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Agaricus pseudoprattensis (Agaricaceae) in the Bulgarian mycota: diversity, distribution, morphology and ecology

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Abstract

This paper summarizes the available data of *Agaricus pseudoprattensis* in Bulgaria. Review of the available previous records is made and new findings are reported. The article brings closer taxonomic profile, ecological requirements and distribution pattern of the both varieties of this species. Detailed macro- and microscopic descriptions are given. Totally 17 localities of the taxa are currently known. An UTM-grid map is appended. An identification key for the taxa is given. The fungus appeared to be widespread, but not common. New additions are included, together with a critical review of data reported in literature for the Bulgarian territory.

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Introduction

The genus *Agaricus* is a diverse, cosmopolitan genus, includes saprotrophic species occurring in a variety of ecosystems. Mushrooms in the genus have a worldwide distribution. There are c. 200 described *Agaricus* species worldwide and approximately 60 of them occurring in Europe (Cappelli, 1984, 1985; Bas, 1991; Calvo-Bado *et al.*, 1999; Kirk *et al.*, 2001, 2008). The genus *Agaricus* includes the most economically important and commercially cultivated mushroom in the world (e.g. *Agaricus bisporus*, *A. campestris*, *A. macrocarpus*, *A. nivescens* etc.), as well as some other inedible species (e.g. taxa in section *Xanthodermatei* Singer). This section comprises only inedible species, including *Agaricus pseudoprattensis* are known to possess unpleasant odours and induce gastrointestinal problems if consumed (Wasser, 1980; Cappelli, 1984, 1985; Calvo-Bado *et al.*, 2000; Lacheva, 2006).

Agaricus pseudoprattensis (Bohus) Wasser is a polymorphic species represented in Europe by two varieties (Wasser, 1980; Cappelli, 1984, 1985; Lacheva, 2006). According to Lacheva (2006) *A. pseudoprattensis* (Bohus) Wasser var. *pseudoprattensis* and *A. pseudoprattensis* var. *niveus* are distributed in Bulgaria. In the latest Bulgarian mycological literature (Denchev and Assyov, 2010), *A. pseudoprattensis* var. *niveus* not reported for the Bulgarian mycota. Our field studies confirm the two varieties for the territory of Bulgaria.

The aim of the paper is to enrich the information about *Agaricus* species and fungal diversity of the Bulgaria's mycota.

This paper provides: (1) a taxonomic revision of *Agaricus pseudoprattensis* s.l. in Bulgaria; (2) a discussion and an expanded description for this taxon; (3) a discussion of the morphological characters previously used to segregated species; (4) descriptions of basidiomata development in the field, and (5) summarize the knowledge about the distribution of these interesting fungal taxa in Bulgaria.

Materials and methods

Collection and keeping of the samples

Air-dried studied specimens of the fungus are kept in the mycological section of the Herbarium of the University of Agriculture - Plovdiv (SOA).

Distribution of the taxa is given according to the floristic regions adopted in the *Flora of the PR Bulgaria* (Jordanov, 1966). The chorological map of the occurrence of each of the species in the country, have been depicted using the program software dSOA (Stoyanov, 2003).

Macro- and microscopic processing methods

The basidiomata were identified according to the keys and descriptions published by Bohus (1939, 1971, 1980), Cappelli (1984, 1985) and Wasser (1980, 2002).

Fruiting bodies of species were photographed with SONY Cyber-shot 5.1Mpix. in standard JPEG format. Microphotographs were taken on Amplival ML. Drawings were made with the aid of a drawing tube under an oil-immersion objective. Description of morphological characters of basidiomata are based on fresh and dried specimens. Microscopic features were observed and measured in fresh and in dry fragments of tissues dehydrated in water, under a Amplival ML light microscope, using a 100× objective. Spores were examined in Melzer's reagent and were taken from the spore print for measurements. Size of spores, basidia, cheilocystidia and pleurocystidia, as well as pileipellis elements dimensions were correspondingly based on: 50, 30, 30 and 20 measurements. Drawings were made with the aid of a drawing tube under an oil-immersion objective. Data marks length and width of microstructures are processed statistically variational (Zaitsev, 1984; Lackey, 1990). Measurement values for basidiospores are presented below as follows: min– (mean±1σ) –max.; of the basidia and cheilocistidia: min-max.

