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On a new Atractiella

R. J. BANDONI[1] and PATRIK INDERBITZIN[2]

[1] Prof. Emeritus, Department of Botany, University of British Columbia,
#3529–6270 University Blvd., Vancouver,
British Columbia V6T 1Z4, Canada (Email: bandoni@interchange.ubc.ca)
[2] Graduate Studies, Dept. of Botany, University of British Columbia,
#3529–6270 University Blvd., Vancouver,
British Columbia V6T 1Z4, Canada (Email: bhpatrik@mail.botany.ubc.ca)

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Atractiella columbiana Bandoni et Inderbitzin sp. nov. is described from felled poplar logs in southwestern British Columbia. The species is characterized by sporocarps typical of the genus, but these can bear either conidia or basidia, or both. The pileolus is globoid to cupulate or discoid; it typically has a differentiated outer hyphal layer of parallel hyphae extending from the base of the stalk to the apex of the capitulum. Basidia are 4-celled; the sessile basidiospores being mostly unilateral and developing synchronously. Conidia develop sympodially on conidiogenous cells arising from assimilative hyphae, hyphae on sporocarp stalks, or in or on the capitulum. All such conidia are of similar appearance and dimensions.

Key words: Atractiellales, Hochnelomycetaceae, *Atractiella*, *A. columbiana*, *Hoehnelomyces*.

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Je popisován nový druh *Atractiella columbiana* Bandoni et Inderbitzin ze spadlých větví topolů v jihozápadní Britské Kolumbii. Tento druh je význačný plodnicemi typickými pro tento rod avšak mohou nést jak konidie tak i basidie. Klobouček je kulovitý, číškovitý nebo terčovitý a má typicky diferencovanou vnější hyfovou vrstvu složenou z paralelních hyf táhnoucích se od báze třeně až do vrcholku hlavičky. Basidie jsou čtyřbuněčné s jednostraně přisedlými basidiosporami, které se vyvíjejí současně. Konidie se vyvíjejí sympodiálně na konidiogenních buňkách, které vyrůstají z asimilativních hyf, z hyf na třeni plodnice nebo na hlavičce. Všechny tyto konidie jsou podobné vzhledem i svými rozměry.

The genus *Atractiella,* based on *A. brunaudiana* Saccardo (Saccardo, 1886), included only three species in the survey of gasteroid auricularioid basidiomycetes by Oberwinkler and Bandoni (1982). A species of *Hoehnelomyces,* possibly a member of this group, was described and illustrated, but not named, by Boidin et al. (1979). During the past two years, several collections have been made of an *Atractiella* similar to *A. brunaudiana* on felled logs of *Populus trichocarpa* Torr. et Gray in a single locality in southwestern British Columbia. The taxon differs from the type species in dimensions of the basidia and spores, in having conidial synnemata

superficially identical to basidiomata, or in having such conidia formed externally on stalks of basidiomata, in the hymenium together with basidia, or on hyphae on the substrate. The species is described here as *A. columbiana* sp. nov. here.

MATERIALS AND METHODS

Material was mounted in 3% KOH-Phloxine and/or 3% KOH-Weak Congo Red for bright field microscopy Spores and other structures were tested for amyloidity using Melzer's reagent, and for cyanophilly by staining with Lacto-phenol Cotton Blue (solutions as indicated in Hawksworth et al., 1995). Weak Congo red solution (Bandoni, unpublished) consists of: distilled water, 30 ml, glycerol, 3 ml, 1% aqueous Congo Red, 3 ml. A drop each of the stain solution and 3% KOH are mixed on the slide and the material is added; the preparation is then allowed to stand for 5–10 minutes for stain penetration. After covering, more stain is added at the edge of the coverslip as drying occurs or, alternatively, 10% glycerine in dist. water is added in this way to make the preparation semipermanent. The weak Congo Red preparation stains hyphal walls clearly, the intensity varying with the amount of stain added and with staining time. These preparations show less crystal formation than often occurs in mounts prepared with 1% Congo Red-KOH.

The collections examined, and the herbaria in which they are deposited, are listed under the specific description. Abbreviations for herbaria where specimens are preserved follow Holmgren et al. (1990).

