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SOME WOOD-STAINING FUNGI FROM PULPWOOD CHIPS

WALLACE E. ESLYN

Forest Products Laboratory, Forest Service, U. S. Department of Agriculture, Madison, Wis. 53705; Maintained at Madison, Wis., in cooperation with the University of Wisconsin

AND

Ross W. DAVIDSON

Colorado State University, Fort Collins, Colo. 80521

Many studies have been conducted on micro-organisms associated with wood chip storage both in North America and abroad, especially in Scandinavia. Wood-staining fungi were generally present, either in the cool, exterior layers of chip piles or were scattered throughout both softwood piles (Lindgren and Eslyn, 1961; Björkman and Haeger, 1963; Bergman and Nilsson, 1966, 1971; Shields and Unligil, 1968; Butcher and Howard, 1968; Kubiak et al., 1969) and hardwood piles (Popescu, 1965; Eslyn, 1967; Bergman and Nilsson, 1967, 1968; Greaves, 197 1, 1973; Gambetta and Orlandi, 197 1; Zielinski, 1973).

This paper describes a new species of *Ceratocystis* from hardwood chips, and provides some observations on other wood-staining fungal species isolated predominantly from stored wood chips or, to a lesser degree, from logs. As the capacity for effecting soft-rot of wood is of paramount importance, each stain isolate was tested for its soft-rot capability.

METHODS

Soft-rot capability was tested by the serial-block method described by Eslyn (1969). Sapwood blocks, 25.4 × 12.5 × 6.3 mm, with the 6.3 mm dimension in the grain direction, were cut from both blackgum (*Nyssa sylvatica* Marsh.) and southern pine (Pinus sp.) wood. In most cases, both wood species were tested to determine capability of test isolates to decay both hardwood and softwood. The blocks were numbered, placed in a conditioning room at 80°F and 70% relative humidity until equilibrium weight was reached, then weighed. Ten blocks of similar wood species were then strung together, serilized, and subjected to decay for 12 weeks at 27°C. The blocks were then removed from test, reconditioned, weighed, and their weight losses calcuated. All tests were run in duplicate. If results deviated substantially between duplicates, the test was rerun. Decay capacity was based on the block exhibiting maximum weight loss within the two replications inoculated with an isolate.

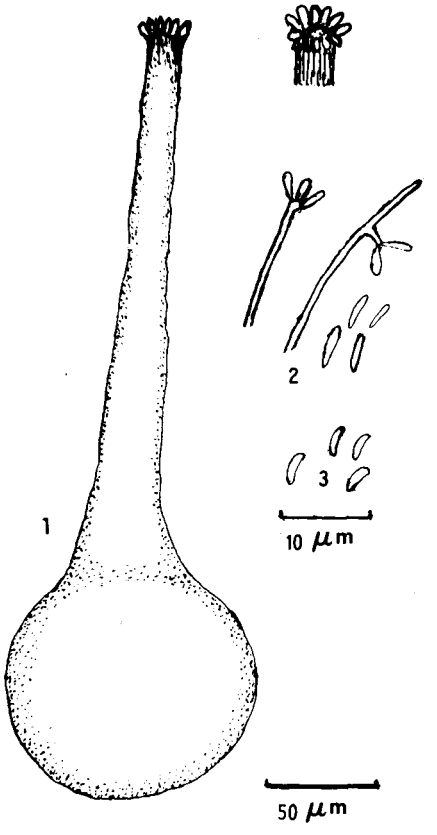
Herbarium abbreviations are according to Holmgren and Keuken (1974).

DESCRIPTIONS AND RESULTS

Species Isolated From Wood Chip Storage Piles.

Ceratocystis rostrocoronata Davidson & Eslyn, sp. nov. Figs. 1–3.