Schäffer reaction was tested by aniline and 65% HNO₃ acid (Schäffer and Møller, 1938) on dried

samples. The abbreviations of the authors of fungal names follow Kirk and Ansell (2004). The taxonomic and nomenclature decisions in the article have been made in conformity with the researches of Cappelli (1984, 1985), Parra (2005), Kirk *et al.* (2008) and Index fungorum (www.speciesfungorum.org: accessed 1 March 2010).

Abbreviations are used throughout the text for the collectors' names as follows: AV - A. Vuchkov; GS - G.

Stoichev; G.P - G. Popgeorgiev; ML - M. Lacheva; OT - O. Todorov.

Results and discussion

The present study of *Agaricus pseudoprattensis* s.l. provides data for a clear delimitation of var. *pseudoprattensis* and *niveus* in the Bulgarian mycota. Their characters are described below.

Table 1. Comparison of the micrometric characteristics of *A. pseudoprattensis* var. *pseudoprattensis* according to different studies.

Author	Basidiospores (μm)	Basidia (μm)	Sterigma ta (μm)	Cheilocystidia (μm)
	1	2	3	4
Bohus (1971)	5-7 × 4-5	15-26 × 5-7	-	16-26 × 7-12
Cappelli (1984)	5-7 × 4-5	15-26 × 5-7	-	16-26 × 7-12
Wasser (2002)	5-7.5 × 3.5-5	24-31 × 7-9.5	2-3	25-36 × 8-13
Lacheva (2008)	4-5 × 3-4.2	17.5-23 × 4-6	2.5-3	16-25.5 × 5.5-10.5
This study	4.5-5.5 × 3.5-4.5	17.5-25.5 × 4.5-6.5	2.5-3	16.5-25.5 × 5.5-11.5

The review of the literature reports, the revision of the herbarium specimens, together with the unpublished collections of the author revealed 24 known records (10 for var. *pseudoprattensis* and 14 for var. *niveus*) of the taxa, corresponding to 16 UTM-Grid squares. They are all listed below and presented on Fig. 11a-b.

Description of the taxa

Agaricus pseudoprattensis

(Bohus) Wasser, Ukrayins'k. Bot. Zhurn., 33(3), 250, 1976.

Basionym

Psalliota pseudoprattensis Bohus, Borbasia, 1: 114, 1939.

Agaricus pseudoprattensis (Bohus) Bohus, Ann. Hst.-Nat. Mus. Nat. Hung., 63, 63:81, 1971.

Icons

Bohus (1939: Tab. 10), Bohus (1971: Fig. 2), Wasser (1980: Fig. 93), Cappelli (1984: Fig. 38, Pl. 68), Cappelli (1985: p. 180), Lacheva (2006: Table 45).

Pileus up to 3.5-6 cm in diameter, thick-fleshy, initially hemispherical, subsequently appanate or slightly umbonate to flat, with a hollow in the center, white, cream white, hazelnut brown to grayish brown, darker in the center, surface cracked with grayish brown scales, larger and thicker in the center. Margin initially involute, then straight, with smaller grayish brown scales, 2-5 mm thick, sometimes with fragments of the partial veil. *Pileipellis* consisting of whitish thick-walled cylindrical hyphae, with clamps, 5-10 μm in diameter. *Gills* free, thin, crowded, initially whitish pink to grayish pink, subsequently pink red to dark brown, with light, sterile edge. *Hymenophoral trama* in young carpophores initially regular, subsequently irregular, consisting of cylindrical, thin-walled hyphae, 5-10 μm in diameter. *Stipe* up to 3.5-5(-6) × 0.7-1.5 cm, central, cylindrical, often narrowed at the base, smooth to silky-fibrillose, white, whitish with a rhizomorph at the base, yellowish and later reddish at touch, staining yellowish to light reddish where cutting. *Ring* in the upper part of the stipe, sometimes in the

