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Biogeography and taxonomy of pyrenomycetous fungi 3. The area around the Sea of Japan

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Abstract — The peculiar species composition of the assemblage of pyrenomycetous fungi known from the area around the Sea of Japan is discussed. Three new species—*Annulohypoxylon orientale*, *Hypoxylon cyanescens*, and *Nectria araliae*—are described and illustrated.

Key words — *Ascomycota*, northeastern Asia, ecological distribution

Introduction

One of the most interesting regions of northeastern Asia is the area around the Sea of Japan. Many pyrenomycetous fungi are restricted to this region of the world, and a number of economically important pathogenic fungi are known only from northeastern China, Japan, Korea and southeastern Russia. Unfortunately, often many of these fungi have been confused with other species, so phytopathologists cannot recognize them and thus develop the appropriate control measures.

At the 2011 Asian Mycological Congress held in Incheon (Korea), Dr. Tsuyoshi Hosoya—during his talk on the reassessment of *Hyaloscyphaceae* (*Helotiales*, *Leotiomycetes*) based on molecular phylogenetic analysis—repeated Dr. Richard Korf's statement that "for the discosystematist, Japan is a relatively unexplored paradise" and that the region is "rich in apparently endemic forms, an understanding of which is essential for phylogenetic and phytogeographic studies" (Korf 1958: 7). However, it seems that not only Japan but rather the entire area around the Sea of Japan is the real 'paradise' for many groups of fungi.

Materials & methods

The specimens reported herein are deposited in the Herbarium of Institute of Biology and Soil Science, Far East Branch of the Russian Academy of Sciences (VLA) and the Herbarium of Cryptogams of the Kunming Institute of Botany, Chinese Academy of Sciences (HKAS). Photographs of ascomata were obtained using a Nikon D40x digital camera and S70 Canon camera. The photographs of ascospores were taken by using a VHX-600E microscope of the Keyence Corporation. External stromatal colors were recorded and coded according to Rayner (1970).

Some pathogenic and nonpathogenic pyrenomycetous fungi restricted to the area around the Sea of Japan

One pathogen restricted to the Sea of Japan area is *Venturia nashicola* S. Tanaka & S. Yamam., the scab fungus of Japanese and Chinese pears. Soon after Tanaka & Yamamoto (1964) described this species, its name was reduced to a synonym of *V. pirina* Aderh., which causes a similar disease in European pears (Sivanesan 1977). Although Khokhryakova (1978) had already discussed that *V. nashicola* has shorter ascospores and conidia than *V. pirina*, her studies and special references to their biogeographical differences (Khokhryakova 1978, 1980) were initially ignored, and only recently have her findings been supported by the data obtained by other researchers (Ishii & Yanase 2000). The differences in ascospore size between *V. nashicola* and *V. pirina* (on *Pyrus*) are exactly the same as between *V. chlorospora* (Ces.) P. Karst. and *V. subcutanea* Dearn. (on *Salix*) or *V. canadensis* M.E. Barr and *V. rumicis* (Desm.) G. Winter (on *Rumex*), and if the latter species pairs are accepted as independent (Barr 1968), there seems little reason to unite *V. nashicola* and *V. pirina*.

Another pathogen restricted to the Sea of Japan area is *Polystigma ussuriense* (Jacz. & Natalyina) Prots., first described from the Russian Far East (Natalyina 1931) but not acknowledged until after almost a half century by Khokhryakova (1978, 1980). This species was re-examined even later (Cannon 1996) and accepted as only a subspecies of *P. rubrum* (Pers.) DC. However, even at the level of subspecies the Sea of Japan area has its own taxon. The ascospores and conidia of *P. ussuriense* are longer than those of *P. rubrum* — the same characters separating *Venturia nashicola* and *V. pirina*. Although the same characters have unequal value in different (especially remote) genera, size differences in ascospores and conidia almost universally discriminate species in numerous pyrenomycetous groups (cf. Rappaz 1987; Rossman et al. 1999; Gryzenhout et al. 2009; Jaklitsch 2011). Consequently, *P. ussuriense* might be considered as an independent species occurring in northeastern Asia and probably replaces *P. rubrum* in this area, although the *P. rubrum* has been reported from China, Japan, and Korea (Cannon 1996; Katumoto 2010).