Perithecia globosa e fusco-atra, 115– 150 μm diam; fimbriae ostiolares brevia, ex hyalino brunneolae, ostiolum in forma coronae circumdantes,



Figs. 1-3. ***Ceratocystis rostrocoronata.*** **1. Perithecium; 2. Conidiophores and conidia; 3. Ascospores.**

1.5–2 × 5–10 μm; ascosporae hyalinae, segmentum mali aurantii conformatae, a latere visae 3.8–5.5 × 1.4–2.5 μm, a margine visae angustiores; conidia hyalina, fere cylindrica, recta subcurvatae, 3–10 × 1–2.5 μm.

HOLOTYPUS—U.S.A.,Maine: In ligno *Betula* sp.—RWD 942 *(ME-B1C-14)* culture isolated from Betula sp. Deposited at National Fungus Collections (BPI). A living culture (ATCC 28826) of the type has been deposited at the American Type Culture Collections, Rockville, Maryland 20852.

Perithecia globose, dark brown to black, with a few brown hyphae over the surface, 115–150 μm diam (Fig. 1); necks dark, slightly roughened, length variable, 160–700 μm, 24–35 μm thick at base to 17–22 μm at tip, at maturity the tips (ostiolar filaments short hyaline to light brown projections surrounding ostiole, crownlike) 1.5–2 × 5–10 μm; ascospores hyaline, shaped like the segment of an orange, 1.4–2.5 × 3.8–5.5 μm in side view (Fig. 3); conidia as in the genus *Sporothrix,* hyaline, borne on normal hyaline hyphae or short to long branches, in groups with separate attachment, nearly cylindrical, straight to slightly curved, 1.0–2.5 × 3–10 μm (Fig. 2).

Cultural characteristics—Culture white cottony to tufted at surface, mat with appressed margins, intermediate to slow growth rate (about 16 mm radius in 10 days), with perithecial primordia visible as dark brown specks at 14–20 days, developing necks and maturing in 5–6 weeks. Perithecia may be in small,

dense, black masses or scattered uniformly over the culture surface. Several of the cultures fail to develop necks on the perithecia or the necks may be considerably shortened. The necks are black, slightly roughened, and sometimes swollen where regrowth occurs. Growth rates and mats are somewhat similar to *C. gossipina* Davidson (1971), but the necks are shorter and ostiolar filaments quite different.

Ceratocystis rostrocoronata differs from most other species in lacking the development of large droplets of ascospores at the tips of the necks. Further, the species is typified by a crown of hyaline to light brown filaments surrounding the ostiole; hence the name "rostrocoronata."

Culture studies were based on isolate RWD 942 from birch chips and RWD 943, 944, and 945 from mixtures of American beech (Fagus *grandifolia* Ehrh.), birth ***(Betula*** sp.), and sugar maple (*Acer saccharum* Marsh.). Isolate RWD 945 did not develop perithecial necks, but was otherwise similar. All were isolated from the same wood chip pile located at Winslow, Maine (Eslyn, 1967).

The species has been found, in laboratory tests, capable of decaying blackgum chips but not southern pine chips. Isolate RWD 942 (Madison isolate ME-B1C-14) caused a maximum weight loss of 25.8% in blackgum, but only 2.3% in pine. Weight losses of 3.0%, or less, such as those encountered here when pine test blocks were used, are considered caused by natural leaching in the blocks during wetting and subsequent incubation.

Ceratocystis piceae (Münch) Bakshi, Trans. Brit. Mycol. Soc. **33:** 113. 1950.—Reported common on softwoods and less common on hardwoods in Europe and North America (Griffin, 1968). Present isolates were obtained from Fagus *grandfolia* (ME-BC-1) and *Acer saccharum* (ME-MC-10) wood chips stored at Winslow, Maine.

