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TAXONOMY OF THREE CANKER-CAUSING FUNGI OF HONEY LOCUST IN THE UNITED STATES

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The anamorph of *Thyronectria austro-americana*, a causal agent of honey locust canker, is shown to be *Gyrostroma austro-americanum* and *Kaskaskia gleditsiae* is reduced to synonymy.

Three fungi are known to cause cankers of honey locust (*Gleditsia triacanthos* L.) in the United States; *Thyronectria austro-americana* (Speg.) Seeler, *Nectria cinnabarina* (Tode:Fr.) Fr., and *Kaskaskia gleditsiae* Born & Crane. Honey locust has many attributes which make it a popular shade tree such as, attractive lacy foliage and fast growth. Cultivars of *G. triacanthos* var. *inermis* (Pursh) Schneid., which are commonly used, lack both seed pods and thorns found on the common honey locust. These seedless, thornless cultivars are often chosen to replace elms that are lost due to Dutch elm disease. Because of the popularity of honey locust as an urban shade tree, canker causing fungi have received attention.

Thyronectria austro-americana was first identified causing cankers of honey locust on Nantucket Island, MA (Seeler, 1940). More recently it has been found causing cankers of honey locust in Tennessee, Mississippi, Alabama, Colorado, and Kansas (Crandall, 1942; Hudler & Oshima, 1976; Crowe, Starkey & Lengkeek, 1982). *Nectria cinnabarina* has been known as a canker pathogen of many hardwoods since 1883 (Mayr, 1883). Pathogenicity on honey locust has only recently been established (Bedker, Blanchette & French, 1982). *Kaskaskia gleditsiae* was reported as a pathogen of honey locust in Illinois (Born & Crane, 1972).

Cankers caused by *N. cinnabarina*, *T. austro-americana*, and *K. gleditsiae* may be confused macroscopically if based on canker morphology. In order to determine the causal agent, the fungi must be examined microscopically. This paper presents a comparison of these three fungi and comments on their taxonomic status.

Isolates of *T. austro-americana* from Colorado

and Illinois were supplied by W. R. Jacobi and E. B. Himelick, respectively. The holotype of *T. austro-americana*, from the Cryptogamic Herbarium, New York Botanical Garden (NY), Bronx, NY, U.S.A., was examined. Additional herbarium material from the Farlow Herbarium of Harvard University (FH), Cambridge, MA, U.S.A., including collections by Seeler, were also examined.

Nectria cinnabarina was isolated on Difco potato dextrose agar (PDA) amended with streptomycin sulfate (20 p.p.m.) from cankers on honey locusts located on the St Paul Campus of the University of Minnesota, and identification was confirmed by Dr C. Booth, CMI, Kew. Fresh canker bearing stems of honey locust were collected and placed in a moist shaded area under a group of trees to stimulate perithecial development. Perithecia were observed after 3 months. Perithecia and sporodochia on host tissue and cultures were placed in herb. IMI and the CMI culture collection where they were assigned the accession numbers IMI 262934, 262935, 262936 and 262937. Perithecia and sporodochia on honey locust were also placed in the herbarium, Univ. Minnesota, Dept Plant Pathology, St Paul (MPPD, coll. no. B81-101).

An isolate of the *K. gleditsiae* (ATCC 122647 type culture) and herbarium material (ILL.S 34832, the holotype) were examined.

Cultures were routinely maintained on PDA at 25 °C in the dark prior to examination. Growth rates of *T. austro-americana*, *N. cinnabarina*, and *K. gleditsiae* were compared at temperatures ranging from 10° to 40° at 5° C intervals in the dark in Petri dishes containing 22 ml of PDA.

