living trees, from 39 recently died trees and from 56 decaying trees.

Identification of isolates

Identification of isolates was performed by: a) the polymerase chain reaction (PCR) and sequencing (Chillali *et al*. 1998), b) haploid – diploid pairings according to the method of Korhonen (1978), and c) identification of basidiomata (Termorshuizen & Arnolds 1987).

Results

Species identification

Armillaria species were found on 34 sites studied (Fig.1), 152 plots or on 81 % of the controlled stands. There were no obvious differences between stands where *Armillaria* species were detected or not. A total of five *Armillaria* species were identified. *Armillaria gallica* was the species most commonly isolated (73 isolations from 27 sites), followed by *A. mellea* (51 isolations from 20 sites), *A. cepistipes* (36 isolations from 12 sites), *A. ostoyae* (25 isolations from 15 sites), and *A. tabescens* (4 isolations from 4 sites). Four isolates could not be identified as any of tested species.

Hosts

Armillaria species were found on 25 tree species that are dominant in the forest ecosystems on the studied sites. Different *Armillaria* species were isolated from 15 hardwood and 10 coniferous hosts (Table 1). Most of isolates were from spruce (45), fir (21), beech (19), and sessile oak (15).

Fifty-three percent of isolates were from conifers and 47 % from broadleaved hosts. Frequencies of isolates from conifers were: *A. cepistipes* (30 %), *A. ostoyae* (26 %), A. *mellea* (23 %) and *A. gallica* (21 %). On hardwoods *A. gallica* was the most common (58 %), followed by *A. mellea* (31 %). The other species were only occasionally found; *A. cepistipes* (7 %), *A. ostoyae* (2 %) and *A. tabescens* (2 %). *Armillaria tabescens* was observed only on hardwoods and only on oaks.

Armillaria gallica was found more frequently than expected by chance on beech and hornbeam, in 40 % of isolates, while *A. ostoyae* and *A. cepistipes* were more frequently observed on conifers. For *A. mellea* there was no statistically significant difference between association with conifers or hardwoods. Sessile oak and Austrian pine were the most frequent hardwood and conifer hosts for *A. mellea. Pinus nigra* was hosting only *A. mellea* and *A. ostoyae,* while *A. tabescens* was isolated only from *Quercus petraea* and *Q. robur*.

Table 1. Number of isolates of *Armillaria* spp. obtained from different tree species in Serbia

Hosts	**No.**
Conifers (10 species)	
Abies alba	21
Abies concolor	2
Cedrus atlantica	2
Larix europea	2
Picea abies	45
Picea omorika	4
Pinus nigra	10
Pinus sylvestris	3
Pinus strobus	7
Pseudotsuga taxifolia	6
Hardwoods (15 species)	
Acer heldreichii	1
Acer pseudoplatanus	3
Carpinus betulus	13
Fagus moesiaca	19
Fraxinus excelsior	3
Prunus domestica	2
Quercus cerris	3
Quercus farnetto	12
Quercus petraea	15
Qurcus robur	12
Quercus rubra	1
Robinia pseudoaccacia	2
Tillia argentea	1
Ulmus carpinifolia	2
Ulmus montana	1

Geographic and altitudinal distribution

Armillaria species were found in the range between 70 and 1820 m above see level (Table 2), where they accompanied trees in major forest ecosystems.

Armilliaria mellea was found in northern lowland forest types, and in eastern hilly region of Serbia with dominant forests of sessile oak, beech and hornbeam. It seems that in these ecosystems the fungus found optimal ecological conditions, characterized by forests with dominating hardwoods, especially oak species.

Armillaria gallica was found in all major regions except in the high mountains of Kopaonik, Stara Planina and Golija. It was present in beech and xerophilous forests of different oak species, but also on conifers at the higher altitudes. *A. gallica* was less frequent above 1.000 m altitude. *A. tabescens* was observed only in dryer forest ecosystems of Hungarian oak and Turkey oak at low altitudes. *A. cepistipes* was found only at altitudes above 590 m, and based on its frequency in different areas, the ecological conditions favouring *A. cepistipes* locate in the mountain areas in the south central and eastern part of country.

