

Pyropyxis, a new pyrophilous operculate discomycete with a *Dichobotrys* anamorph

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The monotypic genus *Pyropyxis* is described based upon the type species, *Peziza rubra* Peck. *Pyropyxis* is distinguished by deeply cupulate, sessile, pink to reddish-orange apothecia. The ectal excipulum is composed of thick-walled cells forming a *textura angularis* tissue with the outer layer of cells occasionally giving rise to hyaline, moniliform hairs. The hymenium contains two types of paraphyses; most are hyaline, but some are filled with orange, crystalline pigments. The ascospores are eguttulate when fully mature but contain two small polar guttules when immature. A *Dichobotrys* anamorph is produced in axenic culture.

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Le genre monotypique *Pyropyxis* est décrit à partir de l'espèce type, *Peziza rubra* Peck. Le *Pyropyxis* se distingue par ses apothèques profondément cupulées, sessiles et roses à orange-rougeâtre. La partie externe de l'excipulum se compose de cellules à paroi épaisse formant un tissu de type *textura angularis* avec une couche externe de cellules donnant occasionnellement naissance à des poils hyalins et moniliformes. L'hyménium contient deux types de paraphyse; la plupart sont hyalines mais quelques-unes contiennent des pigments oranges, cristallins. Les ascospores sont aguttulées à maturité mais contiennent deux petites guttules polaires quand elles sont jeunes. On retrouve, en culture, un anamorphe de type *Dichobotrys*.

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Introduction

While studying pyrophilous discomycetes in eastern Ontario I frequently collected a brightly colored species growing on burnt litter in a 1-year-old prescribed burn. The collections were tentatively assigned to the genus *Geopyxis*. Further examination and cultural studies revealed that this fungus differed from *Geopyxis* in the type and deposition of pigments in the paraphyses, ascospore guttulation, presence of moniliform hairs, and the formation of a *Dichobotrys* anamorph in axenic culture. A new monotypic genus is proposed to accommodate this discomycete.

Materials and methods

This study is based upon collections made by the author during the summer and fall of 1979. Additional herbarium materials were made available by the following herbaria: Oregon State University, Corvallis (OSU) and New York State Museum, Albany (NYS).

Cultures were isolated from fresh ascocarps by harvesting ascospores on potato carrot agar petri plates. Spores typically germinated within 48 h without special treatment. Monospore isolates were obtained by selecting single germinated ascospores and transferring them to potato carrot agar or potato dextrose agar. Cultures were grown at room temperature. Conidial sporulation was induced by irradiating the cultures with long-wavelength ultraviolet light for a period of 6–8 h per day.

A pigment analysis was carried out on dried material of *Pyropyxis rubra* (K.N. Egger No. 279). A crude pigment extract was made by grinding rehydrated material in acetone. Relatively nonpolar pigments (e.g., carotenoids) were isolated by extraction with petroleum ether. The ether phase was removed and washed twice with distilled water. The color of the ether extract was noted and a portion was analysed spectrophotometrically over a frequency range of 300–550 nm.

Description

Pyropyxis gen. nov.

Apothecia profunda cupulata, sessilia, rosea ad rubro-aurantia. Excipulum ectale ex *textura angularis* e cellulis hyalinis, crassitunicatis constitutis cum pilis brevibus, hyalinis,

moniliformibus. Excipulum medullosa ex *textura intricata* e hyphis septatis, hyalinis constitutis. Asci operculati, cylindrici, octospori, J–. Paraphyses biformae: pigmento absenti vel pigmento auranti acrySTALLI capienti. Ascosporae maturae eguttulatae, laeves, hyalinae. Ascosporae immaturae biguttulatae.

ANAMORPHOSUS: *Dichobotrys* Hennebert, *Persoonia*, 7: 193. 1973.

TYPUS: *Peziza rubra* Peck.

Apothecia deeply cupulate, pink to reddish orange. Ectal excipulum composed of thick-walled *textura angularis* tissue with the outer layer of cells giving rise to short, hyaline moniliform hairs. Medullary excipulum composed of septate, hyaline hyphae forming a *textura intricata* tissue. Asci cylindrical with a furcate base, operculate, octosporous, J–. Paraphyses filiform, mostly hyaline, but some are filled with orange, crystalline pigments. Ascospores smooth, hyaline, eguttulate at maturity but biguttulate when immature.

