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## New data about some rare and less known discomycetous fungi from Bulgaria

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### ABSTRACT

New data for six discomycetes from orders *Helotiales* and *Rhytismatales* in Bulgaria are presented herein. Three of them (*Ombrophila violacea*, *Rutstroemia bulgarioides* and *R. calopus*) are of high conservation value.

**Key words:** Ascomycota, Bulgaria, chorology, fungal conservation

## Introduction

Contemporary information on the species diversity, distribution and conservation of discomycetous fungi was collected and published during the past years, as a result of scientific research in different regions of Bulgaria (Stoykov et al., 2014, 2015; Gyosheva et al., 2016; Natcheva & Gyosheva, 2016; Velev et al., 2016).

Order *Helotiales*, usually possessing sessile or stipitate, cupulate, discoid or convex apothecial ascomata, widely known under the names inoperculate discomycetes or cup fungi, consists of 8 families. The *Rhytismatales*, commonly known as tar spot fungi, is an order of ascomycetes with 3 accepted families (Kirk et al., 2008). Its members form apothecial ascomata, immersed or rarely erumpent, usually dark-colored and opening by longitudinal or radial splits. The ascospores of the *Rhytismatales* are almost colourless, usually nonseptate, often elongate and thin.

*Ombrophila violacea* – Endangered (EN), *Rutstroemia bulgarioides* – Vulnerable (VU) and *R. calopus* – Critically Endangered (CR), all from the *Helotiales*, are of high conservation value, included in the Red list of fungi in Bulgaria (Gyosheva et al., 2006). Recently *Ombrophila violacea* and *R. calopus* are enlisted in the Red Data Book of the Republic of Bulgaria also (Dimitrova, 2015a, 2015b). *Ascocoryne turficola* (*Helotiales*) is considered a rare fungus in Europe (Gyosheva et al., 2015). *Colpoma quercinum* (*Rhytismatales*), thought to be common throughout Europe and Central Asia, is still overlooked in Bulgaria. *Rhytisma salicinum* on leaves of *Salix* is less known in Bulgaria.

The establishment and collecting of discomycetes with small ascomata is difficult, therefore the distribution and threat status of these fungi are not well studied in Bulgaria. The aim of this work is to enrich the information about the occurrence of rare and less known discomycetous fungi in our country by adding new data.

## Materials and Methods

The material, used for the present study, was recorded mostly by the authors during the last 5 years (2013-2017) within the framework of different projects. Mycological field studies were carried out using transect methods. The macromorphological features of the fungal specimens were described on the basis of fresh and air-dried materials. The color photographs were made with the aid of Canon PS A460, PS 710IS and PS A1400HD digital cameras under Boeco BM-180/T/SP, Carl Zeiss LM and Boeco BOE3500 dissecting microscope. The threat status of the species follows the Red list of fungi in Bulgaria. The studied specimens are housed in the Mycological Collection of the Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia (SOMF). The identification was confirmed after Moser (1963), Dennis (1968), Breitenbach & Kränzlin (1984), Hansen & Knudsen (2000) and Medardi (2012). Data about the ascospore measurements of *Colpoma quercinum*, obtained on dry twigs of *Quercus thracica*, are given as follows: (minimum) mean  $\pm$  1 $\sigma$  (maximum), n, where “n” - denotes the number of spores used.

## Results and Discussion

### New localities of discomycetes in Bulgaria

#### Helotiales

#### Helotiaceae

#### *Ascocoryne turficola* (Boud.) Korf. (Figure 1)

**Specimens examined:** The Rhodopes, Chairski Ezera Lakes: Golemiya Gyol and Kadirev Gyol, among peat moss, in groups and clusters at the basis of dead stems of *Carex* spp., 19.09.2017, leg. G. Gospodinov, M. Gyosheva, det. M. Gyosheva (SOMF 29757, SOMF 29758).

**Note.** The fungus is a typical inhabitant of peat bogs. It was reported and described for Bulgaria only once by Gyosheva et al. (2015) from Mt Vitoshka (between the Vetrovala ski-track and Ofeliite locality) and from Rhodopes Mts (Smolyanski Ezera lakes – on the floating peat island in Lagera Lake). The second locality of the species in the Rhodopes has been confirmed again by M. Gyosheva in September 2017. The populations of *A. turficola* are vulnerable in the known localities of Rhodopes Mts as a result of habitats changes and degradation during the last decade, connected with the climate aridization. It is necessary to provide regular monitoring of populations numbers. *Ascocoryne turficola* is with high conservation value in Europe. It should be estimated according to IUCN criteria at the next update of the Red list of fungi in Bulgaria.



**Figures 1-4:** 1. Ascoma of *A. turficola*, Rhodopes Mts, Kadirev Gyol, in peat moss (in situ); 2. Ascomata of *R. calopus*, Vitoshka Mt, near Kumata chalet, on dead herbaceous stems; 3. Ascomata of *C. quercinum*, Sofia region, on twig of *Q. thracica*; 4. Tar spots of *R. salicinum*, Vitoshka Mt, below Reznjovete, on leaves of *S. caprea*.

