NEW LOCALITIES AND HABITAT CONDITIONS FOR *CHOIROMYCES MEANDRIFORMIS* VITT. (ASCOMYCOTA, FUNGI) IN NORTHERN ROMANIA

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Abstract: Although *Choiromyces meandriformis* is considered as a wide-spread fungus species in forest habitats of Europe, still it is listed as rare, endangered and even critically threatened in some countries. In Romania the species is sporadically spread throughout the Carpathian Mountains and surrounding areas, in soils under both deciduous and coniferous species. In this study there are presented two new localities for *Choiromyces meandriformis* in northern part of Romania, where the species was identified in spruce communities. The specimens were macroscopically and microscopically characterized and soil analyses were performed. Also, a phytosociological investigation of the forest habitat was performed. Soil analysis highlighted the species preference for acidic soils, with a medium content of humus, total nitrogen, potassium and total phosphorus, and a reduced amount of organic matter. The forest community (*Hieracio transsylvanici-Piceetum*) was characterized by a uniform and species-poor floristic composition.

Keywords: *Choiromyces meandriformis*, ecology, habitat, soil characteristics, hypogeous fungi.

Introduction

Choiromyces meandriformis is a hypogeous fungal species, wide-spread in Europe, from the UK in the western part of the continent to the extreme east of Russia, from Sweden in the north, to Spain, Italy and Greece to the south. Etymology of name derives from the Greek words *choiros* = pig + *mykes* = mushroom (porcine truffle) and Latin *maeander* = meander, sinuous river + *formis* [BRESSION, 1996]. The genus *Choiromyces* is characterized by hypogaeal, sub-globular or irregular shaped sporocarps. Their gleba is solid and has visible veins. The asci are indehiscent, clavated, with 8 spores, and no reactivity to the Melzer reagent. The spores are usually globose, hyaline or yellow-brown and ornamented with thorns, pits, or alveolated cross-links. The species *Choiromyces meandriformis* is collected and consumed as a delicacy in Sweden [WEDEN & al. 2009]. Despite its economic importance, limited information on this fungal genus diversity and distribution is available worldwide. Based on the morphologic similarity, some species within the *Choiromyces* genus may be confused with species of the *Tuber* genus on the market. Thus, some species of *Choiromyces* (e.g. *C. meandriformis*) may be sold as *T. magnatum* Picco, a highly edible species of the *Tuber* genus on European markets for the uninitiated people [MORENO & al. 2012].

Choiromyces meandriformis occasionally occurs under douglas, pines and under *Tsuga* at low altitudes, being slightly widespread in North America (and common in Europe)

[BEUG & al. 2014]. It is apparently a relative common species of truffle and appears to be widespread in the United Kingdom [HOBART, 2005]. In Italy, collected data over the last decade regarding the ecology and distribution of hypogean and semi-hypogean fungi allowed the assessment of their status, and species within *Choiromyces* sp. were considered rare [VENTURELLA & al. 2011]. Also, in Poland, it is considered a rare species [HALAMA & PANEK, 2000], that produces sporocarps at the end of summer and in autumn and occur in mixed oak and beech forests. In France, it is also encountered during the summer season, being one of the largest hypogean mushrooms in the country, which often produces cracks in the soil, thus being easily identified [FOURRÉ, 2000]. In Spain, it has been identified in the Basque Country [CALONGE & al. 1977] and in Navarra at higher elevation (c.a. 1200 m a.s.l.) under *Picea abies* trees [SIERRA & al. 1991]. In Lithuania, the species was found in the vicinity of hardwoods (oaks, birch, alder and willow), in mixed and coniferous forests, from August to October [KUTORGA & KATARŽYTĖ, 2008]. Some localities (Izmir, Uşak, Bolu, and Samsun) have been reported from Turkey [TÜRKOĞLU & CASTELLANO, 2014]. In Bulgaria is included in the Red List of Fungi [GYOSHEVA & al. 2006] as endangered (EN) species. It is also included in the Red Book of Lithuania (2007) as endangered [KUTORGA & KATARŽYTĖ, 2008].