Griffin (1968) reported *C. piceae* caused small streaks of blue stain or failed to cause stain. In our laboratory tests, this fungus caused appreciable decay in blackgum wood blocks; in American beech, a maximum weight loss of about 11.4% and in the sugar maple, 17.3%. No decay tests were conducted on southern pine blocks. However, Bergman and Nilsson's experiments (1968,197 1) showed no weight losses caused by this fungus when tested on pine and spruce. In addition, Liese and Schmid (1961), in electron microscopic studies, found C. *piceae* caused no visible change in the cell walls of *Pinus sylvestris* L. or *Picea abies* (L.) Karst.; Greaves (1973) indicated this fungus is capable of moderate decay of *Eucalyptus regnans* F. Muell. and minor decay (3.3%) of *Pinus radiata* D. Don.

The asexual stage of this fungus has recently been placed into the new hyphomycete genus *Pesotum* Crane & Schoknecht (1973).

Ceratocystis sp.—A low-growing species similar to *C. sparsa* Davidson (197 1) with white graphia and isolated from a mixed Fagus *grandifolia-Betula* sp.-*Acer saccharum* chip pile (ME-MIC-8) at Winslow, Maine. Characterized in culture by white mycelium and abundant black perithecia as in *C. rostrocoronata.* Perithecia here, however, did not mature.

Capable of significant degrade of blackgum wood, causing a maximum weight loss of 20.6% in laboratory tests. Not tested on pine wood.

Graphium sp.—Culture (ME-MC-11) obtained from *Acer saccharum* wood chips stored at Winslow, Maine. Similar to *C. piceae*, but much slower growing and lacking perithecia.

Caused substantial weight loss (25.8%) in blackgum in laboratory tests.

Phialophora sp.—Culture (ME-W3-1) from *Picea glauca* (Moench) Voss wood chips in Wisconsin. A slow-growing culture; white at first, then with a light gray to dark gray center, 6–7 mm radial growth in 7 days. Conidia hyaline, 1.0–2.0 × 4–10 μm, conidiophores 5–15 μm long, 1.5–2.5 μm wide, but tapered to a smaller diameter at tip.

Caused substantial weight loss (27.2%) in blackgum test blocks, but insignificant losses (4.4%) in southern pine test blocks.

Aureobasidium sp.—Culture (ME-W3-2) from *Pinus banksiana* Lamb. wood chips in Wisconsin. Produces abundant conidia which are hyaline, ovoid to elongate, curved: a bacterium also present. Growth slow, about 6–7 mm radius in 7 days. Mycelium very dark and appressed to surface of culture medium.

Caused appreciable blue staining and decay (20.7%) in blackgum wood blocks, and minor decay (3.1%) in southern pine blocks.

Sterile Mycelium.—Culture (ME-W1-2) from *Pinus resinosa* Ait. chips stored in Wisconsin. A very dark, slow growing mycelium (4–5 mm radius in 7 days), dark gray raised mat with pale colored raised margin. No spores were seen attached to hyphae, but globose-to-ovoid brown cells (4–6 × 7–12 μm) were present with associated brown hyphae, some cells being terminal. Also, hyaline yeast-like cells were present in some areas of the culture mat.

Caused blue stain in some blackgum blocks and insigificant weight losses in both blackgum (4.1%) and southern pine (3.8%) blocks.

Sterile Mycelium.—Culture (ME-WE-6) from *Acer rubrum* L. chips in Wisconsin. White to light gray, very fast growth, about 40 mm radius in 7 days. Hyphae variablein size, 1.5–7.0 μm in diam with smaller hyphae mostly hyaline and larger diameter hyphae with brown and rough surfaces. No spores seen.

Caused blue staining in both blackgum and southern pine test blocks, but moderate decay only in blackgum blocks (12.0%).

Aureobasidium pullulans (de Bary) Arnaud, Ann. Mycol. 8: 475. 1910.—Culture (ME-MS-2) from mixed western conifer chip pile, Frenchtown, Montana. The fungus is very common in wood and has been isolated frequently by the authors throughout the years from diverse products such as weathered plywood, painted siding, wood chips, pulpwood logs, utility poles, posts.