Morphological characteristics of *N. cinnabarina*, *T. austro-americana*, and *K. gleditsiae* are given in

Table 1. Comparison of *Tubercularia vulgaris*, *Gyrostroma austro-americanum*, *Kaskaskia gleditsiae* and *Nectria cinnabarina*, *Thyronectria austro-americana* from host tissue*

Anamorphs	<i>T. vulgaris</i>	<i>G. austro-americanum</i>	<i>K. gleditsiae</i>
Conidiomata diam (mm)	1.0 ± 0.3 (0.5-1.8)	1.0 ± 0.3 (0.5-1.7)	1.6 ± 0.4 (1.0-2.3)
Conidiogenous cell length (µm)	5.0 ± 3.2 (2.1-10.6)	10.8 ± 2.5 (5.8-18.0)	8.6 ± 1.1 (6.9-11.7)
Conidiogenous cell width (µm)	0.9 ± 0.2 (0.5-1.3)	1.5 ± 0.3 (1.1-2.1)	1.5 ± 0.3 (1.1-2.1)
Conidium length (µm)	7.0 ± 1.1 (5.3-10.6)	3.2 ± 0.6 (2.4-4.2)	3.4 ± 0.6 (1.3-4.8)
Conidium width (µm)	1.9 ± 0.3 (1.6-2.6)	1.4 ± 0.4 (0.8-2.7)	1.3 ± 0.3 (0.8-1.9)
Teleomorphs	<i>N. cinnabarina</i>	<i>T. austro-americana</i>	
Ascus length (µm)	69.4 ± 12.5 (55.9-88.4)	60.7 ± 5.7 (50.7-74.1)	
Ascus width (µm)	10.2 ± 1.7 (6.5-14.3)	11.0 ± 1.8 (6.5-14.3)	
Ascospore length (µm)	18.1 ± 2.4 (11.7-22.1)	12.5 ± 1.3 (9.8-15.6)	
Ascospore width (µm)	5.2 ± 1.0 (2.6-7.8)	7.1 ± 1.0 (5.2-9.1)	

* The figures in the table represent the averages plus or minus the standard deviations followed by the ranges in parentheses. Averages and standard deviations were determined by taking 50 measurements.

Table 2. Comparison of *Nectria cinnabarina*, *Thyronectria austro-americana*, and *Kaskaskia gleditsiae* in culture

	<i>N. cinnabarina</i>	<i>T. austro-americana</i>	<i>K. gleditsiae</i>
Hyphe diam (µm)	3.0 ± 1.1 (1.6-5.8)	2.9 ± 1.0 (1.6-5.3)	3.4 ± 1.3 (1.6-5.8)
Conidiogenous cell length (µm)	10.4 ± 1.9 (5.8-14.3)	8.4 ± 2.0 (5.3-16.4)	9.4 ± 1.4 (6.9-14.3)
Conidiogenous cell width (µm)	2.1 ± 0.3 (1.6-2.7)	1.9 ± 0.3 (1.3-2.7)	1.6 ± 0.3 (1.1-2.1)
Conidium length (µm)	5.1 ± 0.9 (3.4-6.9)	2.6 ± 0.3 (1.6-3.2)	3.1 ± 0.6 (2.1-4.8)
Conidium width (µm)	1.7 ± 0.2 (1.6-2.4)	1.4 ± 0.3 (1.1-2.1)	1.1 ± 0.3 (0.5-1.6)

* Figures in the table represent the averages plus or minus the standard deviations followed by the ranges in parentheses. Averages and standard deviations were determined by taking 50 measurements.

Tables 1 and 2 (from host material and cultures, respectively) and in Figs 1-13. Growth rates of these 3 fungi are presented in Fig. 14.

