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Pronectria gromakovae, a new lichenicolous fungus on *Lecanora populicola* and notes on other records from Kharkiv region (Ukraine)

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Recent records of lichenicolous fungi from the Kharkiv region are provided. Twenty species were reported as new to the region. *Pronectria gromakovae* is described as a new species on *Lecanora populicola*. *Didymocyrtis cladoniicola*, *D. epiphyscia*, *Epicladonia sandstedei*, *Katherinomyces cetrariae* and *Lichenoconium lichenicola* are new to the forest-steppe zone of Ukraine. *Physcia stellaris* is reported as a new host species for *Cladosporium licheniphilum*. Notes on the currently known distribution of selected species in other Ukrainian regions are provided.

Keywords: biodiversity, *Didymocyrtis*, forest-steppe zone, *Pronectria*, *Trichoconis*

In recent years the interest in lichenicolous fungi in Ukraine is increasing (Darmostuk and Khodosovtsev 2017). The lichenicolous flora in southern Ukraine and the Carpathians is well studied, and there has also been new findings from the Zhytomyr and Ternopil regions (Kapets and Kondratyuk 2019, Darmostuk and Sira 2020).

Kharkiv is one of the most important mycology historical centers in Ukraine. The research of fungi diversity in general as well as lichenicolous diversity has a long history. In early times, only isolated findings of lichenicolous fungi were presented. *Opegrapha physciaria* (= *Celidium varium* (Tul.) Körb.) is one of the few records of lichenicolous fungi which was recorded in Ukraine in the 19th century (Chernov 1895). A new era in research of lichenicolous fungi was started at the end of last century with one report of *Arthonia parietinaria* (as *Arthonia destruens* sensu ukr. auct.) from Gaidary village (Kondratyuk and Khodosovtsev 1997). Currently, only 17 species of lichenicolous fungi were reported from the Kharkiv region (Darmostuk and Khodosovtsev 2017, Gromakova 2018). Currently, 277 species of lichenicolous fungi were reported from Ukraine (Darmostuk unpubl.) instead more than 1600 species of lichens were contributed to Ukraine (Kondratyuk et al. 2010).

It is therefore useful to report lichenicolous fungi new for the region to level biodiversity knowledge gaps and understand how lichenicolous fungi are distributed in Ukraine. In this paper, I contribute twenty species as new to the region, among them *Pronectria gromakovae* described as new for science.

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Material and methods

Specimens were examined by lens (× 10) in situ and by standard microscope techniques using LOMO microscopes Optica and MICROMED-2. Microscopical examination was done in water, 10% KOH and Lugol's iodine solution, directly or after pre-treatment with KOH or Brilliant Cresyl Blue. Measurements were made in water with an accuracy of 0.5 µm for ascospores, asci, conidia, conidiogenous cells, conidiophores, ascomatal and pycnidial wall cells, and 5 µm for ascomata and pycnidia. They were made in magnifications × 640 and × 1600. Sizes are given as (min.) $x - SD - x + SD$ (max.), where x is the average and SD is the standard deviation. All examined specimens are deposited in the lichenological herbarium of Kherson State University (KHER), N.V. Karazin Kharkiv National University (CWU) and author's personal herbarium (herb. VD). Non-collected material is marked as 'non coll.'

Lichenicolous fungi were collected during field trips in 2018–2020 to Kharkiv region. Field surveys were carried out at the following locations (Fig. 1):

1. Ukraine, Kharkiv region, Zmiivs'kyi district, N of Chemuzhivka village, 49°71′20.2″N, 36°34′33.2″E, alt. 102 m a.s.l., *Pinus* forest, 10 Apr 2020, coll. V. Darmostuk.