middle, thick, narrow, patent, often lined along the periphery, white, with or without a brownish silky-fibrillose coating, yellowish to touch. *Flesh* white, initially lemon-yellow at the stipe base when cut, subsequently red-brown to dark wine red in the whole context. *Context* in pileus and stipe quite fragile, whitish. *Smell* slight of carbolic acid or iodine. *Taste* slightly bitter. *Spores* $4.5-(4.5 \pm 0.02)-5.5 \times 3.5-(3.5 \pm 0.01)-4.5 \mu\text{m}$, ovate to ellipsoid, brown, not

ornamented, with fluorescent spots with an apical germ pore. Spore print red brown. *Basidia* $17.5-25.5 \times 4.5-6.5 \mu\text{m}$, inconspicuous clavate, hyaline, with 4 sterigmata. *Sterigmata* $2.5-3 \mu\text{m}$ long. *Cheilocystidia* $16.5-25.5 \times 5.5-11.5 \mu\text{m}$, clavate, numerous, thin-walled, hyaline. *Pleurocystidia* absent. *Macrochemical reactions*: Cross reaction with Schaeffer's reagent positive.

Table 2. Comparison of the micrometric characteristics of *A. pseudoprattensis* var. *niveus* according to different studies.

Author	Basidiospores (μm)	Basidia (μm)	Sterigmata (μm)	Cheilocystidia (μm)
	1	2	3	4
Bohus (1980)	$5.4-6.3 \times 4-4.7$	–	–	$18-22 \times 6-9$
Cappelli (1984)	$5.4-6.3 \times 4-4.7$	–	–	$18-22 \times 6-9$
Lacheva (2002)	$5.3-6 \times 3.8-5$	$15-25 \times 5-7$	3	$23.5-26 \times 9-12$
This study	$4.8-6.5 \times 4-5$	$17-30 \times 4-6.5$	3	$21.5-23.5 \times 7.5-9.5$

Habitat, ecology, phenology

Carpophores of *Agaricus pseudoprattensis* almost exclusively appears in summer to early fall season (May–October), grows solitary or gregarious, mostly in dry, open, sandy places and dunes, in gardens and parks among grassy vegetation; in plantations from *Robinia pseudoacacia* L., and *Populus nigra* Arn., as well as xerothermic or mesothermic broadleaf forests, possibly under broadleaved trees of *Acer*, *Carpinus*, *Fraxinus* and *Quercus*, 1300-1500 m. Humus saprotroph, mostly on basic or sandy soil. There is some evidence that also this species is inedible causing gastrointestinal problems (Cappelli, 1984).

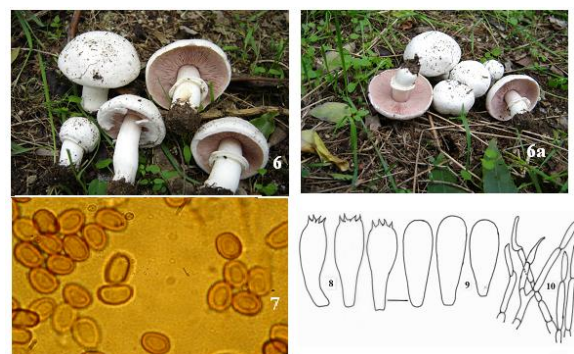


Figs. 1–5. Macro- and microscopic features of *Agaricus pseudoprattensis* var. *pseudoprattensis* from Bulgaria: 1,1a – basidiomata at different stages of

development *in situ*, 2 – basidiospores, 3 – basidia, 4 – cheilocystidia, 5 – generative hyphae of the pileipellis with terminal elements (photos and drawn by Maria Lacheva). Bar = $5 \mu\text{m}$.