Another example, *Valsa mali* Miyabe & G. Yamada, is a very aggressive pathogen that causes canker of apple trees in various countries surrounding the Sea of Japan. Described more than a century ago (Ideta 1911), *V. mali* was confused with other species during the past century, most often with *V. ceratosperma* (Tode) Maire (e.g., Kobayashi 1970; Otani 1995), although the two species can be easily distinguished superficially (Fig. 1). The arrangement of ostioles in *V. mali* as a ring around the margin of an ectostromatic disc is characteristic of *Valsa* sect. *Circinatae* (Nitschke 1867; Urban 1957) or sect. *Valsa* (Spielman 1985). The character of an ectostromatic disc filled with closely packed ostioles places *V. ceratosperma* in section *Monostichae*.

The independent status of *V. mali* began to be discussed only relatively recently (Vasilyeva & Kim 2000), although its anamorph, *Cytospora orientis-extremi* Gvrit., had been described (with reference to *V. mali* as the teleomorph) as a separate species even earlier (Gvritishvili 1973). However, the first molecular study to consider *V. mali* (Adams et al. 2002) suggested that it be combined with *Leucostoma persoonii* (Nitschke) Höhn., since the two species clustered together on a phylogenetic tree. Although they look completely different superficially (Fig. 1), the clustering is actually not surprising, given that these two species share a number of similarities (e.g., the kind and size of ascospores, stromatal shape, *Cytospora* anamorphs, restriction to rosaceous hosts). The problem is that those same similarities comprise a repetitive internal polymorphism within *Valsa* and *Leucostoma* and outweigh the few differences that exist between them. When molecular trees are constructed based on phenetic principles, thus relying upon the quantity of shared characters without discriminating their level in the taxonomic hierarchy, such mistakes in the unification of species from different genera are potentially common (Vasilyeva & Stephenson 2010a). A more recent sequence analysis by Wang et al. (2011)

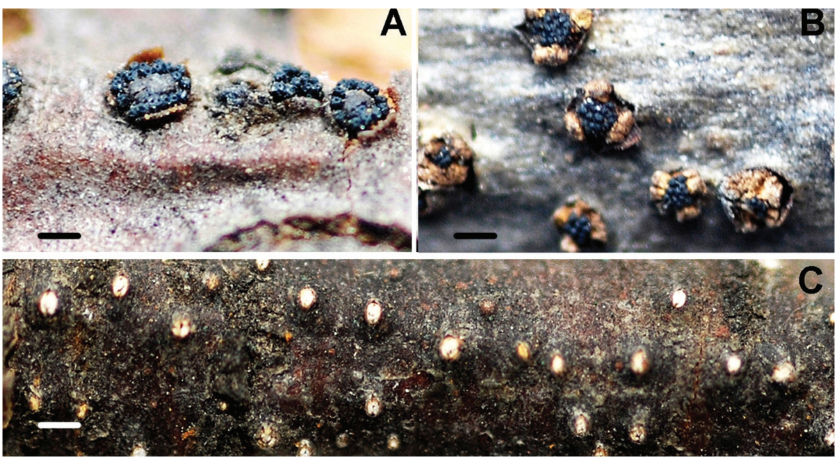

Fig. 1. Stromata. A. *Valsa mali* (VLA P–1981); B. *Valsa ceratosperma* (VLA P–1845); C. *Leucostoma persoonii* (VLA P–230). Scale bars: A, C = 0.5 mm; B = 0.3 mm.