TAXONOMY

Atractiella columbiana Bandoni et Inderbitzin, sp. nov. Figures 1–3.

Basidiomata stilboidea, 750–1100 μm alta, hyalina vel lactea, sicca albolutescentia, caules 400–720 μm longi, ad basim 98–230 μm crassi, attenuati et 55–75 μm ad apicem; tunica e hyphis 3.5–6.5 μm in diam., efibulatis; hyphae internae 2.5–5 μm in diam., efibulatae, compactae. Pilei globosi, 245–390 μm lati, 320–430 μm lati vel discoidei itaque, 120–160 μm lati. Probasidia 50–90 $\times$ 3.5–6 μm demum anguste clavata vel cylindracea, 4-cellularia. Basidiosporae 17–22(-27.5) $\times$ $\times$ 5–7 μm, sessiles, subcylindraceae vel subfusiformes, anguste clavatae, rectae vel curvatae, per hypham germinantes. Conidiomata stilboidea, partim basidiomatibus similia; cellulae conidiogenae plerumque 50–90 $\times$ 4–8.5 μm, ad apicem attenuatae sympodialiter proliferantes, conidia 16–25 $\times$ 6–9 μm, anguste clavata vel subcylindracea, ad basim truncata, per hypham vel per repetitionem germinantia.

Ad truncum *Populi trichocarpae* putrescentem, subcorticale vel in ligno nudo vicino. Delta (Ladner), British Columbia, Canada. A. & R. Bandoni 12992, HOLOTYPUS (DAOM).