General distribution

Rare species in Europe. Described from Italy (Cappelli, 1984, 1985; Lantieri *et al.*, 2009), France (Moreno and Remondo, 1999; ODONAT (Coord.) 2003; Gerault, 2005), Ukraine (Wasser 1980), Hungary (Bohus, 1939, 1971, 1980), and Bulgaria (Lacheva, 2008); In Asia the species has been recorded in Israel (Wasser, 2000, 2002) and Turkey (İşiloğlu *et al.*, 2001; Kaya, 2009b; Solak *et al.*, 2001; Solak and Yılmaz, 2002; Sesli and Denchev, 2009).



Figs 6–10. Macro- and microscopic features of *Agaricus pseudoprattensis* var. *niveus* from Bulgaria: 6,6a – basidiomata at different stages of development

in situ, 7 – basidiospores, 8 – basidia, 9 – cheilocistidia, 10 – generative hyphae of the pileipellis with terminal elements (photos and drawn by Maria Lacheva). Bar = 5µm.

Variability

var. *pseudoprattensis* (Bohus) Wasser. (Figs 1–6)

Pileus hazelnut brown to grayish brown, darker in the center, surface cracked with grayish brown scales a light base, larger and thicker in the center.

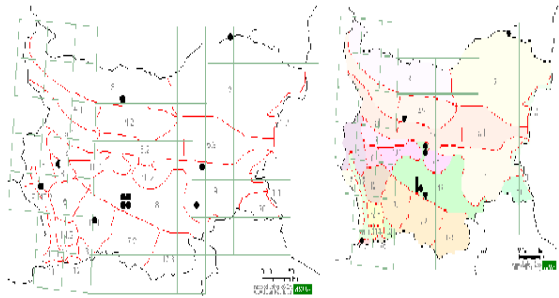


Fig. 11. Distribution of the taxa in Bulgaria: a – *Agaricus pseudoprattensis* var. *pseudoprattensis* in Bulgaria; b – *A. pseudoprattensis* var. *niveus*.

Specimens examined

Northeast Bulgaria: in the park of the Silistra town, on soil, under *Acer campestre*, 13.07.2013, coll. GP, det. ML (SOA 60 003768); Danubian Plain: Pleven town, Kailuka Park, under deciduous trees, 16 Jul 2002, coll. and det. ML (SOA 50 417); Strouma River Valley: Kyustendil distr., near Nevestino village, in a pasture with *Dichanthium ischaemum*, *Poa bulbosa*, and *Chrysopogon gryllus*, along the Struma River, 17 Jul 2004, coll. and det. ML (SOA 50 418); Western Rhodopi Mts: Pazardzhik distr., near Rakitovo village, in a meadow, 26 Sep 2002, coll. and det. ML (SOA 50 419); Thracian Lowland: Plovdiv distr., nearly Kadievo village, in a plantation of *Populus nigra*, 1 Nov 2002, coll. and det. ML, GS (SOA 50 420); Plovdiv distr., above Benkovski village, in a meadow with *Eryngium campestre*, 24 Oct 2004, coll. and det. ML (SOA 50 421); Plovdiv town, nearly Agroecological center, on soil, under *Populus nigra*, 18 Oct 2010, coll. AV, det. ML (SOA 60 00370); Plovdiv town, nearly Lauta Park, on soil, under *Populus nigra*, 10 Oct 2012, coll. and det. ML (SOA 60 00371); Toundzha Hilly Country: Sliven town, in the park of the Sliven Spa Center, under *Paliurus*

spina-christi, 10 Oct 2002, coll. and det. ML, GS (SOA 50 422); Mt Sakar, Topolovgrad distr., above Svetlina village, Roubin Kamuk locality, in a meadow, under *Populus nigra*, 10 Sep 2002, coll. and det. ML (SOA 50 423).

var. *niveus* Bohus, Ann. Hist.-Nat. Mus. Nat. Hung., 72: 96, 1980. (Figs 7–12)

Basionym: Bohus, Ann. Hst.-Nat. Mus. Nat. Hung., 72:96, 1980.