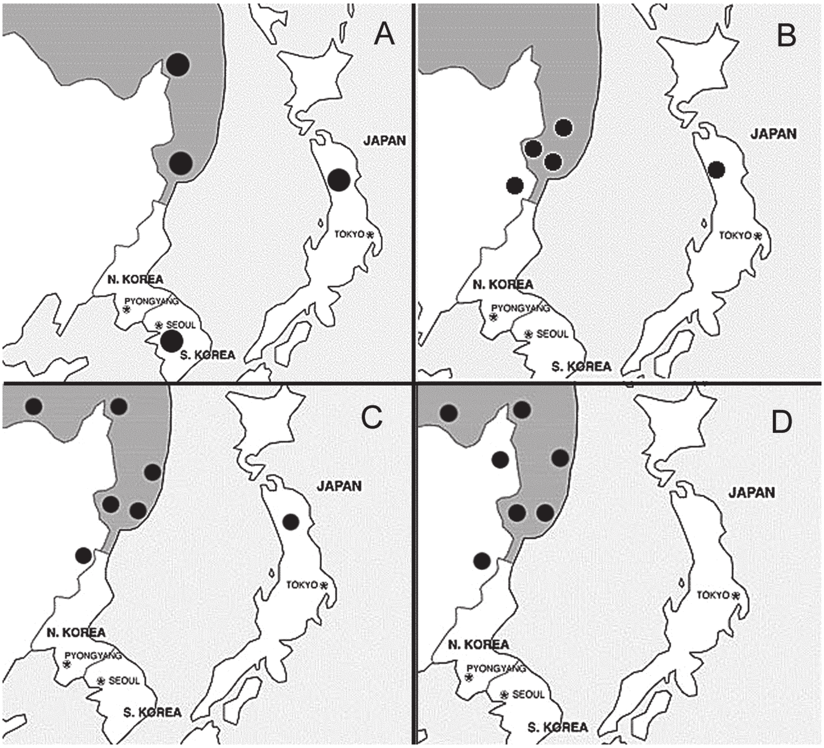

Fig. 2. Known localities of: A. *Biscogniauxia maritima*; B. *Biscogniauxia mandshurica*; C. *Cryptosphaeria venusta*; D. *Cryptosphaeria exornata*. (The map, taken from http://www.fas.org, has been modified to show our data).

supports *V. mali* as independent, and its possible distribution in northeastern Asia might be shared with a number of non-pathogenic pyrenomycetous fungi.

Some species described originally from the Russian Far East (Vasilyeva 1998) — *Biscogniauxia mandshurica* Lar.N. Vassiljeva, *B. maritima* Lar.N. Vassiljeva, *Cryptosphaeria exornata* Lar.N. Vassiljeva, *C. venusta* Lar.N. Vassiljeva — were later found in China, Korea or Japan; since their English descriptions and illustrations of stromata have been provided elsewhere (Vasilyeva et al. 2009), only their known localities are presented here (Fig. 2). *Diaporthella corylina* Lar.N. Vassiljeva is known from only two localities in China and Russia, and *Leucodiaporthe acerina* M.E. Barr & Lar.N. Vassiljeva from only two localities in Korea and Russia (Vasilyeva et al. 2007). *Podostroma giganteum* S. Imai, described from Japan (Imai 1932), was reported much later for the Russian Far East.

Among the species reported previously from northeastern Asia (Tsuneda 1982; Tsuneda & Arita 1984; Vasilyeva 1998) in need of taxonomic reconsideration is *Hypoxylon truncatum* (Schwein.) J.H. Mill. As can be seen from Fig. 3, the stromata of '*H. truncatum*' from the Russian Far East surely differ from those observed for *H. truncatum* specimens from the eastern United States; hence we assign the Russian specimens to *Annulohypoxylon orientale*, a new species described herein. In addition, Vasilyeva (1998) reported *Nectria coryli* Fuckel from the southern Russian Far East but emphasized that it had never been found on either of the two *Corylus* species (*C. heterophylla* Fisch. ex Trautv., *C. mandshurica* Maxim.) that are widely distributed in the region. Instead, the fungus was restricted to the *Araliaceae*—mostly to *Aralia elata* (Miq.) Seem. and *Eleutherococcus senticosus* (Rupr. & Maxim.) Maxim.—and was characterized by the presence of black ascomata similar to those of *Nectria austroamericana* (Speg.) Cooke (Samuels et al. 2006), a marked contrast to the red ascomata in *N. coryli*. Ascospores of *Nectria* species from the Sea of Japan area appear even smaller than those characteristic of *N. coryli*. Therefore, on biogeographical, ecological, and morphological grounds, the former warrants being described as the new species *N. araliae*.

The specimens of the third new species described in this paper were repeatedly found in the southern Russian Far East, mostly on *Ligustrina amurensis* (Rupr.) Rupr. (*Oleaceae*), which is distributed throughout northeastern China, Korea, and the Russian Far East. However, the new species was not described until the same fungus was collected in Jilin Province (northeastern China), as discussed in the Ph.D. dissertation by Haixia Ma (Ma 2011), where it was listed as '*Hypoxylon aeneo-viridis*'; herein it is formally named as *H. cyanescens*.