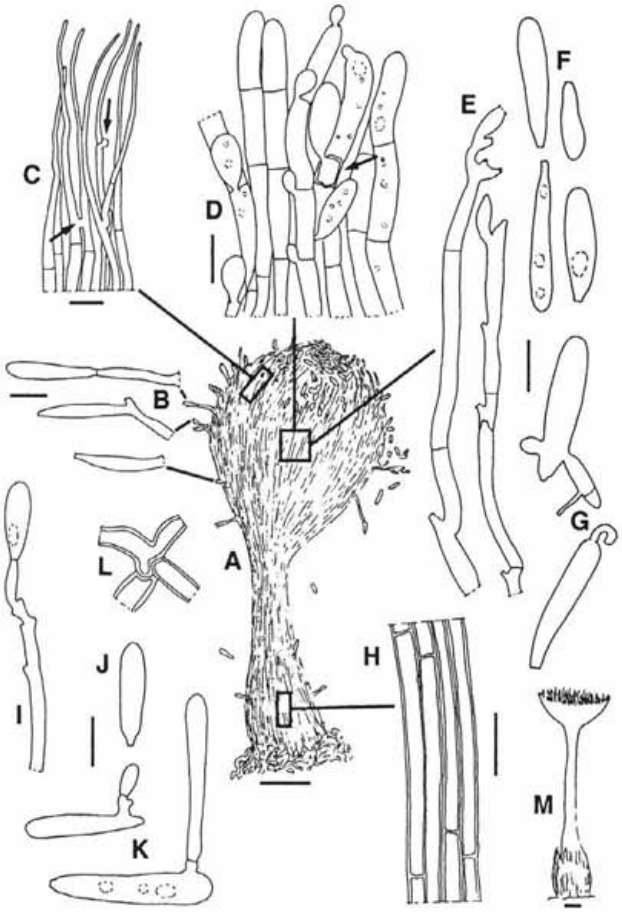

Fig. 1. *Atractiella columbiana* (A-C, H, M, from A. & R. B. 13992, Holotype; I – L from A. & R. B. 12993; E-G, from A. & R. B. 13008). **A.** Habit (drawn from photograph) of a complex sporocarp, the sheath filaments extending over the fertile portion. **B.-L.** Details (camera lucida drawings) of the sporocarp structure and types of reproductive structures in, on, or associated with such sporocarps. **B.** Conidia borne on conidiogenous cells protruding from sporocarp (lines). **C.** tapering apices of sheath hyphae; note clamp-like septum (arrow, above, right) and anastomosis (arrow, lower center). **D.** Hymenium, showing compact basidia of different stages in development. (Note central basidium showing typical unilateral and synchronous development of the 4 basidiospores; arrow, right, indicates chlamydospore-like structure from which a spore appears to be developing). **E.** Conidiogenous hyphae from hymenium of sporocarp, the successive conidiogenous levels suggesting extension to keep pace with extending hymenium. **F.** Conidia. **G.** Germinating conidia. **H.** Hyphae of the sheath showing slightly thickened walls and close, parallel arrangement. **I.** Conidiogenous cell and attached conidium, from hyphae on substratum adjacent to basidiome. **J.** Conidium, and **K.** germinating conidia, the lowermost germinating by a repetitive process. **L.** Clamp-like septum of hypha producing conidiogenous cells on substrate surface. **M.** Sporocarp with a discoid pileolus, that sketched had both basidia and conidiogenous cells in the hymenium. The volva-like basal portion probably is a rudimentary sheath. (Bars all equal 10 μm)