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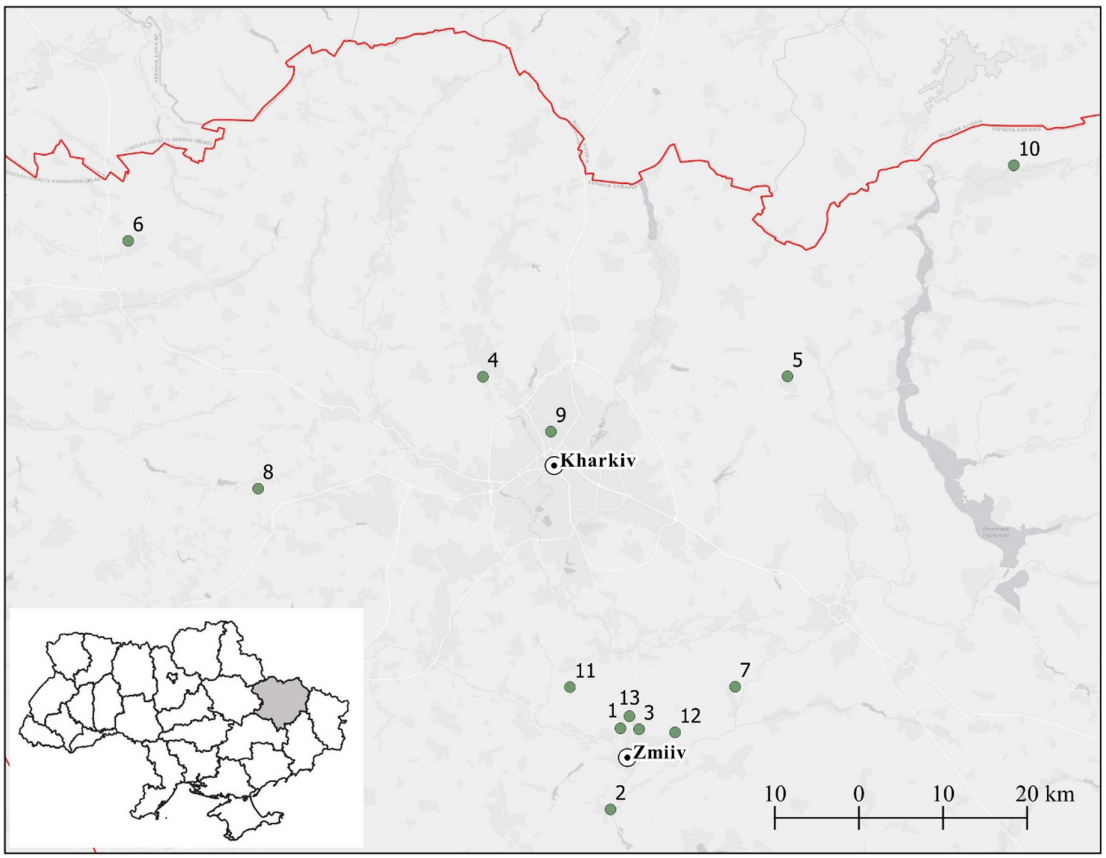


Figure 1. Map of collection sites in the Kharkiv region.