Taxonomy

Annulohypoxylon orientale Lar.N. Vassiljeva & S.L. Stephenson, **sp. nov.** Fig. 3B

MycoBank MB 802460

A Annulohypoxylone truncato *in disco ostiolato plano ejusdem typi ac* A. bovei *differt.*

Type: Russia: Primorsky Territory, Khasan District, Trinity Bay, on dead branches of *Quercus mongolica* Fisch. ex Lebed. (*Fagaceae*), 3 Nov. 1995, L. Vasilyeva (**Holotype** VLA P–382).

Etymology: The epithet refers to the eastern part of Asia where this species was collected.

Stromata solitary to densely aggregated, semi-globose, 2–4 mm diam., 1.3–1.5 mm high, with perithecial mounds ½–¼ exposed, surface initially brown with shining-black ostiolar caps, then becoming blackened, with KOH-extractable pigments olivaceous (48); the tissue below the perithecial layer blackish. Perithecia spherical, 0.25–0.35 mm diam., ostioles finely papillate, encircled with a flat, *bovei*-type disc 0.2–0.25 mm diam. Asci in the spore-bearing

Fig. 3. Stromata. A. *Annulohypoxylon truncatum* from Texas, USA (VLA P-2446); B. *Annulohypoxylon orientale* (VLA P-381). Scale bars: A = 1.2 mm; B = 2.7 mm. (Photo E.M. Bulakh).

portion 70–80 × 4.5–5.5 µm, stipitate, stipes 30–50 µm long, with an apical ring bluing in Melzer's iodine reagent, discoid, about 1 µm broad. Ascospores brown, unicellular, ellipsoid-inequilateral, with narrowly rounded ends, 8–10 × 4–4.5(–5) µm, with a straight germ slit shorter than the length of the spore; perispore dehiscent in 10% KOH, smooth.

Additional specimens examined: **RUSSIA**: **Primorsky Territory**, Khasan District, Ryazanovka vicinity, on *Quercus mongolica*, 30 Jun 1982, M. Nazarova (VLA P–381); Trinity Bay, on *Quercus mongolica*, 4 Aug 2006, L. Vasilyeva (VLA P–1803). **KOREA**: vicinity of Seoul, Suwon, on *Quercus* sp., 30 May 1999, L. Vasilyeva (VLA P–1649)

Comments—In the Russian Far East, this species has been found only in the Khasan District, at the very southern extreme of the Primorsky Territory. It differs from *A. truncatum* (Schwein.) Y.M. Ju et al. in having smaller ostiolar discs that are of the *bovei*-type instead of the *truncatum*-type. The general appearance of the stromata is characterized by the presence of the shining-black and convex ostiolar caps, which persist for a long time but finally fall off to expose the flat ostiolar discs. The image of stromata of a specimen from Japan (Tsuneda & Arita 1984: Fig. 1) corresponds to that noted for specimens from southeastern Russia.

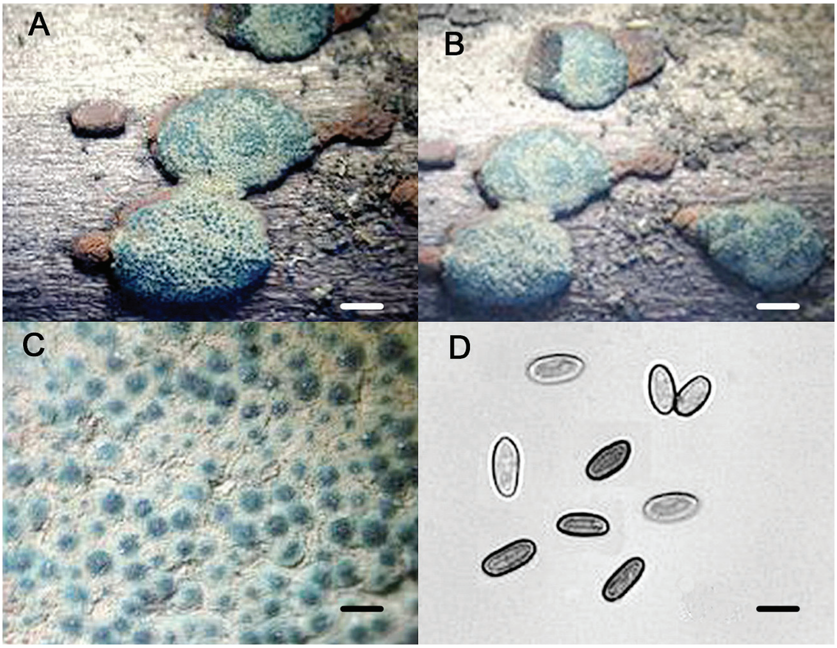

Fig. 4. *Hypoxylon cyanescens*: A-B. Stromata; C. Stromatal surface; D. Ascospores. Scale bars: A = 3.6 mm, B = 2.7 mm, C = 1.4 mm, D = 9.4 µm.