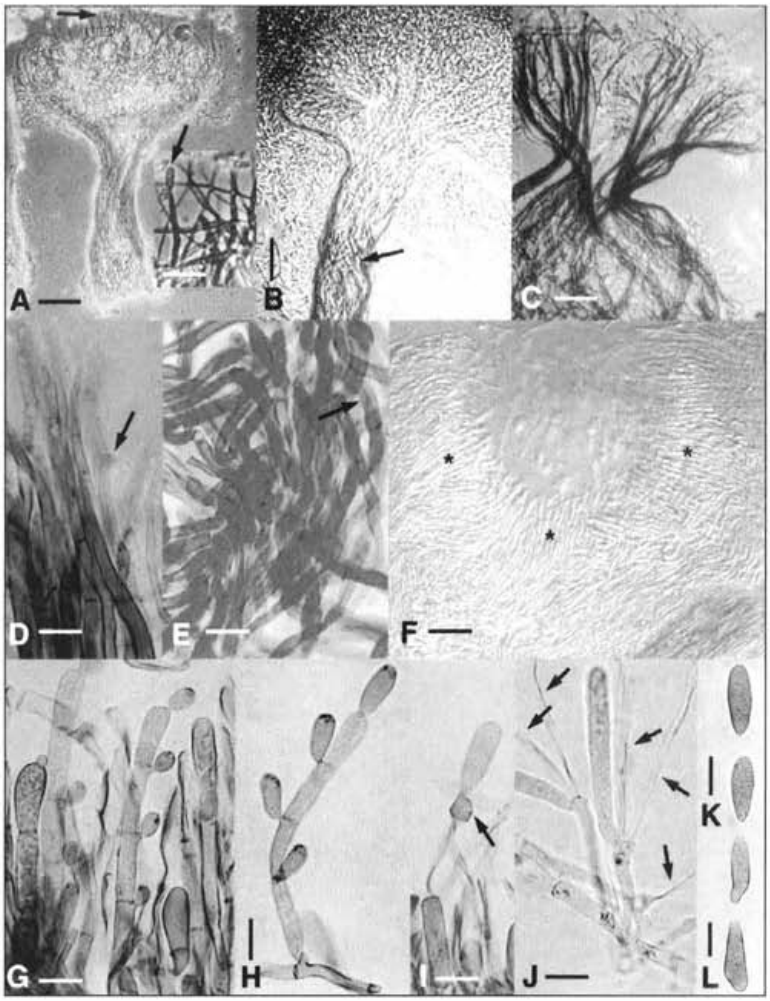

Fig. 2. *Atractiella columbiana* (all figures from A. & R. B. 12993, Holotype). **A.** Sporocarp with discoid pileolus, lightly stained with KOH – Weak Congo Red. Enlarged portion, from margin of disk, the same attached conidium indicated by arrows in the habit and inset. Note the tapered apices of sheath hyphae in both insert and main print. **B.** Sporocarp stained as in A, the sheath hyphae of the stalk showing characteristic spiral arrangement found in most. **C.** Sheath, from crushed mount, separated from most of the stalk core and hymenium, showing continuity of sheath hyphae around stalk (below central constriction) and those extending around pileolus. **D.** Detail showing stained sheath hyphal apices from their widest points (ca. midpoint on capitulum) upward. Arrow indicates clamp like septum on one hypha. **E.** Thick-walled, irregularly arranged hyphae of emplacement, the lumen almost occluded in spots (arrow). **F.** Hymenium of a basidiome showing the compact, concave form in some basidiomata, the external form not discoid. **G.** Hymenium with developing basidia and basidiospores. **H.** Typical basidium with 4 unilateral, sessile, synchronously-formed basidiospores. **I.** Atypical basidium, a basidiospore apparently developing from a chlamydospore-like remnant of protoplasm in a partially emptied basidial cell (below arrow). **J.** Probasidium, upper center, and numerous collapsed basidia (arrows) showing proliferation pattern. **K.** Basidiospores of the most common form, showing inconspicuous attachment scars. **L.** Curved basidiospores. (Bars in A, left = 10 μm; right bar = 30 μm. All other bars = 10 μm)

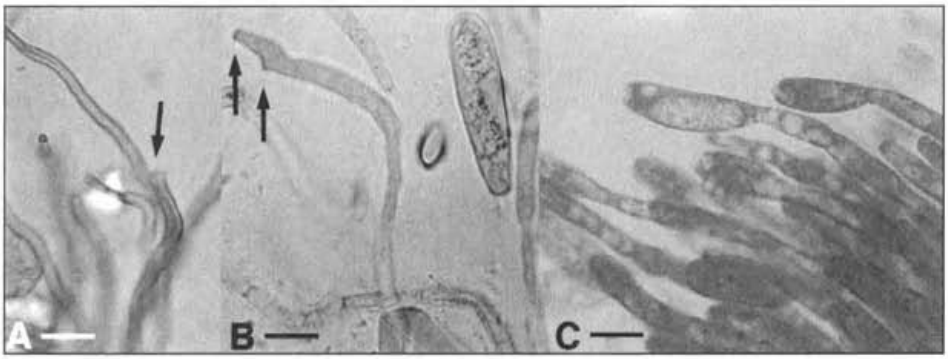

Fig. 3. *Atractiella columbiana* (from A, & R. B. 12992). **A.** Sheath hypha showing locus (arrow) where conidium developed during elongation and **B.** hymenial conidiogenous cell from basidiome, two sporogenous loci indicated by arrows. **C.** Hymenium of conidial sporocarp showing young conidiogenous cells and attached conidia. (Bars = 10 μm).

Basidiomata scattered to gregarious, often intermixed with conidiomata of similar form or differing slightly, most 750–1100 μm high, stipitate-pileolate (Fig. 1A, M; 2A-B); stalk 400–720 μm long, 98–230 μm wide basally, tapering to 55–75 μm below the pileolus, the latter 245–390 μm wide, ca. 120–160 μm high centrally if discoid, 320–430 μm high if globose or oval, hyaline to milky when fresh, the basal half sometimes appearing gelatinous, with a slime drop above surrounded by paraphysis-like tapered tips of the sheath hyphae (Fig. 2A), these enclosing numerous basidiospores and/or conidia, drying inconspicuous, whitish. Stalk arising from a pulvinate emplacement of irregularly arranged hyphae (Fig. 2E), 3.5 to 6.5 μm in diam., the walls to 2 μm thick. A sheath like layer commonly surrounds the stalk; it is composed of hyphae 2.5–5 μm in diam., their walls thickened to ca. 1 μm, these hyphae closely adherent laterally, typically arranged in a steep spiral around the stalk (Fig. 2B); they are septate, efibulate, branched, the main axis swollen immediately below the infrequent branch points, branches recurving and adhering to the parent; the sheath typically continuous upward around the capitulum, the hyphae branching more frequently and separating from one another there, swollen to 5–7 μm at the level of the hymenium, then tapering upward, arching to form a loose enclosure over the fertile zone, the tips narrowed tp 1–1.5 μm in diam., the terminal cell slightly larger, rounded apically; some such hyphae conidiogenous, producing a conidium, then extending below its base, the locus visible after secession (Fig. 3A); sheath lacking entirely in some basidiomata and conidiomata, otherwise extending to varying distances up the stalk, to the base of the pileolus, or to the edge of the disk and appearing as a marginal fringe (Fig. 2A); central core hyphae of stalk 3.5–6 μm in diam., thin walled, septate, some septa adventitious, without clamps, compactly arranged, parallel, strongly adherent, infrequently branched, continuing to the hymenium to