Icons: Bohus (1980: Fig. 3), Cappelli (1984: Fig. 39, Pl. 69), Cappelli (1985: p. 181), Lacheva (2006: Table 46).

Pileus up to 4–5.5 cm in diameter, snow-white, subsequently light cream, smooth, without scales in the center. *Stipe* up to 2–4 × 0.7–1 cm, cylindrical, smooth, without narrowing in the base, with or without a rhizomorph at the base. *Spores* ovate, 4.8–(5.3 ± 0.01)–6.5 × 4–(3.7 ± 0.01)–5 µm. Spore print dark brown. *Basidia* clavate, 17.0–30.0 × 4–6.5 µm. *Sterigmata* 3 µm long. *Cheilocystidia* clavate, 21.5–23.5 × 7.5–9.5 µm.

Specimens examined

Northeast Bulgaria: Silistra distr., nearly Popina village, on soil, among grasses in culture of *Populus nigra*, along the Dunav River, 16 Jul 2004, coll. and det. ML (SOA 50424); Central Stara Planina Mts: Teteven Municipality, Topilishte neighborhood nearly Glojene village, Kolyov Oreh locality, on soil, under *Populus nigra*, 28 Aug 2013, coll. and det. ML (SOA 60 00369); Belasitsa: Petrich distr., near Samuilovo village, on soil, 5 Sept 2008, coll. OT, det. ML (SOA 60 00377); Mt Sredna Gora: in the park of the Karlovo town, 23 Aug 2011, coll. and det. ML (SOA 60 00378); Karlovo Municipality, in the park of Banya town, on soil, 23 Aug 2011, coll. and det. ML (SOA 60 00374); Central Rhodopi Mts: Plovdiv distr., near Brestnik village, above monastery, on soil, under *Populus canescens* and *Cerasus avium*, 10 Oct 2011, coll. and det. ML (SOA 60 00372); Plovdiv distr., above Brestnik village, on soil, under 10 Oct 2011, coll. and det. ML (SOA 60 00373); Thracian Lowland: nearly Plovdiv town, on soil, among grasses, nearly

pump house "Sever", 30 Jul 2002, coll. and det. GS (SOA 50425); Plovdiv town, in the park of Agricultural University, on soil, under *Syringa vulgaris*, 5 Sept 2002, coll. and det. ML, GS (SOA 50426); Plovdiv town, Lauta Park, on soil, under *Populus nigra* and *Fraxinus* sp., 15 Oct 2002, coll. and det. ML (SOA 50427); Plovdiv town, Lauta Park, on soil, among grasses under *Gleditschia triacanthos*, 8 Oct 2004, coll. and det. GS, ML (SOA 50428); Plovdiv distr., above Benkovski village, on soil, among grasses in culture of *Populus nigra*, 24 Oct 2004, coll. and det. ML (SOA 50429); Plovdiv distr., nearly Krasnovo village, under *Populus nigra*, 21 Sept 2012, coll. and det. ML (SOA 60 00375); Plovdiv distr., nearly Kadievo village, in culture of *Populus nigra*, 21 Sept 2012, coll. and det. ML (SOA 60 00376).

Identification key

1 Pileus up to 3.5–6 in diameter, hazelnut brown to grayish brown, darker in the center, surface cracked with grayish brown scales; spores 4.5–5.5 μm long, 3.5–4.5 μm wide, ovate to ellipsoid var..... *pseudopratisensis*

1* Pileus up to 4–5.5 cm in diameter, snow-white to light cream, surface smooth without scales in the center; spores 4.8–6.5 μm long, 4–5 μm wide, ovate..... var. *niveus*

Note

Nearly similar fruiting bodies produces *A. campestris* L. : Fr. known to be common in Europe, which can be separated with certainty from *A. pseudopratisensis* in the cap being generally whitish or off-white, soft ring, colour of flesh when cut, macrochemical reactions as well as mainly on the base of microscopic characters and differences in habitat (terrestrial fungus occurs mainly in grasslands on clayey soil), sweet fungoid smell and taste (Bohus, 1939, 1971; Cappelli, 1984).