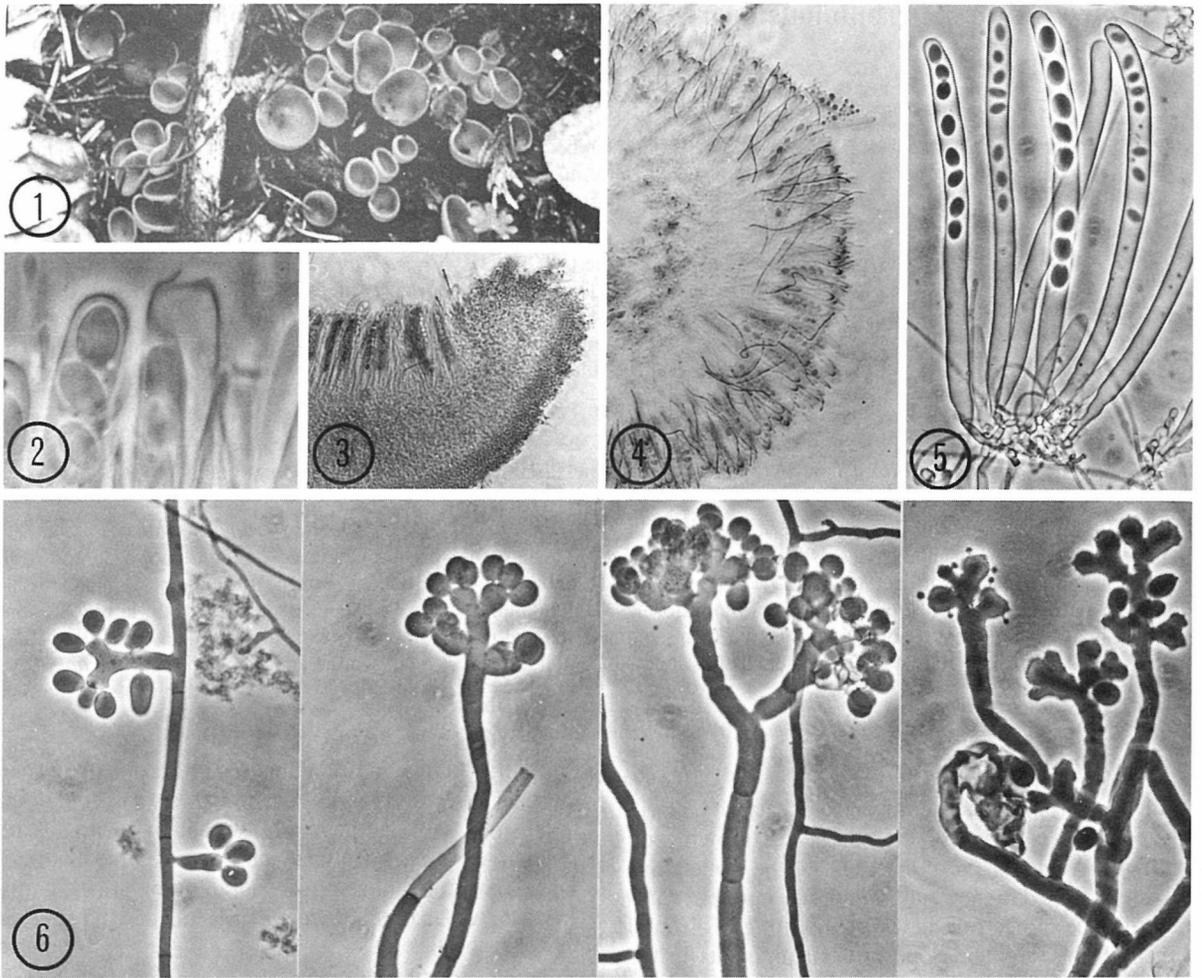
ANAMORPH: *Dichobotrys* Hennebert, *Persoonia*, 7: 193. 1973.

TYPE SPECIES: *Peziza rubra* Peck.

ETYMOLOGY: Greek, *pyros* = fire; Latin, *pyxis*, referring to the segregation of this genus from *Geopyxis*.

Pyropyxis rubra (Peck) Egger, comb. nov. Figs. 1–10
BASIONYM: *Peziza rubra* Peck, *Annu. Rep. N.Y. State Mus.* 24: 95. 1872.