2. Ukraine, Kharkiv region, Zmiivs'kyi district, near Gaidary village, 49°62′57.4″N, 36°32′70.2″E, alt. 132 m a.s.l., *Quercus* forest, 17 Apr 2019, coll. V. Darmostuk.
3. Ukraine, Kharkiv region, Zmiivs'kyi district, N of Zmiiv town, 49°71′11″N, 36°37′4″E, alt. 119 m a.s.l., *Pinus* forest, 17 Apr 2020, coll. V. Darmostuk.
4. Ukraine, Kharkiv region, Derhachivs'kyi district, near Luzhok village, 50°08′48.4″N, 36°11′72.5″E, alt. 107 m a.s.l., 20 Jun 2019, coll. V. Darmostuk.
5. Ukraine, Kharkiv region, Vovchans'kyi district, near Shestakove village, 50°08′52.0″N, 36°61′80.1″E, alt. 150 m a.s.l., 26 Jul 2019, coll. V. Darmostuk.
6. Ukraine, Kharkiv region, Bohodukhivs'kyi district, near Zariabene village, 50°22′81.2″N, 35°53′38.8″E, alt. 177 m a.s.l., 6 Jul 2019, coll. V. Darmostuk.
7. Ukraine, Kharkiv region, Zmiivs'kyi district, near Mokhnach village, 49°75′63″N, 36°53′19.1″E, alt. 166 m a.s.l., 2 May 2020, coll. V. Darmostuk & A. Gromakova.
8. Ukraine, Kharkiv region, Valkivs'kyi district, near Staryi Merchyk village, 49°96′65.4″N, 35°74′75.7″E, alt. 173 m a.s.l., 4 Jul 2019, coll. V. Darmostuk.
9. Ukraine, Kharkiv region, Kharkivs'ka city council, Sarzhyn Ravine, 50°02′68″N, 36°22′89.6″E, alt. 127 m a.s.l., 8 Apr 2019, coll. V. Darmostuk.
10. Ukraine. Kharkiv region, Vovchans'kyi district, near Vovchansk city, 50°30′76.4″N, 36°99′01.9″E, alt. 145 m a.s.l., 6 Apr 2019, coll. V. Darmostuk & A. Gromakova.
11. Ukraine, Kharkiv region, Zmiivs'kyi district, near Konstantynivka village, 49°75′59.2″N, 36°26′03.3″E, alt. 135 m a.s.l., 13 May 2020, coll. V. Darmostuk.
12. Ukraine, Kharkiv region, Zmiivs'kyi district, near Lazukivka village, 49°70′77.7″N, 36°43′31.6″E, alt. 127 m a.s.l., 21 Jun 2020, coll. V. Darmostuk.
13. Ukraine, Kharkiv region, Zmiivs'kyi district, near Aksiutovka village, 49°72′49.9″N, 36°35′80.5″E, alt. 130 m a.s.l., 28 Apr 2020, coll. V. Darmostuk.

Results and discussion

Arthonia apotheciorum (A. Massal.) Almq.

Site 8: on *Polyozosia albescens* on concrete (non coll.).

Cladosporium licheniphilum Heuchert & U. Braun

Specimens examined: Site 3: on *Physcia stellaris* on *Quercus robur* bark (herb. VD 778); Site 12: on *Xanthoria parietina* on *Pyrus* bark (herb. VD 768).

This species was reported from a few localities, but is probably overlooked elsewhere. It was known from Mykolaiv, Kherson and Ternopil region (Khodosovtsev and Darmostuk 2016, Khodosovtsev et al. 2019a, Darmostuk and Sira 2020). *Physcia stellaris* is a new host species.

Didymocyrtis cladoniicola (Diederich, Kocourk. & Etayo) Ertz & Diederich

Specimens examined: Site 1: on *Cladonia uncialis* on sand (herb. VD 460); Site 13: on *Cladonia* sp. on soil (herb. VD 775).

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This species was reported only from the steppe zone of Ukraine (Darmostuk and Khodosovtsev 2017, Khodosovtsev et al. 2019b). This record is new for the forest-steppe zone.

Didymocyrtis epiphyscia Ertz & Diederich

Specimens examined: Site 2: on *Physcia aipolia* on *Quercus robur* (CWU 202971), on *Xanthoria parietina* on *Salix* (CWU 202975); Site 4: *X. parietina* on *Pyrus* twig (CWU 203005).

This lichenicolous fungus is a poorly known species in Ukraine. It was reported only from Kherson region and Autonomous Republic Crimea (Darmostuk and Khodosovtsev 2017). Probably, *Didymocyrtis epiphyscia* is a common but rarely collected species in Ukraine.

Didymocyrtis foliaceiphila (Diederich, Kocourk. & Etayo) Ertz & Diederich

Specimen examined: Site 3: on *Cladonia mitis* on sand (herb. VD 497).

Recently, *Didymocyrtis foliaceiphila* was reported from Ternopil region on *Punctelia subrudecta* (Darmostuk and Sira 2020).