Table 2. Altitudinal distribution of *Armillaria* species in Serbia

***Armillaria* sp.**	**Altitude (m)**		
	Minimum	**Optimum**	**Maximum**
cepistipes	590	1.000–1.500	1.820
gallica	60	– 1.000	1.450
mellea	70	– 800	1.040
ostoyae	850	900–1.600	1.820
tabescens	70	– 250	250

Armillaria ostoyae was predominantly found in southern part of Serbia between 44 and 43 ° N, which corresponds to the extension of Dinaric Alps and Balkan mountains. Distribution of this species overlaps with the occurrence of conifer species at higher altitudes.

Discussion

Five *Armillaria* species were now found during a survey of forest ecosystems in Serbia. Up to three *Armillaria* species were found in single sites, but on most sites two *Armillaria* species were coexisting. Combinations of *Armillaria gallica/A. mellea* and *A. ostoyae/A. cepistipes* were most frequently observed, and on some mountain sites the combination of *A. ostoyae/A. cepistipes/A. gallica* was common.

Armillaria species occurring in European forests have a wide distribution throughout the continent. *Armillaria borealis* has the northernmost distribution, its northern limit coinciding with the limit of woody vegetation in Scandinavia (Roll-Hansen 1985). The species has been found only in Europe, and the most eastern record is from Ural region in Russia (Korhonen 2004), while the southern limit is somewhere in Slovenian part of Alps (Munda 1997) and plains of Hungary (Szanto 1998).

Armillaria cepistipes has a very wide distribution from the Arctic Circle (66 °N) (Korhonen 1978) to the mountain Vernon (40°40' N) in Greece. In Serbia and Montenegro *A. cepistipes* follows the high mountain massif between 44° and 43° N. According to the data from Balkan (Tsopelas 1999; Lushaj *et al*. 2001) and Serbia, this species follows the woody vegetation to its disappearance, which has been also observed in the Alps in central Europe (Rigling 2001).

Armillaria ostoyae occurs independently of latitude or altitude in European coniferous forests with continental or oceanic climate type (Guillaumin *et al*. 1993). As observed in Mediterranean countries, *A. ostoyae* was now found only at high altitudes in Serbia. High mountains of Dinaric Alps (south-western part of Serbia) and Balkan Mountains (south-eastern part of Serbia) massifs were the only sites where this species was recorded. *A. ostoyae* appeared above 800 m, but its optimal growth conditions seem to locate between 1000–1600 m. On higher altitudes its occurrence decreased, but still it accompanied coniferous forest types to the end of vegetation. It seems that the altitudinal distribution of *A. ostoyae* is similar between southern and central part of Europe and influenced by the distribution of conifers.

Armillaria gallica is widely distributed throughout the European continent, but its distribution is highly dependent on altitude (Guillaumin *et al*. 1993). In the French Massif Central *A. gallica* is predominant in forests up to 850 m, but becomes rare at higher altitudes, though it still is present up to an altitude of 1100m. Because of the continental climate type prevailing in northern and central part of Serbia this species is rare at altitudes above 1000 m and absent from altitudes above 1400 m.

Armillaria mellea occurs in central and south Europe, but is common only in the southern and western parts of this area (Korhonen 2004). In central part of France the species is present in all predominant forest types at altitudes below 900 m (Legrand & Guillaumin 1993) but further south the species can occur at altitudes up to 1400 m in Albania (Lushaj *et al*. 2001) and up to 1750 m in Greece (Tsopelas 1999). Records from Serbia show that this species is distributed throughout the country, except in high mountain region.

Armillaria tabescens is the most thermophilic species and it was found in Serbia only in the altitude range between 70–250 m. This does not correspond with the data from Greece (Tsopelas 1999) and Albania (Lushaj *et al*. 2001), where the species has been found at altitudes up to 1150 m and 1300 m, respectively. Climatic conditions may explain this difference since Serbia has a more continental climate than the others.

Due to their wide host range *Armillaria* species can survive for a long time on an occupied forest area (Kile *et al.* 1991). These fungi can successfully survive on plant remains and wait for an opportunity to colonize new substrate, either as opportunists or primary pathogens. A simplistic view of interactions between hosts and *Armillaria* species is that *A. mellea*, *A. gallica* and *A. tabescens* occur primarily on hardwood species, while *A. ostoyae*, *A. cepistipes* and *A. borealis* prefer conifers (Kile *et al*. 1991, Fox 2000). However, it should be kept in mind that all these species can successfully colonize both conifers and broadleaved trees.

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