Apothecia solitary to gregarious, deeply cupulate, sessile, 5–20 mm in diameter; hymenium at first yellowish pink turning reddish orange to brownish orange with age; receptacle yellowish pink to orange turning brownish orange, paler than the hymenium. Ectal excipulum composed of hyaline, thick-walled cells 8–14 μm in diameter forming a *textura angularis* tissue, the outer cells occasionally giving rise to short, hyaline, simple or branched, moniliform hairs 34–76 \times 5–13 μm . Medullary excipulum composed of hyaline, septate, thin-walled, branched hyphae 4.5–8.5 μm wide forming a *textura intricata* tissue. Asci cylindrical with a furcate base, 180–225 \times 10–14 μm , operculate, octosporous, not blueing in iodine.



FIGS. 1-6. *Pyropyxis rubra* (Peck) Egger. Fig. 1. Apothecia. $\times 0.5$. Fig. 2. Ascus apex showing operculum. $\times 700$. Fig. 3. Section of apothecium. $\times 65$. Fig. 4. Squash mount of hymenium showing pigmented paraphyses. $\times 65$. Fig. 5. Asci and ascospores. $\times 265$. Fig. 6. *Dichobotrys* anamorph. $\times 400$.

Paraphyses filiform, $2.4-3 \mu\text{m}$ wide with the apex slightly swollen up to $5 \mu\text{m}$ wide. Most paraphyses hyaline, but some filled with orange, crystalline pigments (see Fig. 4). Ascospores smooth, hyaline, elliptical, $(12.5-13.5) \times 7.5-9.5 \mu\text{m}$, eguttulate when mature but containing two small polar guttules when immature.

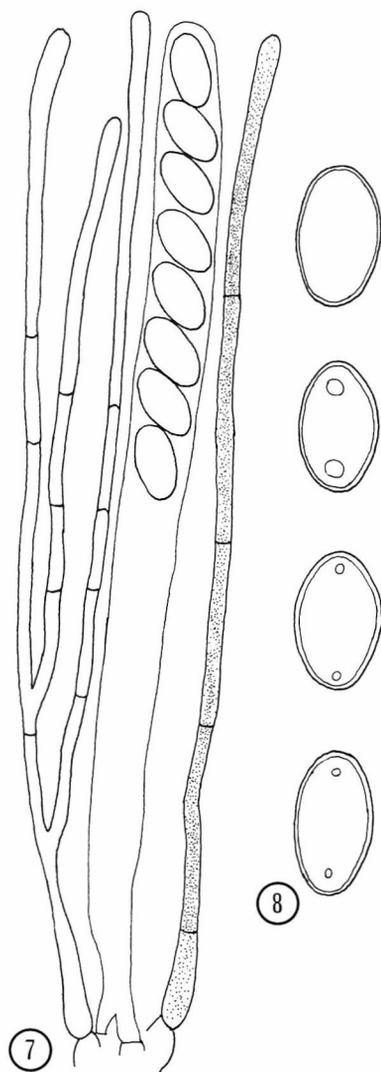
Colonies on potato dextrose agar fast growing, reaching 8 cm diameter in 5 days at 22°C , effuse with a floccose overgrowth of white mycelium, developing a pale-orange pigmentation when exposed to ultraviolet light. No conidia produced. Colonies on potato carrot agar reaching 9 cm diameter in 5 days at 22°C , effuse with a loose, thin overgrowth of white mycelium, developing a very pale orange pigmentation when exposed to ultraviolet light. Conidial production sparse under normal room lighting but becoming abundant under ultraviolet illumination. Mycelium variable in width, $2.4-16.8 \mu\text{m}$, immersed and superficial, sparingly septate, not or slightly constricted at the septa, hyaline to pale brown, smooth to echinulate or verrucose with roughening consisting of short, broad warts. Occasionally forming clumps of pale-brown, moniliform hyphae immersed in the agar and black stromata on the surface. Conidiophores $30-125 \times 5-11 \mu\text{m}$, smooth, hyaline, septate, slightly constricted at the septa, usually dichotomously branched at the apex with swollen ampullae up to 14

μm wide. Conidia produced blastically from short denticles on the ampullae, subglobose to elliptical, $7.5-13.5(-18) \times 5.5-9 \mu\text{m}$, pale brown at maturity, nonseptate, smooth to echinulate, with a short basal apiculus.