Nectria cinnabarina and *T. austro-americana* can be differentiated by ascospore morphology,

morphology of the asexual fruiting structures, and cultural characteristics. Both *N. cinnabarina* and *T. austro-americana* have perithecia that are clustered on a stroma (Figs 1, 2). The ascospores of *N. cinnabarina* are characterized by having a

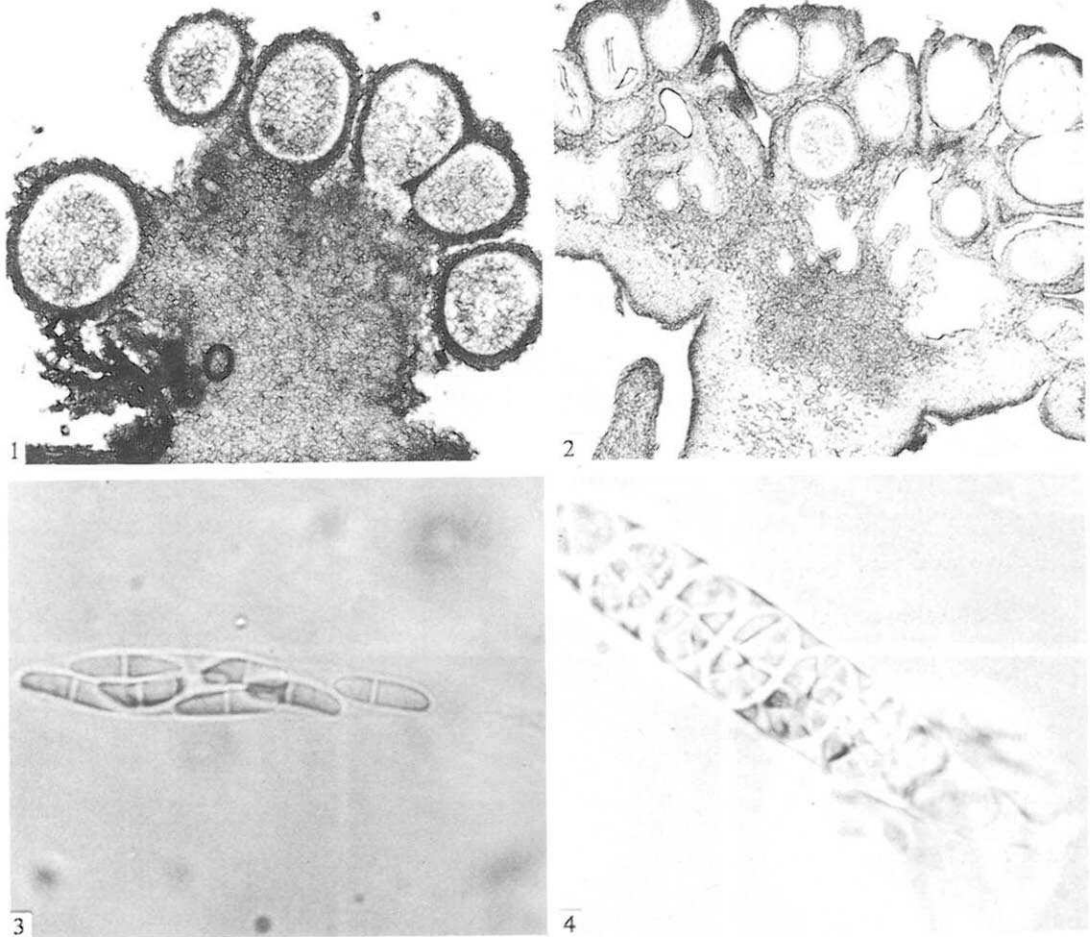


Fig. 1. Perithecia (v.s.) of *Nectria cinnabarina* clustered on a stroma, $\times 55$.

Fig. 2. Perithecia (v.s.) of *Thyronectria austro-americana* clustered on a stroma, $\times 40$.

Fig. 3. Ascus and ascospores of *Nectria cinnabarina*, $\times 910$.