Hypoxylon cyanescens H.X. Ma, Lar.N. Vassiljeva & Yu Li, **sp. nov.** Fig. 4

MycoBank MB 802459

A Hypoxylone aeruginoso *in ascosporis majoribus et a* H. aeruginoso *var.* macrosporo *in granulis in KOH dissolutis differt.*

Type: China: Jilin Province, Changbai Mountain, alt. 1300 m, on rotten wood, 7 Sept 1998, Pei-gui Liu (**Holotype** HKAS32650).

Etymology: The epithet refers to the color of the stroma.

Stromata pulvinate or effused-pulvinate, flat or with conspicuous perithecial mounds 2–8 mm diam. and 0.5–1 mm thick; surface brown to dark vinaceous at the sterile margins, dark cyan blue, isabelline or gray olivaceous over the ostiolar portion, later becoming darkened, with KOH-extractable pigments bay (6) to livid purple (81). Perithecia 0.1–0.3 mm, ostioles finely papillate. Asci not observed. Ascospores brown, unicellular, ellipsoid-inequilateral or almost equilateral, with narrowly or broadly rounded ends, 11.5–13.5 × 5–6 µm, with a straight germ slit extending the length of the spore; perispore indehiscent in 10% KOH, smooth.

Additional specimens examined: **RUSSIA**: **Primorsky Territory**, Lazovsky State Nature Reserve, on *Ligustrina amurensis* (*Oleaceae*), 2 Aug 1986, L. Vasilyeva (VLA

P–2638); **Amur Region**, in the vicinity of Kundur, 24 Aug 1992, L. Vasilyeva (VLA P–2639). **KOREA**: Gangwon province, Pyeongchang county, Mt. Odaesan, 20 Sep 2006, L. Vasilyeva (VLA P–2746).

Comments—*Hypoxylon aeruginosum* J.H. Mill. and its large-spored variety *H. aeruginosum* var. *macrosporum* J.D. Rogers are the only previously known taxa with a dark cyan blue stromatal surface. Both varieties of *H. aeruginosum* are known from the western hemisphere (Guyana, Mexico, and Louisiana in the United States). *Hypoxylon cyanescens* is similar to the large-spored variety of *H. aeruginosum* in ascospore size but differs in the presence of bay to livid purple KOH-extractable pigments, whereas the latter variety has stromata apparently without KOH-extractable pigments.

Nectria araliae Lar.N. Vassiljeva & S.L. Stephenson, **sp. nov**.
MycoBank MB 802461

A Nectria coryli *in ascosporis minoribus et ascomatis nigrescentibus differt.*

Type: Russia: Primorsky Territory, Khasan District, Kedrovaya Pad Nature Reserve, on *Aralia elata* (*Araliaceae*), 25 Oct 1987, L. Vasilyeva (**Holotype** VLA P-1383).

Etymology: The epithet refers to the host genus of the holotype specimen.

Ascomata crowded in clusters, superficial or slightly embedded on a stromatic basis, dark-grey or black, globose to turbinate, becoming collapsed when dry, smooth, 200–250 µm diam., with small papillate ostioles. Asci cylindrical, 70–80 × 4–4.5 µm. Ascospores biseriate, two-celled, about 8–10 × 3–4 µm, with small setose appendages, usually budding in the asci.

Additional specimens examined: **RUSSIA**, **Primorsky Territory**, Ussuriysk District, Mountain-Taiga Station, on *Aralia elata*, 23 Sep 1984, M. Gvritischvili (VLA P-1993); Sikhote-Alin Nature Biosphere Reserve, on *A. elata*, 3 Aug 1985, L. Vasilyeva (VLA P-1381); **Kurile Islands**, Kunashir Island, on *A. elata*, 31 Jul 1987, L. Vasilyeva (VLA P-1386); **Khabarovsk Territory**, Komsomolsk Nature Reserve, on *Eleutherococcus senticosus*, 2 Jul 1986, L. Vasilyeva (VLA P-2139); **Jewish Autonomous Region**, Bastak Nature Reserve, on *E. senticosus*, 19 Aug 2004, L. Vasilyeva (VLA P-2124).