The species identity of this and other listings of *Aureobasidium* are difficult to obtain, but we believe that cultures of this type could be several separate species. The descriptions given for *Hormiscium gelatinosum* Hedgc. (Hedgcock, 1906) and for *Hormonemu dematioides* Lager. & Melin (Lagerberg et al., 1927) more nearly agree with this fungus. However, both of these species have been listed as synonomous with *A. pullulans* (Cooke, 1962). From 7-day-old cultures on malt extract agar medium, the conidia are hyaline and very abundant. Older cultures contain some brown one- or two-celled sporelike bodies, but the hyaline one-celled conidia are most conspicuous.

Caused a small amount of decay (6.2%) in blackgum test blocks, but no decay in southern pine. The culture was mixed with a bacterial-like slime.

Phialophora sp.—Culture (ME-MS-3) from mixed western conifer chip pile, Frenchtown, Montana. Growth slow on malt agar medium, about 7 mm radius in 7 days; pure white dense powdery mat appressed to surface of medium and with a very pronounced fimbriate margin; center of older cultures turning light gray and under surface of mat variable light brown to dark brown. Hyphae hyaline 1.5–2.5 μm thick; conidiophores numerous, slender, about the same thickness as hyphae but tapering slightly to the tip, 15–30 μm long, perhaps some much longer, sometimes appearing slightly enlarged at the tip. Conidia very abundant, elongate, straight, or slightly curved, usually larger at apical end, variable in size from 2.5–8.0 × 1.0–2.5 μm.

Caused a small amount of decay in blackgum blocks (6.8%), but no decay in southern pine.

Species Isolated From Fresh, Unstored Chips or Logs.

Ceratocystis pluriannulata (Hedgcock) C. Moreau, Rev. Mycol., Suppl. Colonial 17: 22. 1952.—From aspen (*Populus* sp.) pulpwood chips, Oconto Falls, Wisconsin, 1972 (RWD-936).

Chips were received in mid-December, still moist (green), and contained dark stain and numerous Ceratocystis perithecia on the surface.

Isolations from perithecia on numerous chips were identified as C. pluriannulata. Cultures produced mature perithecia in 7 days. This is the most common species on hardwood logs and lumber in the southern states and is widespread in North America and Europe.

Hunt (1956) states that it is readily identified by the pure white cultures, but this is not usual for fresh isolates. Cultures may be dark, usually with dark segments, in contrast to solid dark substrata of *C. pilifera* (Fr.) C. Moreau, which is common on pine lumber. Hunt (1956) also compares it to *C. stenoceras* (Robak) C. Moreau, which is usually pure white and much slower growing than C. pluriannulata. Robak (1932) points out the rather striking differences between these two species.

Caused minor decay (3.8%) in blackgum test blocks, and none in southern pine blocks.

Ceratocystis pilifera (Fries) C. Moreau, Rev. Mycol., Suppl. Colonial 17: 22. 1952.—This is a common and widespread species on pine lumber, and has been isolated from pine chips from Texas and Colorado. As mentioned, it differs from *C. pluriannulata* by its uniformly dark substratum and slightly slower growth, slightly longer period for perithecia to mature, and on the lack of abundant annuli on somewhat shorter necks. Cultures (RWD-947-2B) usually have a white powdery-to-cottony surface layer. *Ceratocystis pluriannulata* perithecia may mature in 7 days, whereas those of *C. pilifera* usually take 9 to 11 days. Griffin (1968) lumps these species together, but we consider them to be distinct.

Caused only minor amount of decay (5.6%) in blackgum test blocks, and none in southern pine blocks.

Ceratocystis ips (Rumbold) C. Moreau, Rev. Mycol., Suppl. Colonial 17: 22. 1952.—This species was one of the most common in blue-stained pine chips from Texas. Possibly much of the infection in green chips originated in pine logs invaded by *Ips* sp. beetles. However, the fungus was reported very common in

stained lumber at mills in the southern United States (Davidson, 1935; Verrall, 1939, 1941) and was not associated with log stain.