Fig. 4. Ascus and ascospores of *Thyronectria austro-americana*, $\times 1470$.

single transverse septum (Fig. 3) while those of *T. austro-americana* have both transverse and longitudinal septa (Fig. 4). The anamorph of *N. cinnabarina*, *Tubercularia vulgaris* Tode:Fr., has long flexuous curled conidiophores (Fig. 5), whereas those of the anamorph of *T. austro-americana*, *Gyrostroma austro-americanum* Seeler, are straight and branched with long sterile attached structures (Fig. 6). The conidiophores of *K. gleditsiae* (Fig. 7) are similar to those of *G. austro-americanum*. The conidiophores of *T. vulgaris* are produced in sporodochia (Fig. 8), in contrast those of *G. austro-americanum* and *K. gleditsiae* are produced in stromatic conidiomata

(Figs 9, 10, respectively). *Nectria cinnabarina* and *T. austro-americana* can also be differentiated by cultural appearance and their growth rates. Based on these same characteristics it is difficult to differentiate *T. austro-americana* from *K. gleditsiae*. Seven-day-old cultures of *N. cinnabarina* are white and fluffy in appearance (Fig. 11), whereas the same age cultures of *T. austro-americana* have a white waxy margin with a distinct slimy orange appearance near the centre due to abundant sporulation (Fig. 12). Cultures of *K. gleditsiae* (Fig. 13) are very similar to those of *T. austro-americana*. *Nectria cinnabarina* has an optimum growth at 20° and does not grow at