Comments—The similarity of *Nectria araliae* with *N. coryli* is impressive, and the ascospores have setose appendages and are usually budding in both species. Only recently, the ecological preferences of the north-Asian specimens and their distribution peculiarities were taken into consideration. Superficially, the black ascomata of *N. araliae* are similar to those of *Nectria austroamericana* (Samuels et al. 2006: photo on p. 120), but the latter seems to be restricted to members of the *Fabaceae* (*Acacia* and *Gleditsia*), has ascospores that are irregularly septate, with 3–6-transverse and one longitudinal septa, and has been reported for North and South America.

Discussion

The area considered in this paper is known as "the Eastern Asiatic Region (Oriasiaticum, Sino-Japanese Region, East Asian Region, Temperate Eastern Region)" and has been described as "the richest floristic region within the Holarctic Kingdom situated in temperate East Asia." Moreover, "it has been recognized as a natural floristic area since the publication in 1872 of August Grisebach's volume 'Die Vegetation der Erde' and was later delineated by such geobotanists as Ludwig Diels, Adolf Engler (as the Temperate Eastern region), Ronald Good (as the Sino-Japanese Region), and Armen Takhtajan" (http://en.wikipedia.org).

This special center of biodiversity is said to comprise "the southern part of the Russian Far East, southern part of Sakhalin, Manchuria, Korea, Japan, Taiwan, the relatively humid eastern part of mainland China extending from Manchuria and the coastline to the Eastern Himalaya and Kali Gandaki Valley in Nepal, including Sikkim, northern Burma (Myanmar) and northernmost Vietnam (parts of Tonkin)" (http://en.wikipedia.org). Although knowledge of the mycobiota in many of the countries in southern Asia (such as Burma or Vietnam) is rather poor, the scattered data reported in the rather limited literature and our preliminary observations indicate that the fungal biodiversity in countries (e.g., China, Japan, Korea, and southeastern Russia) located around the Sea of Japan differs from that of more southern portions of this world region. Moreover, it also should be noted that some current studies of vegetation also exclude the south of China, Burma, and Vietnam from the Sino-Japanese Floristic Region (Yih 2012: map at p. 16). It seems that the apparently unique area in question is primarily limited by the distribution in northeastern Asia of what are referred to as 'cool-temperate rainforests' (Krestov et al. 2011; Nakamura et al. 2011), whereas the excluded countries are characterized by different types of climate and vegetation.

Many pyrenomycetes have been collected exclusively in the Sea of Japan area and assigned to new species or genera (Vasilyeva 1990, 1993, 2001, 2007, 2010; Ju et al. 1999, 2009; Stadler et al. 2005; Vasilyeva & Mel'nik 2006; Vasilyeva & Stadler 2008; Vasilyeva et al. 2007, 2010; Hirooka et al. 2011). The main purpose of the Vasilyeva & Stephenson (2010b, 2011) series (for which this is the third contribution) is to emphasize that by neglecting biogeographical patterns, mycologists often have reached the wrong conclusions regarding application of species names to fungi in northeastern Asia. For example, *Cryptosphaeria eunomioides* (G.H. Otth) Höhn., *Diatrype disciformis* (Hoffm.) Fr., *Diatrypella quercina* (Pers.) Cooke, *Hypoxylon fragiforme* (Scop.) J. Kickx f., and *Rosellinia aquila* (Fr.) Ces. & De Not. have all been reported from the Russian Far East (e.g., Koval 1972) but actually do not occur there, and any of these names applied to Chinese fungi (Teng 1996) should be checked.

It is possible that species restricted to the Sea of Japan area had a wider distribution prior to the Glacial Age but were pushed by glaciers to the edge of the Pacific Ocean where the Sea of Japan formed after separation of the island arc of Japan from the Asian mainland about 20 million years ago. It is possible that this area became a kind of 'trap' for fungi that depend upon air currents for spore dispersal, whereas the air currents themselves often depend upon sea currents. If one looks at the map of sea currents around the Japanese Archipelago (http://valeriecourreges.blogspot.com), one can see that the main water circulation pattern in the Sea of Japan is created by two main currents, with the Tsushima Current being warm and the Liman Current being cold. In fact, these two currents create a closed system of water as well as the air currents responsible for transferring fungal spores. The partial exit from this closed system toward Sakhalin Island and the southern Kurile Islands is provided by the Soya Current. This might explain why some fungal species (e.g., *Loranitschkia viticola* Lar.N. Vassiljeva, *Nectria araliae*) are known not only from the Asian mainland near the Sea of Japan but also from Kunashir Island.