produce basidia and/or conidia. Hymenium of compactly arranged probasidia, and basidia, these developing continuously, older basidiomata with numerous collapsed basidia present (Fig. 1J); probasidia narrowly clavate (Fig. 1J), finally 4-celled, 50–90 × 3.5–6 μm, the 3 upper cells cylindrical, the basal cell longer and tapering to 2.5–3.5 μm; predominantly clearly simple septate basally (Fig. 2G, H, J), but infrequently appear to be clamped there; basidial walls thin, each cell producing a sessile basidiospore (Fig. 1D; Fig. 2G, H), then collapsing as the protoplast migrates into the spore. Basidiospores 17–22(-27.5) × 5–7 μm, subcylindrical to subfusiform, narrow clavate, straight or variously curved (Fig. 2K, L), some approaching allantoid or sigmoid; walls smooth, thin, inamyloid, not cyanescent; germination by germ tube. Conidia produced in or on some basidiomata (Fig. 1B, E-G, I-L; Fig. 3B-C) in purely conidial structures essentially like the basidiomata, on tapered apices of sheath hyphae (Fig. 3A), or sometimes along stalks of both synnemata and basidiomata; conidia also develop on hyphae in culture and on the substrate surface in nature (Fig. 1I-L); these can be born on single, separate conidiogenous cells, but rudimentary synnemata are more common on the natural substrate, often enclosing conidiophores of associated dematiaceous hyphomycetes; synnemata variable, many indistinguishable from the basidiomata superficially, but others often under 500 μm high, the capitulum globose, oval, cupulate or discoid to irregularly clavate; stalk structure as with basidiomata, the sheath present or absent, if present, either extending to the margin of the discoid or cupulate portion of the synnema (as in Fig. 2A) and forming a marginal fringe, or otherwise ending below the capitulum, the hyphae and their arrangement as in basidiomata. Conidiogenous cells proliferating sympodially in all cases, many of those on the stalk and fertile portion of sporocarps 50–90 × 4–8.5 μm, tapered apically, producing a terminal conidium, then proliferating immediately below it (Fig. 1I; Fig. 3B), leaving a conspicuous conical locus and often bent at an angle below this; conidia 16–25 × 6–9 μm, thin-walled, smooth, narrowly clavate to subcylindrical (Fig. 1B, F, J; Fig. 3B), indistinguishable from basidiospores in general form, but most visibly truncate basally or with an abruptly broadened shoulder a short distance above the attachment point; germination by germ tube (Fig. 1G) or by a repetitive process (Fig. 1K).

Habitat: Growing on felled trunks of *Populus trichocarpa* (cut 2–3 years previously), beneath the loose bark, on the inner bark layer, or immediately adjacent to the bark on recently exposed wood.

Collections examined: All collections decaying logs of *Populus trichocarpa* (felled, on the ground for 2–3 years); on wood beneath loosened bark, on inner bark, or on exposed wood immediately adjacent to bark edges; all came from a a single locality, as follows:. Canada: British Columbia; Delta (Ladner), Lagoon Walk, off Ferry Rd. A. & R. Bandoni, 12992, 14 Oct. 2000, HOLOTYPE (DAOM); A. & R. Bandoni 12993, 19 Oct. 2000 (DAOM); 12997, 15 Oct. 2000 (DAOM);

13004, 25 Oct. 2000 DAOM); 13007, 2 Nov. 2000 (UBC); 13008B, 2 Nov. 2000 (UBC); 13028, 30 Nov. 2000 (UBC).