The taxonomic status of the *A. pseudopratisensis* var. *niveus* is formulated in various ways. For example, Wasser (1980, 2002), Parra (2005) and (Denchev and Assyov, 2010) include it in the *A. pseudopratisensis* var. *pseudopratisensis*, whereas Bohus (1980), Cappelli

(1983, 1984) and Lacheva (2006) accepted *A. pseudopratisensis* var. *niveus* as an autonomous taxon. We also support this standpoint. The both varieties differ apart among mainly in macroscopic characters. *A. pseudopratisensis* var. *niveus* can be separated with certainty from *A. pseudopratisensis* var. *pseudopratisensis* in the cap and stipe being generally snow-white, smooth, without scales, narrow and strong ring as well as and the slightly larger spores (Bohus, 1980; Cappelli, 1984; Lacheva, 2006).

The Bulgarian collection of both varieties corresponds both macro- and microscopically to the descriptions, available in the literature (Bohus, 1971, 1980; Cappelli, 1983, 1984; Gerault, 2005). Comparison of the micrometric characteristics of both varieties according to different studies are outlined in Tables 1–2. Although slightly different, the measurements of the Bulgarian specimens are close to this last figure. This difference could be probably due to the different sampling sizes – 50 basidiospores in this paper and 250 measured by Lacheva (2006, 2008), but it could be also due to simply individual variability caused by specific environmental conditions, genetic variability, etc.

According to some authors the difference between the two varieties is also given by habitat, namely Lantieri *et al.* (2009) reported *A. pseudopratisensis* var. *pseudopratisensis* in consolidated inner dunes with reforestation of *Acacia saligna* Auct. et some other authors reported the taxa on sandy places (Wasser, 1980; Cappelli, 1984) and mixed forest (Kaya, 2009). According to this investigation, the both varieties should be considered as a species developing among grassy vegetation at lower elevation, only under broadleaved trees of *Populus*, *Robinia*, *Acer*, and *Fraxinus*, possibly also under other broadleaved trees, as well as mostly on basic to neutral or sandy soil, as *A. pseudopratisensis* var. *pseudopratisensis* prefers open places, until *A. pseudopratisensis* var. *niveus* is occurring mainly on sandy soil (Cappelli, 1984, 1985).

Conclusion

The both varieties of *Agaricus pseudoprattensis* s.l., distributed in Europe, occurring in Bulgaria. So far, both varieties are reported in Bulgaria only from Danubian Plain, Strouma River Valley, Rhodopi Mts, Thracian Plain, and Toundzha hilly region Lacheva (2002, 2006, 2008). Totally 24 records, corresponding to totally 17 localities of the taxa are currently known.

Observations point out that the taxa should be searched for especially in plains and lowlands regions of the country, and its presence in other parts of Bulgaria cannot be excluded; mostly among grasses in a sandy place under *Populus nigra* Arn. or *Robinia pseudoacacia* L., and only exceptionally found in some broadleaf woodlands of *Acer* and *Fraxinus*.

Although present with a number of records in the country, *A. pseudoprattensis* var. *niveus* does not seem to be a common fungus in Bulgaria. Observations confirm it is never abundant and is usually present by single or some basidiomata in its localities. It is a rare taxa, that must be further monitored and conscientiously recorded.

The discovery of new locality of *Agaricus* species for Bulgaria, and also rare species throughout in Europe, indicating a great biodiversity that is yet to be discovered in this country moreover, the lack of specific studies in the given area.

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