#### *Ombrophila violacea* (Hedw.) Fr.

**Specimens examined:** Rila Mts, near Belmeken dam-lake, a small group on decaying plant remnants, in damp place, 28.09.2016, leg. T. Nedelin, det. M. Gyosheva & D. Stoykov (SOMF 29759).

**Note.** *O. violacea* is the type species of the genus. It has been reported only once in the country, more than 40 years ago, only from two localities in Mt Vitoshka: under Bor chalet, on dead moss; and in Kominite locality, on decaying leaves of *Sorbus aria* (L.) Crantz. (Aleksandrov, 1971). Short description and illustration of the species were given by Dimitrova (2015a). Our collection is a new record for Rila Mts.

**Endangered** (EN) fungus in Bulgaria. The known localities are situated in sites of European ecological network Natura 2000 in Bulgaria (Dimitrova, 2015a).

#### Rutstroemiaceae

#### *Rutstroemia bulgarioides* (Rabenh.) P. Karst.

**Specimens examined:** Rila Mts, near Belmeken dam-lake, alt. 1930 m, gregarious on damp spruce cones, on soil, 14.06.2016, leg. T. Nedelin, det. M. Gyosheva.

**Note.** This species grows on decaying cones of conifers (*Picea abies* (L.) Karst., *Abies alba* Mill., and *Pinus sylvestris* L.). It has been published previously under the synonymous names of *Chlorociboria strobilina* (Alb. & Schwein.) Sacc., *Ombrophila strobilina* (Alb. & Schwein.) Rehm, and *Piceomphale bulgarioides* (P. Karst.) Svrček (Hinkova, 1955, 1958; Aleksandrov, 1968; Dimitrova 1996, 1999) and under the current name of *R. bulgarioides* (Denchev et al., 2006; Dimitrova & Gyosheva, 2010) from Mt Vitoshka, Eastern Rila Mts and Western and Central Rhodopes Mts. All specimens are very old, and only these published from Central Rhodopes in Dimitrova & Gyosheva (2010) are relatively recent. Short description and illustration of the species were given by Dimitrova (1996).

**Vulnerable** (VU) fungus in Bulgaria. It is necessary to provide studies on ecology and monitoring of the species in all of the localities.

#### *Rutstroemia calopus* (Fr.) Rehm. (Figure 2)

**Specimens examined:** Vitoshka region: Mt Vitoshka, near Kumata chalet, on dead herbaceous stems, alt. 1700 m, 14.08.2013, leg. R. Natcheva, det. M. Gyosheva (SOMF 29780); above Aleko chalet, by the track to Bistrishko Branishte Reserve, on decaying wet herbaceous stems, alt. 1820 m, 29.07.2017, leg. D. Stoykov, det. M. Gyosheva & D. Stoykov (SOMF 29781).

**Note.** The species was reported in Bulgaria until now only from Mt Vitoshka: near Aleko chalet, on decaying leaves of *Geum rivale* L. (Dimitrova, 1996, Dimitrova & Gyosheva, 2010) and above Edelvais chalet, on dead branches of *Carex*

*acuta* L. (Dimitrova, 2002; Dimitrova & Gyosheva, 2010). The specimens are very old, collected more than 45 years ago. Short description and illustrations of the species were given by Dimitrova (1996, 2015b).

*Critically Endangered* (CR) species in Bulgaria. The known localities are situated in sites of European ecological network Natura 2000 in Bulgaria (Dimitrova, 2015b).

### *Rhytismatales*

### *Rhytismataceae*

### *Colpoma quercinum* (Pers.: Fr.) Wallr. (Figure 3)

**Apothecia** crateriform, often regularly ellipsoidal, developed in the outer layers of plant tissues. **Asci** (85-)100-140 × 7.5-10 μm, hyaline, straight, elongate fusiform, somewhat transparent in water, 8-spored. **Ascospores** (35-) 45-55(-65) × (1-)1.5-2 μm, hyaline, straight, one-celled, needle-shaped, slightly tapering at the ends.

**Specimens examined:** Sofia region: between Tsaritschina and Tsibaovtsi villages, 16.05.2011, leg. I. Assyova, det. D. Stoykov & B. Assyov (SOMF 29181), on dead twig of oak; Sofia city, Darvenitsa quarter, in the yard of the Institute of Forest, Bulgarian Academy of Sciences, 15.06.2006, D. Stoykov, on dead twig of *Quercus thracica* Stefanov & Nedjalkov (specimen not conserved, macro- and microphotographs available); Vitosha region: Mt Vitosha, above Boyana, near the track to Boyansko Ezero lake, 42°38'27.6"N, 023°16'02.5"E, alt. 890 m, 22.07.2017, D. Stoykov (SOMF 29782), on dead twig of oak; Rila Mts: Rilomanastirska Gora Reserve, above Ilijna Reka River, 42°06'33.6"N, 023°20'30.4"E, alt. 1445 m, 02.06.2015, D. Stoykov (SOMF 29783), on twig of oak.