SUBSTRATE AND HABITAT: Scattered to caespitose on charred litter in recently burned mixed coniferous and deciduous forest.

DISTRIBUTION: CANADA: Ontario, British Columbia. U.S.A.: New York.

COLLECTIONS: U.S.A., NEW YORK, Ulster Co. near Highland, June 1870, C.H. Peck, holotype at NYS. CANADA: ONTARIO, Renfrew Co. ($77^\circ 35' \text{N}$, $45^\circ 14' \text{W}$), 21 August 1979: DAOM 178727 (K.N. Egger 268); DAOM 178728 (K.N.E. 270); DAOM 178729 (K.N.E. 272); DAOM 178730 (K.N.E. 278); 2 September 1979: DAOM 178731 (K.N.E. 279); DAOM 178732 (K.N.E. 283); DAOM 178733 (K.N.E. 289); DAOM 178734 (K.N.E. 290); 16 September 1979: DAOM 178735 (K.N.E. 320); DAOM 178736 (K.N.E. 323); DAOM 178737 (K.N.E. 325); 7 October 1979: DAOM 178738 (K.N.E. 328); DAOM 178739 (K.N.E. 329); DAOM 178740 (K.N.E. 333); DAOM 178741 (K.N.E. 334); DAOM 178742 (K.N.E. 335); BRITISH COLUMBIA: 1 mi (1 mi = 1.609 344 km) south of Sicamous, 26 June 1971, OSU 33348 (H. Larsen F-206). Cultures of K.N.E. numbers 268, 272, 320, and 334 are deposited in the culture collection at DAOM.



FIGS. 7, 8. *Pyropyxis rubra* teliorph. Fig. 7. Unpigmented paraphyses, ascus with ascospores, pigmented paraphysis. $\times 700$. Fig. 8. Ascospores: one mature and three immature with guttules. $\times 1400$.

Discussion

A pale yellowish orange pigment was present after extraction of dried ascocarps with petroleum ether. Spectrophotometric analysis indicated that the predominant compound present had an absorption peak at 326 nm, which is well below the absorption range for carotenoids. Oxidation of the carotenoid pigments upon drying and storage of the ascocarps may be responsible for the low reading. Conclusive proof of the presence of carotenoids in the fruiting bodies of *Pyropyxis* will have to await pigment extraction from fresh ascocarps.

Cooke (1875–1879) placed *Peziza rubra* Peck in synonymy with *Peziza araneosa* Bull. after examining Fuckel's Fungi Rhenani Exsiccati No. 2389. I have examined this exsiccati specimen and disagree with his decision. The spores of No. 2389 are larger than *P. rubra* (up to $10 \times 20 \mu\text{m}$), the paraphyses do not contain crystallized pigments, the ectal excipulum is not composed of thick-walled *textura angularis*, and there is no reference to *P. araneosa* occurring on burnt substrates.

Pfister (1979) examined the type of *P. rubra* and placed it in synonymy with *Geopyxis carbonaria* (Alb. & Schw.) Sacc.

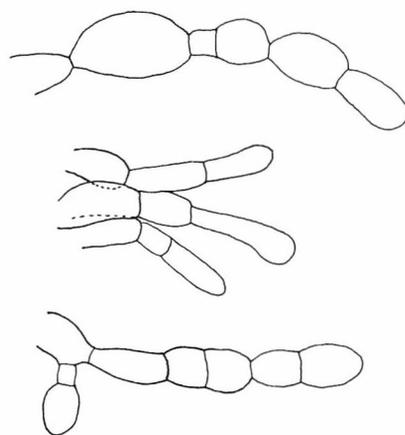


FIG. 9. Moniliform excipular hairs. $\times 700$.

However, I follow the concept of *G. carbonaria* as described by Rifai (1968) and *Pyropyxis* can be distinguished from *Geopyxis* sensu Rifai. At maturity *Pyropyxis* and *Geopyxis* share a number of similarities: the structure of the ectal excipulum, eguttulate ascospores, and pyrophilous habit. However, there are a number of significant differences between the two genera. *Pyropyxis* produces a *Dichobotrys* anamorph in axenic culture, while *Geopyxis* produces a *Hansfordia* anamorph. The ascospores of *Pyropyxis* contain guttules when immature, while *Geopyxis* ascospores do not contain guttules at any stage of their development. This suggests that the eguttulate spores of *Pyropyxis* and *Geopyxis* differ in some aspects of development and metabolism. The pigments of *Geopyxis* are dissolved in lipid globules in the paraphyses and are evenly distributed throughout the hymenium. The pigments of *Pyropyxis* are crystalline and are only present in a small proportion of the paraphyses in the hymenium. In balance I feel that there are enough significant differences to warrant segregation of *Pyropyxis* from *Geopyxis*.