In Romania, *Choiromyces meandriformis* is encountered from July to September [SĂLĂGEANU & SĂLĂGEANU, 1985]. The species is widespread throughout the Carpathian Mountains and surrounding regions, and can be identified in soils under both deciduous and coniferous species. Comprehensive information on distribution of hypogean fungi in Transylvania (and Harghita County in particular) was published in the work of PAP & al. (1983). Also, according to PAZMANY (1990) *Choiromyces meandriformis* is found in Alba County (Zlatna), Caras-Severin County (Anina, Băile-Herculane and Moldova Nouă), Cluj County (Cluj-Napoca and surrounding areas), Covasna County (Sânzieni and Ojdola), Harghita County (Gheorgheni, in forests dominated by fir, Borsec, Ditrău, Mihăileni, Toplița, Rugănești, Cristuru-Seuciesc), Maramureș County (Ocna-Șugatag, in areas dominated by oak and Maramureș depression), Salaj County (Romanași). It was also identified in Sibiu County, in Tufa Mohului Forest [NEGREAN & DRĂGULESCU, 2005].

The main objective of this article was to present new localities where the species was identified and to characterize the habitat in respect to soil properties and forest characteristics.

Material and methods

The specimens were identified based on the analysis of macroscopic and microscopic characters, using literature: GERHARDT (1999), SĂLĂGEANU (1985), ASTIER (1998), COURTECUISSE & DUHEM (2013), TRAPPE & CASTELLANO (2007). The nomenclature followed Index Fungorum [<http://www.indexfungorum.org/Names/Names.asp>]. The soil analyses were performed at The Offices of Pedological and Agrochemical Studies (OSPA) Iași. The trees age was estimated according to the diameter and the forest documents. In order to characterize the floristic composition of habitats, phytosociological relevés have been performed according to the method of the Central European School for the study of vegetation [BRAUN-BLANQUET, 1964; BORZA & BOȘCAIU, 1965].

Results and discussions

In this study are presented two new localities for *Choiromyces meandriformis* from northern part of Romania (Suceava County), where the species was identified in spruce young forests (*Picea abies*), in the first half of July 2013 and 2014 (Figure 1). The first spruce community was situated near Capu Câmpului village (47°30'48.42"N, 25°57'18.73"E) at approximately 500 m a.s.l., on a terrain with an inclination of 3-5°, eastern aspect, and had trees cover of c.a. 90%. The second point was near Izvoarele Sucevei locality (47°43'53.52"N and 25°08'49.16"E), at 1050 m a.s.l., in a terrain of 7-10° inclination with northwestern aspect. The specimens of porcine truffle have been identified close/under to the spruce trees. The investigated truffle species has an important scientific and practically role because of their importance in mycorrhizal associations with different tree species. At the establishment of spruce plantation, seedlings were brought together with this porcine truffle. Later, after a certain number of years, it produced sporocarps which emerged to soil surface through the soil cracks under favorable conditions of development.

Figure 1. Map indicating the new localities for *Choiromyces meandriformis* in Northern region of Romania

The spruce forest from Capu Câmpului was about 40 years old, while the spruce forest at Izvoarele Sucevei area was approx. 50 years old.

The sporocarp was white at the beginning, then its colour became light yellow-brown, with a scented smell. Peridia was thin, smooth, often cracked. The gleba was fleshy, soft, white at first, and then marbled in yellow-brown by sinuous bands of pedunculated axes arranged in palisades (Figure 2). They were found in soil, under coniferous trees, in July-September period. In Bulgaria it was also found at the beginning of October, in soil, in habitats with *Picea abies* and *Abies alba* in the Rhodope Mountains [LACHEVA, 2012].

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The favourable conditions for this species in Northern Romania were represented by the temperate climate, more humid during all seasons, soil characteristics and vegetation type (forest vegetation with *Picea abies* in different age stages). Soil analysis (Table 1) highlighted the species' preference for acidic soils (pH 5.06), with a medium content of humus, total nitrogen, potassium and total phosphorus, and a reduced amount of organic matter. These results in agreement (acid soil, wet site, spruce forest) with RODRIGEZ'S (2008) findings who identified the species (in Spain) also in acid and clay soils, in areas with increased rainfall, forming mycorrhizae with common oak (*Quercus robur* L.) and spruce (*Picea abies*).