Discussion

All previously described species of *Atractiella* are clampless, and almost all septa appear simple in *A. columbiana*. However, rare septa at the bases of basidia and elsewhere are possibly clamped (Figs. 1C, L; Fig. 2D). The basidia are similar to those in other species of the genus, typically producing passively released sessile basidiospores unilaterally and synchronously. Exceptions with respect to synchrony were observed several times; these might represent delayed development of a single spore on a basidium. In each such case, a small chlamydospore-like compartment within the basidium giving rise to a typical basidiospore (Fig. 1D, 2I, arrows)

The species is distinctive in having conidia often present superficially on the stalks of basidiomata and conidiomata, on the capitulum surface (Fig. 1B; Fig. 2A), mixed with basidia (Fig. 3B), free on the substratum or in synnematous conidiomata. The surface sometimes arising from the growing sheath hyphae (Fig. 3A). Conidial synnemata can be stilbelloid, cupulate, or discoid, the conidiogenous cells often occurring in a hymenium-like layer (Fig. 3C) on the planate to convex or concave distal surface. Similar conidia are produced on the mycelium in nature (Fig. 1L) and in culture; sporocarps developed in culture were mostly anamorphic, but no attempt was made to determine conditions necessary for formation of basidiomata.

Since conidia and basidiospores are of similar form, overlap in dimensions, and have the same wall characteristics, they can easily be confused with one another. Generally, however, the conidia are symmetrical and are relatively conspicuously truncate basally. The basidiospores are attached asymmetrically, and the scar left after secession is inconspicuous.

Known species of *Atractiella* fall into two groups: In *A. brunaudiana* Saccardo (type species) and *A. columbiana*, hyphae of the sheath terminate in narrowed hyphidium-like filaments apically. These species lack hyphidia in the hymenium. The sheath sometimes is absent or is only partially developed, but it generally is present and surrounds the stalk, the same hyphae extending upward around the capitulum. The sheath hyphae of the stalk and basal portion of the capitulum are closely parallel and are more strongly adherent to one another than to the inner core of cells. Pressure on the coverslip of a whole mount often results in separation of the sheath from the central core, as seen in Figure 2C. The apices of individual sheath hyphae are free (Figs. 1C; 2D), and they sometimes appear to be present in the hymenium when whole mounts of single basidiomes are flattened. However, in *A. columbiana*, they are restricted to the area around the hymenial margin.

A. brunaudiana appears to have a sheath like that in *A. columbiana* , but true hyphidia are present in hymenia of other species in the genus In *A. columbiana,* the sheath hyphae can be conidiogenous (Fig. 3A). In *A. solani* (Cohn et Schroet.) Oberw. et Bandoni and *A. delectans* (Moeller) Oberw. et Bandoni, a sheath is present around the stalk, where it is at least superficially similar to that described above, but, the tapered extensions over the fertile area are absent. Sporocarps of these species do have hyphidia that develop from the fertile hyphae (Oberwinkler et Bandoni, 1982). It is not clear at this time whether the two groups are closely related (i.e., actually congeneric) as the study of most of these species is based upon limited herbarium material, some (e.g., *A. brunaudiana*) from a single poorly preserved collection. Occasional stalked conidiomata of *A. columbiana* lack both a sheath and a capitulum; here the stalk terminates in a ring of conidiogenous cells through which the central core of cells sometimes protrudes. In typical sporocarps, the central core of the stipe extends to the hymenial area, where they produce the fertile hyphae bearing basidia or conidia.

Conidial production has been reported only on germ tubes arising from basidiospores in *A. delectans* and *A. solani;* they occur on conidiophores in a *Hoehnellomyces* sp. reported on olives from Italy (Boidin et al., 1979). The description and figures of the last named fungus appear to be *Atractiella – like.* Although the dimensions of basidia and spores given for this species are within those noted here for *A. columbiana,* the conidia are much smaller and the conidiophores and conidiogenos cells also differ sharply. Thus far, no other atractyelloid taxa have been found which produce synnematous anamorphs that closely resemble their basidiocarps. Nor are conidium-bearing cells known on the stipes, on the tapered hyphae of the sheath, and on conidiogenous cells in the hymenium in species other than *A. columbiana.*

In conclusion, the unusual habitat in which much of the *A. columbiana* material grew, (i. e. beneath the slightly loosened bark of relatively sound trunks of felled or fallen trees) is rich in arthropods, worms, bacteria, and fungi. The presence of arthropods, and the sticky spore drops characteristic of Atractiellas suggest transport by such animals. Although we contemplated erecting a new genus for the species described here, the similarities to *A. brunaudiana* dictate against such a course at this time.

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