**Note.** According to Fakirova (1978) and the materials housed in SOMF it was known only from dry twigs of oak in Western Sredna Gora Mts (Lozenska Planina) and Stara Planina Mts, but it seems to be overlooked. Data about the ascospores' length and width, obtained in the examined material on *Q. thracica*, are as follows (33) 44.6±5.2 (55) × (1) 1.6±0.2 (2.1) μm, n=50. Usually known to inhabit living oak twigs, therefore is supposed to be an endophyte. Sometimes is reported as causing diseases of twigs and branches. It is known as the only European member of the *Rhytismatales* producing large ascomata on oak twigs. Native, so far rarely recorded species in Bulgaria.

### *Rhytisma salicinum* (Pers.: Fr.) Fr. (Figure 4)

**Tar spots** on leaves, ellipsoid to irregularly oblong, up to (10-)15-20(-22) × (5-)6-10(-11) mm in diam, black, pustulate. **Asci** (90-)95-100 × 7.5-9(-11) μm, clavate. **Paraphyses** 50-70 × (2-)3-4 μm. **Ascospores** not observed.

**Specimens examined:** on leaves of *Salix caprea* L.: Vitosha region, Mt Vitosha (in different points in Bistrishko Branishte Biosphere Reserve and in several places in

direction from Aleko chalet towards Goli Vrah lift station), Vitosha Nature Park: near Aleko chalet, near the track to Zlatnite Mostove, 42°35'01.7"N, 023°17'26.4"E, alt. 1812 m, 29.10.2016, D. Stoykov (SOMF 29790), idem., 20.11.2016, D. Stoykov (SOMF 29784); idem., on overwintered leaves, 21.04.2018, D. Stoykov (SOMF 26631); Bistrishko Branishte Biosphere Reserve: towards Bistritsa village, 42°34'40.1"N, 023°18'03.5"E, alt. 1794 m, 09.10.2016, D. Stoykov (SOMF 29792); idem., 42°35'01.8"N, 023°17'26.3"E, alt. 1811 m, 09.10.2016, D. Stoykov (SOMF 29791); idem., on overwintered leaves, 42°34'40.0"N, 23°18'02.4"E, alt. 1798 m, 21.04.2018, D. Stoykov (SOMF 26755); Bistrishko Branishte, after Reznjovete, 42°33'29.8"N, 023°17'47.9"E, alt. 1968 m, 24.09.2016, idem., below Golemiya Rezen, 42°34'03.6"N, 023°17'53"E, alt. 1898 m, 26.08.2017; 42°34'39.5"N, 023°18'04.7"E, alt. 1797 m, idem., below Reznjovete locality, 42°34'23.1"N, 023°18'01.6"E, alt. 1845 m, 26.08.2017, D. Stoykov (SOMF 29788), idem., after Golemia Rezen, 29.07.2017, D. Stoykov (SOMF 29785); below Reznjovete, along the track to Akademika chalet, 29.07.2017, alt. 1989 m (SOMF 29786); idem., alt. 1851 m, D. Stoykov (SOMF 29787), below Reznjovete, D. Stoykov, 29.07.2017 (SOMF 29789); Pirin Mts: below Banderishka Polyana, nearly 1 km away from the ski-track, 04.10.2014, leg. B. Assyov & I. Assyova (SOMF 29793).

**Note.** Obviously more common, but it seems to be under-recorded. Part of the localities in Bistrishko Branishte Biosphere Reserve are given with GPS data available, confirming the locations of *R. salicinum* during the 2016 year. The members of genus *Rhytisma* Fr.: Fr. are known to form distinctive tar spots on leaves of deciduous trees such as *Acer*, *Salix* (Kirk et al., 2008). The tar spots of *R. salicinum* are formed on living leaves attached to the trees during the summer season, observed in late July (Figure 4). Ascomata were fully ripe usually in April, after the fall, in dead leaf tissues on the snow-covered soil after the winter season. So far known from Rila and Rhodopes Mts on leaves of *Salix alba* L., *S. caprea*, *S. herbacea* L. and *S. purpurea* L. (Malkoff, 1910; Dimitrov, 1926; Klika, 1926; Hinkova, 1959; Denchev et al., 2006, 2011).

### Conclusion

New data concerning the distribution of *A. turficola* (a vulnerable fungus in peat bogs, confirmed and recorded from one more new locality in Rhodopes Mts), *O. violacea* (with new record in Rila Mts), recently recorded *Rutstroemia bulgarioides* (Rila Mts) and *R. calopus* (Mt Vitosha), *C. quercinum* (recorded for the first time in Sofia region, Vitosha and Rila Mts) and new records of *R. salicinum* (Vitosha and Pirin Mts), are presented and well documented.



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