Figure 2. Macroscopic aspect of analyzed specimens of *Choiromyces meandriformis* (Photo C. Bîrsan)

Table 1. Results of chemical analysis of soil samples collected in plots with *Choiromyces meandriformis*

Soil characteristic	Value	U/M	Evaluation
1. pH	5,06	-	Acid soil
2. Humus	2,89	%	Middle
3. Total Nitrogen	0,126	%	Middle
4. Phosphorus	46,0	ppm	Good
5. Accessible Potassium	83,0	ppm	Middle
6. Total Phosphorus (P ₂ O ₅)	0,205	%	Middle

7.	Total Potassium (K ₂ O)	0,187	%	Middle
8.	Organic Matter	5,70	%	Reduced
9.	Total Zinc	52,0	mg/kg	Max. val. 2000
10.	Total Copper	19,15	mg/kg	Max.val. 500
11.	Total Lead	26,0	mg/kg	Max.val. 300
12.	Total Iron	956,0	mg/kg	-

The forest habitat (Figure 3) was characterized by a uniform and species-poor floristic composition, typical to *Hieracio transsivanicus-Piceetum* Pawl. et Br.-Bl. 1939 community (Table 2). Trees layer presented high cover (90%) and was dominated by *Picea abies* while *Sorbus aucuparia*, *Fagus sylvatica*, *Betula pendula*, *Acer pseudoplatanus* were sporadically identified. The shrubs layer (with *Corylus avellana* and *Spiraea chamaedryfolia*) had very low cover (3-5%). Herbs layer was also species-poor, and had low cover (2-10%) with *Luzula luzuloides*, *Salvia glutinosa*, *Geranium robertianum*, *Euphorbia amygdaloides*, *Veronica officinalis* etc. among the most frequent species.

Figure 3. General aspect of the habitat (young *Picea abies* forest) where *Choiromyces meandriformis* was identified (Photo C. Bîrsan)

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Table 2. Phytosociological table of *Hieracio transsylvanici-Piceetum* Pawl. et Br.-Bl. 1939 forest communities where the specimens of *Choiromyces meandriformis* were identified

No. of relevé	1	2	3	4	5	6	7	
Altitude (m a.s.l.)	1050	1040	1000	1050	1040	500	510	
Aspect	NW	NW	NW	NW	NW	E	E	
Slope (°)	7	10	7	8	10	5	3	K
Trees cover (%)	90	90	85	90	90	95	90	
Shrubs cover (%)	5	3	3	5	3	5	3	
Herbs cover (%)	2	5	10	10	5	5	10	
Plot area (m ²)	500	500	500	500	500	1000	1000	
<i>Car. ass.</i>								
<i>Picea abies</i>	5	5	5	5	5	5	5	V
<i>Picea abies</i> (juv.)	1	-	+	+	+	+	-	IV
<i>Hieracium transsylvanicum</i>	-	+	-	-	+	-	-	II
<i>Vaccinio-Piceetea</i>								
<i>Luzula luzuloides</i>	+	+	-	+	+	-	-	IV
<i>Oxalis acetosella</i>	+	+	+	+	+	-	-	IV
<i>Melampyrum sylvaticum</i>	-	+	+	-	+	-	-	III
<i>Luzula sylvatica</i>	-	-	+	+	+	-	-	III
<i>Calamagrostis arundinacea</i>	-	+	+	-	-	-	-	II
<i>Sorbus aucuparia</i>	-	+	-	-	+	-	-	II
<i>Sorbus aucuparia</i> (juv.)	-	-	-	-	+	-	-	I
<i>Juniperus communis</i>	-	+	-	-	-	-	-	I
<i>Campanula abietina</i>	-	-	+	-	-	-	-	I
<i>Quercu-Fagetea</i>								
<i>Fagus sylvatica</i>	-	+	+	-	-	+	+	IV
<i>Fagus sylvatica</i> (juv.)	+	-	+	+	-	+	+	IV
<i>Geranium robertianum</i>	-	-	+	+	-	+	+	IV
<i>Spiraea chamaedryfolia</i>	+	-	+	+	-	+	-	IV
<i>Daphne mezereum</i>	-	+	+	-	+	+	-	IV
<i>Euphorbia amygdaloides</i>	-	+	-	-	+	+	+	IV
<i>Pulmonaria rubra</i>	-	+	+	-	-	-	+	III
<i>Salvia glutinosa</i>	-	-	-	+	-	+	+	III
<i>Veronica urticifolia</i>	-	-	+	+	+	-	-	III
<i>Corylus avellana</i>	-	+	-	-	-	+	+	III
<i>Dryopteris filix-mas</i>	-	-	+	+	+	-	-	III
<i>Mycelis muralis</i>	-	-	-	+	-	+	+	III
<i>Poa nemoralis</i>	-	-	-	-	-	+	+	II
<i>Acer platanoides</i> (juv.)	-	-	-	-	-	+	+	II
<i>Epilobium montanum</i>	-	-	-	+	-	-	-	I
<i>Lonicera xylosteum</i>	-	-	-	+	-	-	-	I
<i>Brachypodium sylvaticum</i>	-	-	-	-	-	-	+	I
<i>Glechoma hirsuta</i>	-	-	-	-	-	-	+	I
<i>Carex sylvatica</i>	-	-	-	-	-	+	-	I
<i>Epilobietea angustifolii</i>								
<i>Fragaria vesca</i>	-	+	-	+	-	+	+	IV
<i>Galeopsis speciosa</i>	-	-	+	-	-	-	-	I
<i>Betula pendula</i>	-	-	+	-	-	-	-	I
<i>Silene dioica</i>	-	-	-	+	-	-	-	I
<i>Tussilago farfara</i>	-	-	-	-	-	-	+	I

