

**14030203** March 2, 2014. Leg. Jason Karakehian. Det. JK. United States, Massachusetts, Worcester Co., Princeton, Wachusett Meadow Wildlife Sanctuary, West Trail. *Permission to collect verbally granted for this date only in conjunction with the Boston Mycological Club winter excursion.* On well-rotted wood (white-rot).

Sections made on freezing stage to 20 µm and 10 µm, dilute gum arabic used as a matrix. Observations made on Olympus BX40, photos and measurements made on MicroSuite Special Edition. Observations made from living material. Material kept in a plastic box in a refrigerator – further observations made on March 15 and 23. Ascospore measurements from living spores collected on a coverglass placed onto an apothecium, mounted in water and also observed in phloxine for septation.

**Ascomata** apothecia sessile, superficial, surrounded by a scanty white subiculum, single to gregarious, ~ 0.4 – 0.5 mm diam, **disc** distinctly orange to yellow – a few pale yellow; releasing a small amount of yellow exudate in 3% KOH, with single or fascicles of short pale yellow hairs on the flanks and margin. In cross-section the pigmented region is restricted to the hymenium and mostly the inner layer of the ectal excipulum which also forms the margin – the basal tissues are hyaline and conspicuously gelatinized. An apothecium placed in water or 3% KOH and observed under magnification will appear to have a highly pigmented hymenium encased in a receptacle of transparent tissue.

**Hairs** on margin and flanks, single or in fascicles of 6 – 10 or more, straight, septate, sparingly to strongly encrusted with yellow crystals which dissolve in 3% KOH, hyaline with sparse yellow pigmented globules within, ~ 100 µm long at most, single hairs: 3.8 – 4.5 µm wide at the bases and apices. **Hymenium** ~ 88 µm thick, in sections from living material the hymenium and inner ectal excipulum become pale J+ in Melzer's (the same in 3% KOH and Lugol's) though fading after ~ 5 min. **Asci** clavate, 8-spored, pore J+ in Melzer's and I+ in 3% KOH & Lugol's (asci protoplasm turns brick red in this treatment), apex slightly thickened ~ 1.7 µm, 77 – 87 x 8 – 10 µm, dehiscence via an apical pore leaving a protruding collar at the apex, arising from repeating croziers. **Paraphyses** filiform, dichotomously branching anywhere along their length, straight, not enlarged but somewhat contorted at the apices, not extending beyond the length of the asci, filled with plentiful yellow-golden pigmented globules, ~ 1.7 µm diam. **Ascospores** biserial, varied in form and size: elliptical to cylindrical with fusoid or obtuse ends, 0 – 3 septate (this condition occasionally observed even within the same ascus) mostly 1-septate, smooth, hyaline, with 2 – 4 refractive bodies in each cell, septa not easily observable in water, no appendages observed, budding to form ascocidia in older apothecia (as observed from living material on 3/23), 50 ascospores at 1000x in water: (9.9-) 13.8 (-22.4) x (2.9-) 3.9 (-4.8) µm. **Subhymenium** indistinct. **Ectal excipulum** (as observed in water and phloxine mounts) consisting of 3 intergrading layers: a basal layer of thick and refractive-walled (gelatinized) *textura intricata* of elongate-inflated cells (~ 6.7 µm) which gives rise to the subicular hyphae and contains a small amount of orange pigment within the hyphae; above this a layer of *textura oblita* of anastomosing thick-walled hyphae running perpendicularly to the hymenium which gives rise to the hairs of the flanks, containing still more orange pigment within the hyphae; running parallel to this and directly below the hymenium is a layer (14-19 µm thick) of thin-walled *textura prismatica*, cohering and highly orange pigmented and forming the inner layer of the **margin** (~ 39 µm wide), the outer layer of which is the second layer of *textura oblita* which runs up and forms the hairs of the margin. **Subicular hyphae** 2.5 – 3.2 µm diam, septa scarce – as observed in long and straight hyphae, thick-walled (in phloxine and water), in water having that “glassy” or refractive look to them, anastomosing occasionally, straight or intertwining at the base of the apothecia, surface smooth to slightly roughened.

*Possibly A. trabinellinoides or possibly somewhere near A. major and A. cornuta.*

**Culture** 3/6 - Two apothecia placed at opposition to each other on damp pieces of filter paper in the inside of a petri dish lid with the dish of water agar media with chloramphenicol and streptomycin suspended over them to collect ascospores forcibly ejected upwards. After twelve hours ascospores germinated from both poles and frequently laterally. 3/7 – germinating ascospores transferred to PDA. For single spore transfers, the water agar plates were inverted on the stage of compound microscope and isolated ascospores were located through the bottom of the dish which was then marked. From this marked area, under sterile conditions with a scalpel, a 2 mm square block of agar bearing the single spore was excised and transferred to the PDA, then reexamined under magnification to be sure only one spore was captured. For polysporous cultures the same method was employed. 3/15 – small sections of polysporous culture from plate A-PDA placed in two eppendorf tubes of 250 ml CTAB buffer.