Epicladonia sandstedei (Zopf) D. Hawksw.

Specimens examined: Site 1: on *Cladonia rangiformis* on sand (herb. VD 769); Site 13: on sand (herb. VD 773).

In Ukraine, *Epicladonia sandstedei* was known from Kherson and Zakarpattia regions (Khodosovtsev and Darmostuk 2016, Khodosovtsev et al. 2018, Darmostuk et al. 2020).

Homostegia piggotii (Berk. & Broome) P. Karst.

Specimen examined: Site 5: on *Parmelia sulcata* on *Quercus robur* (herb. VD 366).

This species was reported from a few localities in different regions of Ukraine. In the forest-steppe zone, *Homostegia piggotii* was found in Poltava region (Darmostuk et al. 2017).

Katherinomyces cetrariae Khodos.

Specimen examined: Site 1: on *Cetraria aculeata* on sand (herb. VD 461).

Katherinomyces cetrariae was described on *Cetraria aculeata* from Kherson region (Khodosovtsev et al. 2016). Further research has shown that the species is not host specific and can infect other Parmeliaceae species as well as *Lecidea fuscoatra* and *Rhizoplaca chrysoleuca* (Darmostuk and Khodosovtsev 2019, Zhurbenko et al. 2020).

Illosporiopsis christiansenii (B. L. Brady & D. Hawksw.) D. Hawksw.

Specimens examined: Site 2: on *Physcia tenella* on *Quercus robur* (CWU 202969), on *P. sulcata* on *Q. robur* (CWU 202973); Site 3: on *Physcia adscendens* on *Acer* (non coll.); Site 6: on *P. adscendens* on *Acer* (non coll.); Site 7: on *P. adscendens* on *Q. robur* (non coll.); Site 8: on *P. adscendens* on *Fraxinus* (non coll.).

Illosporiopsis christiansenii is a common species in Ukraine, but there is no previous report from Kharkiv region (Darmostuk and Khodosovtsev 2017, Khodosovtsev and Darmostuk 2017b, Darmostuk and Sira 2020).

Laetisaria lichenicola Diederich, Lawrey & Van den Broeck

Specimen examined: Site 8: on *Physcia adscendens* on *Quercus* twig (non coll.).

This species is rarely collected but probably overlooked (Darmostuk and Khodosovtsev 2017, Khodosovtsev and Darmostuk 2017b, Darmostuk and Sira 2020).

Lichenochora obscuroides (Linds.) Triebel & Rambold

Specimens examined (all on *Phaeophyscia orbicularis*): Site 2: on *Populus* (CWU 202980, 2028487); Site 3: on *Acer* (herb. VD 476); Site 6: on *Populus* (non coll.); Site 9: on *Fraxinus* (herb. VD 119); Site 11: on *Populus* (herb. VD 767).

Lichenochora obscuroides probably is a common species within the forest-steppe zone of Ukraine (Darmostuk and Khodosovtsev 2017, Pleskach and Kondratyuk 2019).

Lichenoconium erodens M. S. Christ. & D. Hawksw

Specimens examined: Site 2: on *Evernia prunastri* on *Quercus* (CWU 202841); Site 3: on *Parmelia sulcata* on *Acer* bark (herb. VD 674); Site 5: on *P. sulcata* on *Q. robur* (herb. VD 361); Site 13: on *P. sulcata* on *Q. robur* (herb. VD 777).

Lichenoconium lecanorae (Jaap) D. Hawksw.

Specimen examined: Site 6: on *Lecanora carpinea* on *Q. robur* (herb. VD 200).

Lichenoconium lichenicola (P. Karst.) Petr. & Syd.

Specimen examined: Site 2: on *Physcia tenella* on *Q. robur* twig (herb. VD 118).

This coelomycetes fungus may be overlooked and not frequently collected. Confirmed records are known only from Kherson and Mykolaiv region (Darmostuk 2019).

Lichenoconium xanthoriae M.S. Christ.