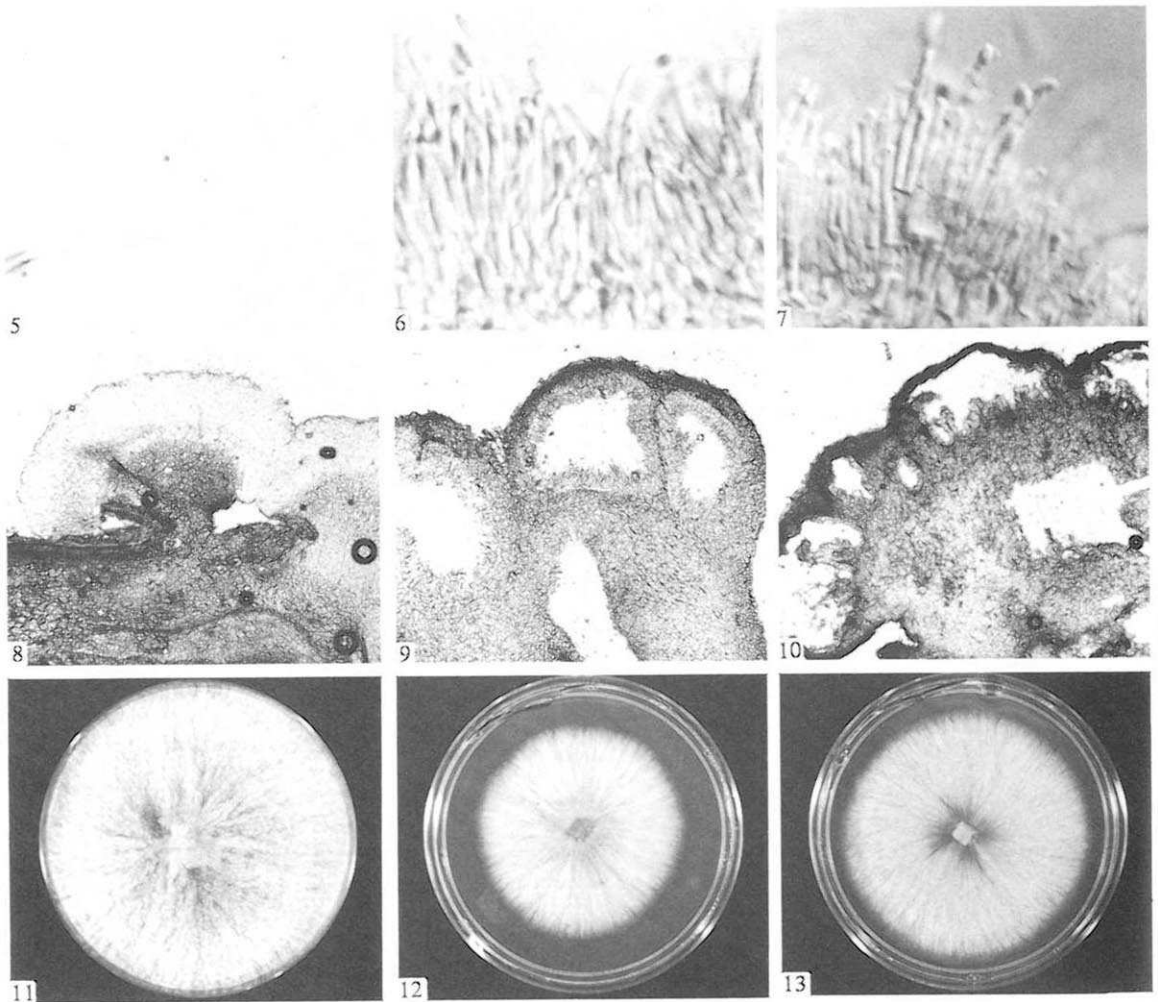


Fig. 5. Conidiophores of *Tubercularia vulgaris*, $\times 380$.
 Fig. 6. Conidiophores of *Gyrostroma austro-americanum*, $\times 1500$.
 Fig. 7. Conidiophores of *Kaskaskia gleditsiae*, $\times 1500$.
 Fig. 8. Sporodochium (v.s.) of *Tubercularia vulgaris*, $\times 40$.
 Fig. 9. Stroma (v.s.) of *Gyrostroma austro-americanum*, $\times 80$.
 Fig. 10. Stroma (v.s.) of *Kaskaskia gleditsiae*, $\times 40$.
 Fig. 11. Seven-day-old culture of *Nectria cinnabarina*.
 Fig. 12. Seven-day-old culture of *Thyronectria austro-americana*.
 Fig. 13. Seven-day-old culture of *Kaskaskia gleditsiae*.

temperatures above 35° (Fig. 14). *Thyronectria austro-americana* in contrast, has an optimum growth at 30° and is relatively thermotolerant with growth occurring at 40° . *Kaskaskia gleditsiae* has a growth curve that is very similar to *T. austro-americana* (Fig. 14).

The observations made in this study lead us to

conclude that the anamorph of *T. austro-americana* and *K. gleditsiae* are conspecific based on conidial ontogeny, size (Tables 1, 2), shape (Figs 9, 10), and colour. This is further supported by their similarities in cultural appearance (Figs 12, 13) and growth rates (Fig. 14). We therefore propose the following synonymy:

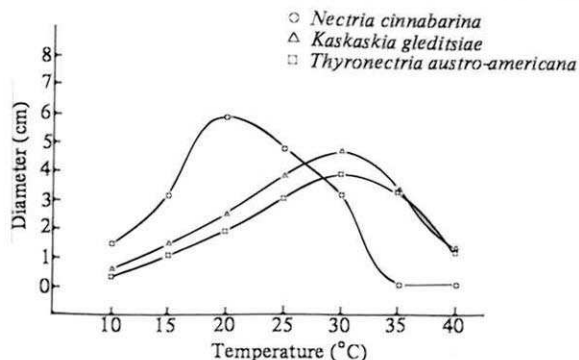


Fig. 14. Comparison of growth rates (diam) of -○- *Nectria cinnabarina*, -□- *Thyronectria austro-americana*, and -△- *Kaskaskia gleditsiae* at various temperatures in culture after 4 days.

GYROSTROMA AUSTRO-AMERICANUM Seeler (as '*austro-americana*'), *J. Arnold Arbor. Harv. Univ.* 21: 447 (1940).

Kaskaskia gleditsiae Born & Crane, *Phytopathology* 62: 926 (1972).

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