<i>Digitalis grandiflora</i>	-	-	+	-	-	-	-	I
	<i>Variae syntaxa</i>							
<i>Veronica officinalis</i>	+	-	-	-	+	+	+	IV
<i>Urtica dioica</i>	-	-	-	+	+	-	-	II
<i>Acer pseudoplatanus</i>	-	-	-	+	-	+	-	II
<i>Geum urbanum</i>	-	-	+	-	-	+	-	II
<i>Tilia cordata</i> (juv.)	-	-	-	-	+	-	-	I
<i>Populus tremula</i> (juv.)	+	-	-	-	-	-	-	I
<i>Prunella vulgaris</i>	-	-	-	-	-	-	+	I
<i>Pulmonaria officinalis</i>	-	-	-	-	-	-	+	I

Place and date of relevés: Izvoarele Sucevei: 10.07.2014 (rel. 1-5);

Capu Câmpului: 12.07.2013 (rel. 6-7).

In a phytosociological perspective, increased constancy presented the diagnostic species for *Piceion* alliance, *Piceetalia* order and *Vaccinio – Piceetea* class (*Melampyrum sylvaticum*, *Luzula luzuloides*, *Oxalis acetosella*, *Sorbus aucuparia* etc.). Also, the floristic composition included some diagnostic species for *Quercio – Fagetea* (*Daphne mezereum*, *Corylus avellana*, *Dryopteris filix-mas* etc.) and *Epilobietea angustifolii* (*Galeopsis speciosa*, *Fragaria vesca* etc.) classes. Most of the plant species were sciophytes, preferring the cool mountain areas or temperate sub-mountain areas with moderate humid, acid and nutrient poor soils. Canopy closure in investigated areas was high, maintaining soil moisture by not directly exposing it to sunlight. Rainfall and soil moisture were very important in investigated habitats for development of porcine truffle sporocarps.

Conclusions

This study presented new localities and highlighted similarities with other findings regarding the ecological requirements of the species *Choiromyces meandriformis*. This species produced hypogeous sporocarps in summer and autumn, in moderate acid and moist soils in some spruce forests from northern part of Romania. Unlike other truffle species, *Choiromyces meandriformis* was much more easily to be identified because a part of their sporocarps came to the surface without the need for dogs or other animals specially trained to find them. According to the Romanian literature, this species is edible with caution, while in other countries it is considered as toxic species, producing gastrointestinal disorders.

Notes on contributors

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How to cite this article:

BÎRSAN C., MARDARI C. & TĂNASE C. 2019. New localities and habitat conditions for *Choiromyces meandriformis* Vitt. (Ascomycota, Fungi) in Northern Romania. *J. Plant Develop.* **26**: 173-180. <https://doi.org/10.33628/jpd.2019.26.1.173>