Specimens examined (all on *Massjukiella polycarpa*): Site 2: on *Prunus* (CWU 202981); Site 3: *Pyrus* (herb. VD 776); Site 5: on *Betula* (non coll.); Site 10: on *Crategus* bark (herb. VD 123); Site 13: on *Xanthoria parietina* on *Acer* twig (herb. VD 771).

This is a common species in Ukraine (Darmostuk 2019).

Pronectria diplococca Kocourk., Khodos., Naumovich, Vondrák & Motiej.

Specimens examined: Site 12: on *Enchylium tenax* on soil (herb. VD 676); Site 13: on *Enchylium* sp. on soil (herb. VD 774).

This rarely collected species was described on *Collema* sp. from southern Ukraine (Khodosovtsev et al. 2012).

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In forest-steppe zone this species was reported only from Poltava region (Darmostuk et al. 2017).

Pronectria gromakovae Darmostuk sp. nov. (Fig. 2)

MycoBank MB 838716

Type: Ukraine, Kharkiv region, Bohodukhivskyi district, near Zariabene village, small *Populus* forest, 50°23′12.7″N, 35°53′16.9″E, alt. 195 m a.s.l., on thallus and apothecia margins of *Lecanora populicola*, on *Populus tremula* bark, 8 Jul 2019, V. Darmostuk (Holotypus KHER 14203, Isotypus herb. VD 195).

Diagnosis

Morphologically similar to *Pronectria leptaleae*, but differing in the larger asci (60–)65–75(–80) × (11.4–)11.8–13.2(–14.0) µm, larger ascospores (10.4–)11.5–15.0(–18.2) × (5.8–)7.4–10.2(–12.6) µm and different host (*Lecanora* versus *Physcia*).

Description

Ascomata perithecia, numerous, immersed in discoloured host thallus and apothecia, globose to pyriform, 180–200 µm in diam, orange-brown to reddish-brown. Perithecial apex distinctly visible on host surface, ± reddish-brown, urceolate to flat, (30–)40–50(–65) µm diam. [n = 20]. Perithecial wall (14.2–)15.6–18.2(–19.6) µm wide [n = 15] in its medium and lower part, up to 30 µm in the upper part, rose to light orange to almost colourless, homogeneous, composed typically of 6–8 rows of cells, K–, with scattered orange oil droplets. Cells fusiform, thin-walled, rectangular, (3.6–)6.2–8.8(–11.2) × (1.6–)2.4–2.8(–3.2) µm [n = 20]. Paraphyses absent. Periphyses (8–)10–12(–14) long [n = 10] and ca 1.5 µm wide. Asci unitunicate, 8-spored, cylindrical to subclavate, (60.0–)65.0–75.0(–80.0) × (11.4–)11.8–13.2(–14.0) µm [n = 15], with apical ring. Ascospores uniseriate to biseriate, hyaline, (0)–1 septate, broadly ellipsoid to subglobose, slightly constricted at septa, with rounded apex, mostly smooth, only a few observed ascospores were slightly verrucose, with one oil drop, (10.4–)11.5–15.0(–18.2) × (5.8–)7.4–10.2(–12.6) µm [n = 50], the ratio of ascospore length/width (1.2–)1.3–1.7(–2.0). Conidiomata not observed.

Etymology

The epithet honours the Ukrainian lichenologist Dr Alla Gromakova at the occasion of her 55th birthday.

Ecology and distribution

The new species is known from two localities in Kharkiv region, where it grows on the thalli of *Lecanora populicola* on *Populus tremula*. The infection does not induce gall formation, but causes a discoloration of the host thallus and apothecia.