Caused minor decay (4.3%) in blackgum, and none in southern pine test blocks.

Ceratocystis sagmatospora E. F. Wright & Cain, Canad. Jour. Bot. 39: 1226. 1961.—Similar to *C. olivacea* (Mathiesen) Hunt and *C. obscura* (Davidson) Hunt. However, separated from *C. olivacea* mainly on the basis of ascospore shape and from *C. obscura* on ascospore size (Wright and Cain, 1961). A new genus, *Phialographium*, has recently been proposed by Upadhyay and Kendrick (1974) to accommodate the asexual stage of this fungus.

Previously described from bark beetle galleries on the sapwood and freshly cut surfaces of *Picea mariana* (Mill.) B.S.P., *Pinus resinosa* and *P. strobus* L. (Wright and Cain, 1961). Present isolate (RWD 947-A) was obtained from pine wood chips from Texas.

Caused minor weight losses (5.2%) in blackgum wood test blocks, and no loss in similar blocks of southern pine.

Ceratocystis stenoceras (Robak) C. Moreau, Rev. Mycol., Suppl. Colonial 17: 22. 1952.—This species was originally described by Robak (1932) from recently processed pulp at mills in Norway and Sweden. Although an authentic culture was not available for comparison, our isolate (RWD 922) from a pine root fits the original description well. It is not the same as the species listed under this name from hardwoods by Davidson (1942).

The fact that it caused appreciable weight loss (16.8%) in blackgum wood test blocks, but no weight loss in southern pine blocks, is of considerable interest.

Ceratocystis tremulo-aurea Davidson & Hinds, Mycologia **56:** 794. 1964.—Similar to C. ***stenoceras,*** but ascospores of C. *tremulo-aurea* are 4.7–5.9 × 1.1–2.4 μm (Davidson et al., 1964), almost twice as long as those of *C. stenoceras.* Isolated from cankers on *Populus* sp. (RWD 565-A).

Weight losses in both blackgum and southern pine test blocks do not exceed those lost in controls due to normal leaching in culture.

Leptographium sp.—Isolated from *Pinus contorta* Dougl. wood chips from Oregon. This culture has growth rates similar to *Leptographium terborantis* Barras & Perry (RWD 985-B).

Caused minor weight losses (5.0%) in blackgum wood blocks, and no loss in southern pine test blocks.

Aureobasidium pullulans (de Bary) Arnaud, Ann. Mycol. 8: 475. 1910.—Culture (ME-RP-1) from stored *Pinus resinosa* pulpwood log prior to chipping, Madison, Wisconsin.

Caused minor weight losses (4.8%) in blackgum test blocks, and no loss in southern pine.

Phialophora sp.—Culture (ME-AL-1) from *Alnus rubra* Bong. chips stored in simulated laboratory chip piles (towers).

Culture dark gray with white margin, margin proper finely fimbriate. Mat slightly raised, tufted; growth slow, about 5 to 7 mm radius in 7 days. Conidiophores abundant, in bunches on short side branches, 7–20 μm long, tapered to a

smaller apex; conidia abundant 2–3× 3–6μm, hyaline, ovoid to short cylindrical, slightly narrowed to basal end.

Caused moderate decay (18.0%)in blackgum wood test blocks, and none in southern pine test blocks.

SUMMARY

A new species of *Ceratocystis* isolated from *Betula* sp. wood chips has been named and described. The species is typified by a crown of hyaline to light brown filaments surrounding the ostiole; hence the specific epithet "rostrocoronata" has been applied to it. In laboratory test the fungus was found capable of causing significant decay in blackgum wood but none in southern pine wood. Observations are provided on 17 other fungi isolated from stained wood chips or logs, including information on their capacity to produce soft-rot in blackgum and southern pine wood

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