Only a few species have distribution patterns that extend southward from the Sea of Japan area, and the currents around the Japanese Archipelago are probably responsible for this distribution, too. The prevailing winds associated with the warm waters of the Kuroshio Current, which begin along the eastern shores of Taiwan, might carry fungal spores to Japan, and there are interesting examples of distribution patterns that extend along the eastern coast of Asia from the Kurile Islands to an area of the mainland at approximately the same latitude as Taiwan. For example, *Spirodecospora melnikii* (Lar.N. Vassiljeva) K.D. Hyde & Melnik, described from Kunashir Island (Vasilyeva 1990: as *Anthostomella melnikii*), also has been found in Korea (Mel'nik et al. 2005) and near Hong Kong (Lu et al. 1998). The Hong Kong fungus was described originally as *Spirodecospora bambusicola* B.S. Lu et al., now regarded as a synonym of *S. melnikii* (Mel'nik & Hyde 2003). The mainland coastline of China is mentioned above as one characteristic area of the Sino-Japanese Floristic Region, and further investigations may reveal a whole complex of species with the same distribution.

Similar distributional regularities also occur in a very distantly related group of fungi. Fifty-three of 123 rust species (43%) found in Russia belonging to *Chaconiaceae*, *Coleosporiaceae*, *Cronartiaceae*, *Melampsoraceae*, *Mikronegeriaceae*, *Phakopsoraceae*, and *Pucciniastraceae* are restricted to the Sea of Japan area (Azbukina & Karatygin 2010). Within this group, a number of species seem to have had their spores carried by the air current associated with the Soya Current from Japan to Sakhalin Island and southern Kurile Islands. Consequently, *Coleosporium yamabense* (Saho & I. Takah.) Hirats. f., *Naohidemyces fujisanensis* S. Sato et al., and *Uredinopsis woodsiae* Kamei are shared by Japan and Sakhalin Island (Azbukina 2005); *Pucciniastrum hydrangeae-petiolaris* Hirats. f. is known from Japan, Sakhalin Island, and Kunashir Island; *Thekopsora tripetaleiae* Hirats. f. has been reported from Japan and Iturup Island; and *T. menziesiae* Hirats. f. is known from Japan, Kunashir Island, and Iturup Island. The Taiwanese connection is also found in the rust fungi: *Blastospora itoana* Togashi & Onuma and *Milesina miyabei* Kamei are found both in the Sea of Japan area (China, Japan, the southern portion of the Russian Far East) and Taiwan (Ono et al. 1987; Azbukina 2005).

Zoogeographical studies clearly also distinguish the so-called 'Sino-Japanese realm' (Holt et al. 2013). This realm does not encompass such southern countries as Burma, Nepal, or Vietnam but extends further to the west of northern China than suggested for the center of fungal biodiversity around the Sea of Japan. In this context, a rather interesting fungus described recently by Zhuang & Bau (2008), namely *Sinofavus allantosporus* W.Y. Zhuang & T. Bau, merits attention. One specimen was collected in 2003 in the Magadan region (north-eastern Russia) by Nina Sazanova and sent to Larissa Vasilyeva but not described. A second collection found again in 2007 by Tolgor Bau in the westernmost Chinese province of Xinjiang (Tuomer Peak) was designated as the holotype of the species. *Sinofavus allantosporus* seems to be restricted to *Populus* species, the substrate of the Nina Sazanova specimen; the illustration provided in Zhuang & Bau (2008: Fig. 1) shows the branch (apparently from *Populus*) upon which the Chinese specimen fruited. As such, a double restriction—first to substrate and then to the northern mountainous region of northeastern Russia and northern China—is characteristic of *S. allantosporus*, and it is extremely surprising that such a peculiar discomycete with compound fruitbodies remained completely unnoticed until the 21st century. All these preliminary observations indicate the need for further investigation of several species complexes in north-eastern Asia that appear to have their own biogeographical patterns.

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