Notes

The genus *Pronectria* in strict since is characterized by orange, immersed perithecia without setae, mostly 1-septate ascospores and K– perithecium wall (Rossman et al. 1999, Diederich 2003). Including 44 species, *Pronectria* is one of the genera with the largest number of described lichenico-

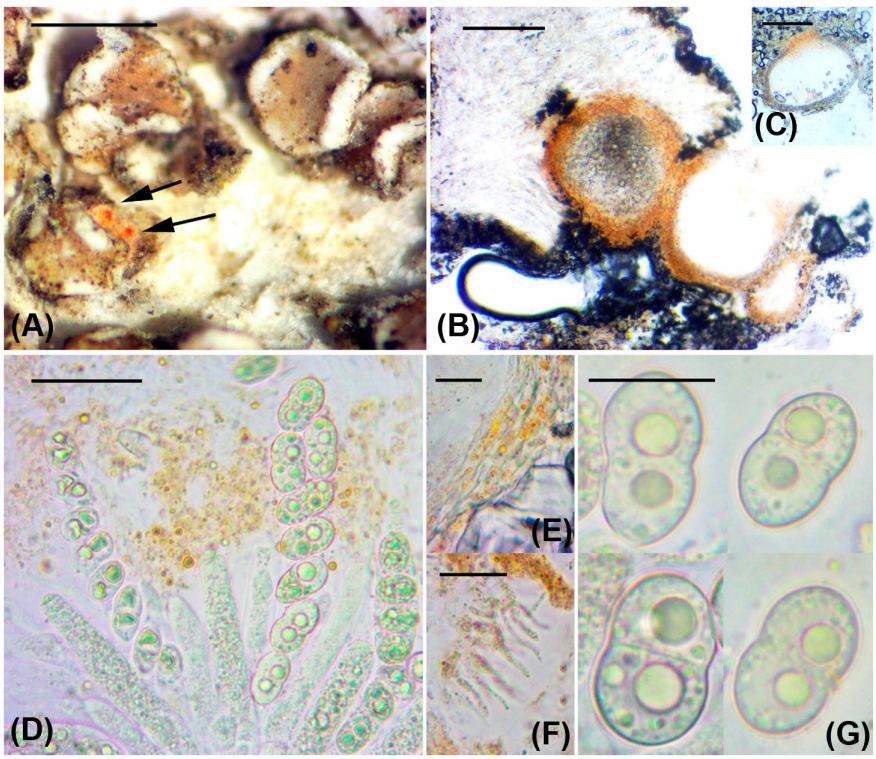


Figure 2. *Pronectria gromakovae* (holotype). (A) Ascomata immersed in apothecia of *Lecanora populicola* (arrow), (B–C) cross-section of the ascomata, (D) ascus with ascospores, (E) cross-section of the ascomatal wall, (F) periphyses, (G) ascospores. Scale bar: (A) 1 mm, (B) 100 µm, (C) 50 µm, (D) 25 µm, (E–G) 10 µm.

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lous fungi (Diederich et al. 2018). All *Pronectria* species are more or less specific to one host species or genus. No *Pronectria* species has been described on *Lecanoraceae*. Several *Pronectria* species with broadly ellipsoid to subglobose ascospores have to be compared with the new species. *Pronectria diplococca* has similar ascomata size and ascospores shape, but differs in having much smaller asci ((30.0–)34.0 ± 2.0(–37.0) × (4.0–)6.0 ± 1.6(–9.0) µm versus (60–)65–75(–80) × (11.4–)11.8–13.2(–14.0) µm in *P. gromakovae*), smaller disintegrating ascospores [(8.75–)10.95 ± 1.39(–13.50) × (4.75–)6.66 ± 0.90(–8.75) µm versus (10.4–)11.5–15.0(–18.2) × (5.8–)7.4–10.2(–12.6) µm in *P. gromakovae*) and different host (Collemataceae versus Lecanoraceae) (Khodosovtsev et al. 2012)]. The new species is also closely related to *Pronectria leptaleae* has similar ascomata and asci, but can be distinguished by smaller ascospores (6.0–11.5 × 5.5–6.5 µm in type description versus (10.4–)11.5–15.0(–18.2) × (5.8–)7.4–10.2(–12.6) µm in *Pronectria gromakovae*), narrower asci (8.0–11.5 µm versus (11.4–)11.8–13.2(–14.0) µm in *Pronectria gromakovae*) and *Physcia* as the host (Steiner 1900, Rossman et al. 1999, Berger et al. 2020). Within this study, we compare the ascospore length of two *Pronectria gromakovae* specimens (KHER 14192, herb. VD 766) with three specimens of *P. leptaleae* (KHER 10280, 10322, herb. VD 700). All three speciemens of *P. leptaleae* had significantly shorter ascospores than both specimens of *P. gromakovae* (Fig. 3).