Dissing and Sivertsen (1983) proposed a new genus, *Rhodotarzetta*, to accommodate *Pustularia rosea* Rea. Immature ascocarps of *Pyropyxis* may be confused with *Rhodotarzetta rosea* (Rea) Dissing and Sivertsen because of the biguttulate spores and pyrophilous habit. However, the ectal excipulum of *P. rubra* is composed of thick-walled *textura angularis* tissue rather than *textura intricata*, and the spores are eguttulate when mature.

Pyropyxis shares some characters with *Pulvinula*. Some members of the genus *Pulvinula* have hairs, the asci are usually furcate at the base, ascospores vary from eguttulate to multi-guttulate or uniguttulate, and many are pyrophilous (Pfister 1976). However, *Pulvinula* has globose spores, the paraphyses are usually curved, and the ectal excipulum is composed of thin-walled *textura globosa*. *Aleuria* also has hyaline hairs. However, *Aleuria* has reticulate or warty spores with prominent guttules and an ectal excipulum composed of thin-walled cells. *Octospora* has an ectal excipulum composed of *textura intricata* or *textura epidermoidea* which distinguishes it from *Pyropyxis*. *Caloscypha fulgens* produces a blastic anamorph, *Geniculodendron*, and has eguttulate spores. However, the ascospores are globose, the ascocarps stain greenish when bruised, and it is not pyrophilous.

Dichobotrys anamorphs have been reported in the genus *Trichophaea* (Hennebert 1973). The anamorph of *Pyropyxis* has echinulate conidia, which differentiates it from other

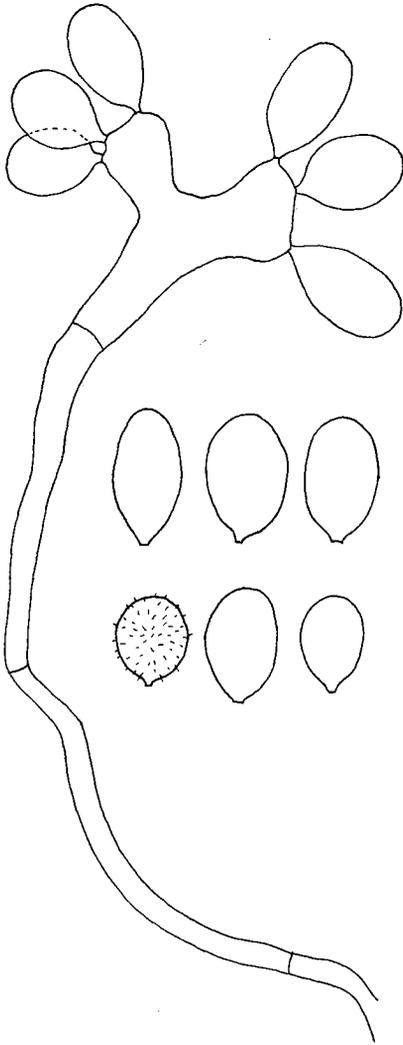


FIG. 10. *Pyropyxis rubra* (Peck) Egger conidial anamorph. Conidiophore and six conidia, one showing echinulate roughening. $\times 1400$.

Dichobotrys species. *Trichophaea* and *Pyropyxis* do not appear to be closely related.

I tentatively place *Pyropyxis* within the group of carotenoid-producing Pezizales: family Pyronemataceae, tribe Aleurieae sensu Korf (1973); family Aleuriaceae sensu Arpin (1969). Conclusive proof for this position must await pigment analysis of fresh ascocarps. It appears to be most closely related to *Geopyxis* but is also allied with *Pulvinula*, *Aleuria*, and *Rhodotarzetta*. *Pyropyxis* may provide a link between *Geopyxis* and the guttulate genera of the tribe Aleurieae.

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