Pronectria biglobosa Etayo has smaller ascomata (100–150 µm versus 180–200 µm in diam in *Pronectria gromakovae*), slightly narrower ascospore strongly constricted at the septa [(5.0–)6.0–6.5(–7.5) µm versus (5.8–)7.4–10.2(–12.6) µm in *Pronectria gromakovae*) and a different host (*Hypotrachyna* versus *Lecanora*) (Etayo 2017)]. Lastly, the new species is similar to the recently described *Pronectria etayoi* E. Zimm. & F. Berger which differs in the larger ascomata (200–300 µm versus 180–200 µm in diam in *Pronectria gromakovae*), distinctly echinulate and larger ascospores that are not constricted at the septum (versus mostly smooth, constricted at the septum in *P. gromakovae*) and the different host (*Physcia* versus *Lecanora*) (Berger et al. 2020).

Additional examined specimens

Pronectria gromakovii (paratype): Ukraine, Kharkiv region, Zmiivs'kyi district, near Konstantynivka village, 49°75′59.2″N, 36°26′03.3″E, 13 May 2020, coll. V. Darmostuk (herb. VD 766).

Pronectria leptaleae s.s. (all on *Physcia stellaris*): Ukraine, Mykolaiv region, Voznesens'kyi district, near Trykraty village, 47°70′65.5″N, 31°40′90.3″E, on *Quercus robur* twig, 21 Oct 2016, V. Darmostuk, A. Khodosovtsev (KHER 10322); Brats'kyi district, near Vysoka Hora village, Dubova Balka Lendmark, 47°88′95.5″N, 31°61′29.8″E, on *Acer campestre* twig, 21 Oct 2020, V. Darmostuk (herb. VD 700); Sumy region, Seredyno-Buds'kyi district, Desniansko-Starogutsky National Nature Park, near Ochkino village, 52°26′22.5″N, 33°38′92.7″E, on *Salix* twig, 1 Aug 2016, V. Darmostuk, A. Khodosovtsev (KHER 10280).

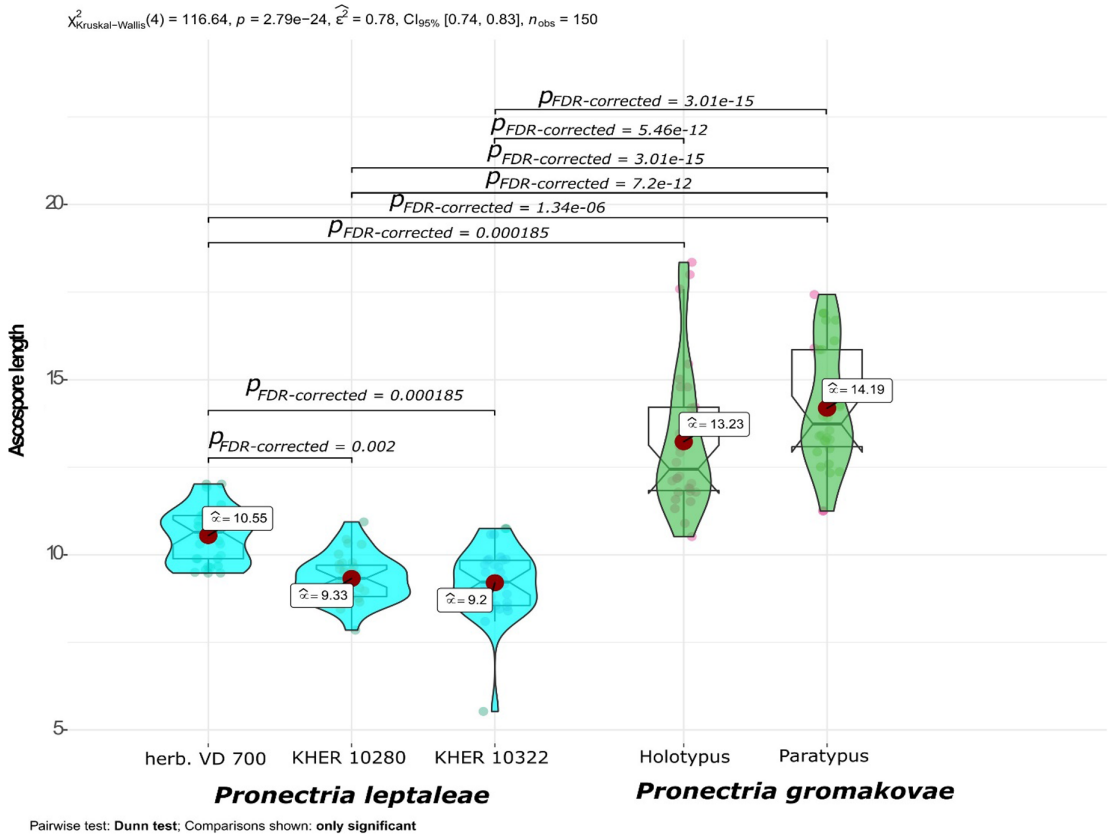


Figure 3. Differences in ascospores length of *Pronectria gromakovii* and *P. leptaleae* specimens.

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Pyrenochaeta xanthoriae Diederich,

Specimens examined: Site 2: on *Xanthoria parietina* on *Populus tremula* bark (CWU 202848, 203002).

Pyrenochaeta xanthoriae is a common fungus dwelling on *Xanthoria*, which was reported from a few localities in Ukraine (Darmostuk and Khodosovtsev 2017, Kapets and Kondratyuk 2019, Khodosovtsev et al. 2019a).

Refractohilum intermedium Cl. Roux & Etayo

Specimen examined: Site 2: on *Gyalecta fagicola* on *Populus tremula* bark (CWU 203446).

Refractohilum intermedium is a rare species growing on *Pachyphiale* species (Roux et al. 1997). In the forest-steppe zone of Ukraine it was reported from Sumy and Ternopil regions (Khodosovtsev and Darmostuk 2017a, Darmostuk and Sira 2020).

Taeniolella phaeophysciae D. Hawksw.

Specimen examined: Site 2: on *Phaeophyscia orbicularis* on *Acer* bark (CWU 2028486).

Trichoconis hafellneri U.Braun, Khodos., Darmostuk & Diederich

Specimen examined: Site 2: on *Xanthoria parietina* on *Q. robur* bark (CWU 2028481).

Trichoconis hafellneri was described from Kherson and Poltava region on thallus and apothecia of *Athallia pyracea* and *Xanthoria parietina* (Braun et al. 2016). Currently, only records from Austria are known in addition to the type localities (Berger 2019).

Conclusion

Including the 21 species presented in this study, in total 38 species of lichenicolous fungi are currently known in the Kharkiv region. *Pronectria gromakovae* is described as new for science on thallus and apothecia margin of *Lecanora populicola*. It is the first *Pronectria* species described from Lecanoraceae. *Didymocyrtis cladoniicola*, *D. epiphyscia, Epicladonia sandstedei*, *Katherinomyces cetrariae* and *Lichenoconium lichenicola* are new to the forest-steppe zone of Ukraine. Currently, the Kharkiv region harbours the greatest diversity of lichenicolous fungi in the Left Bank Forest-Steppe of Ukraine. However, we suggest that the 38 species currently known are only a fraction of all species present, and further studies on lichenicolous fungi in the Kharkiv region will likely result in more new findings.

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