

# Pigment chemistry and morphology support recognition of *Cortinarius austrocinnabarinus* sp. nov. (Fungi: Cortinariaceae) from Australia

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## Introduction

The late Melvyn Gill (1946-2004) (Burns *et al.* 2005) and co-workers carried out extensive research on the pigment chemistry of Australian macrofungi, especially on anthraquinones from the basidiomycete genus *Cortinarius* (Pers.) Gray (Gill 1995, 2001). One of the groups of pigments discovered contained novel anthraquinones (including austrocorticin) based on a propionate-triggered octaketide assembly, in contrast to the more usual acetate-primed biosynthesis (Gill & Giménez 1988). Many of the collections investigated by Gill were not identified to species at the time, due to the lack of monographic treatments of the genera concerned. The material from which austrocorticin was isolated (Watling 19352 in E) was indicated by Gill and Giménez (1988) as belonging to a 'new taxon' in *Cortinarius* subgenus *Dermocybe* (Fr.) Loudon. Gill and Giménez (1988) further noted that the fungus was the same as that 'wrongly' illustrated under the name *Cortinarius cinnabarinus* Fr. by Cole *et al.* (1978) and Fuhrer (1985).

*Cortinarius cinnabarinus*, a species originally described from Europe, was placed by Moser (1974) and earlier workers in *Cortinarius* subgenus *Dermocybe* due to the presence of anthraquinone pigments. However, on the basis of pigment chemistry, morphology, cytology and ecology, Høiland (1983) proposed that a more appropriate placement for *C. cinnabarinus* was alongside *Cortinarius bulliardii* (Pers.: Fr.) Fr. in section *Armillati* Moser, within subgenus *Telamonina* (Fr.) Loudon, and molecular data confirm this placement (Garnica *et al.* 2005). A second species, *C. californicus* A.H.Sm., is considered to be close to *C. cinnabarinus* based on morphology and pigment profile (Keller 1982; Høiland 1983; Keller & Ammirati 1983; Ammirati 1989; Høiland & Holst-Jensen 2000). In various studies based upon DNA sequence data (Liu *et al.* 1997; Chambers *et al.* 1999; Høiland & Holst-Jensen 2000; Seidl 2000), *C.*

## Abstract

*Cortinarius austrocinnabarinus* R.H.Jones & T.W.May sp. nov. is proposed to accommodate Australian collections previously assigned to either *Cortinarius cinnabarinus* or *Dermocybe cramesina*. Among these collections is the voucher from which novel anthraquinone pigments, including austrocorticin, were initially isolated. The presence in Australia of *Cortinarius cramesinus* comb. nov. is confirmed. Thin-layer chromatography patterns differ significantly between *C. austrocinnabarinus* and *C. cramesinus*, and there are also macro- and micro-morphological differences.

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*californicus* is placed in a clade with other members of subgenus *Telamonia* outside subgenus *Dermocybe*. Thus, there would appear to be at least two distinct groups within *Cortinarius* in which a range of species produce anthraquinone pigments.

The image reproduced under *Cortinarius cinnabarinus* in Fuhrer (1985) was included in a later reprinting of *The Field Companion to Australian Fungi* (Fuhrer 1993) captioned *Dermocybe cramesina* E.Horak. Horak (1988) described *D. cramesina* from New Zealand, with a 'cinnabar red or crimson red' pileus. However, Gill (1995) and Morgan (1998) examined pigment extracts from the type collection of *D. cramesina* by thin-layer chromatography, and found that there were no features in common with extracts from the Australian material designated as *C. cinnabarinus* (Watling 19352 in E) from which austrocorticin and related compounds had been isolated.

Pigment chemistry, molecular data, morphology and ecology were incorporated in a comprehensive investigation of Australian species of *Cortinarius* subgenus *Dermocybe* and other *Dermocybe*-like members of *Cortinarius* (Jones 2003). On the basis of that study, plus evidence from published work on pigment chemistry (e.g. Gill & Giménez 1988; Gill 1995; Morgan 1998), we propose a new species for Australian material previously misidentified as *Cortinarius cinnabarinus* or incorrectly assigned to *Dermocybe cramesina*, and also confirm the occurrence of *C. cramesinus* in Australia.

## Materials and Methods

### Morphology

Macroscopic information was recorded from fresh material collected from areas of natural vegetation in Victoria and southwest Tasmania. The *Methuen Handbook of Colour* (Kornerup & Wanscher 1978) was used to record basidiome colour from fresh material prior to air-drying collections using a Hydraflow Ezidri Ultra 1000 dehydrator. Collections are lodged at MEL. Representatives from herbarium collections of mainly Australian material on loan from CSIRO–Wembley, E, MEL, and ZT were also examined, as were collections provided by D.A. Ratkowsky and G. Gates (Tasmania) and K. Syme (Western Australia).

Data on microscopic characters were recorded from

dried material rehydrated in 3% aqueous KOH (w/v). Following initial examination, 0.5% aqueous Congo Red (w/v) was added to these preparations to enhance contrast. Radial sections through the pileipellis were initially observed in water. Drops of 3% KOH were then added to the slide at the edge of the cover slip, and the KOH drawn under the slide by placing absorbent paper on the other side of the cover slip. Data on spores and basidia were taken from lamellae fragments of mature basidiomes. The designation A/B in square brackets before spore measurement data indicates A measurements from B collections. At least ten spores were measured from one or more basidiomes of each collection. Spore measurements were to the nearest 0.5  $\mu\text{m}$  and excluded ornamentation and the hilar appendage. The spore quotient (Q) for an individual spore is the spore length divided by the spore width. For each collection the mean values for spore length, spore width and spore quotient were calculated, and the range of means across collections is presented along with the grand mean across the means from each collection. Basidia measurements did not include sterigmata.

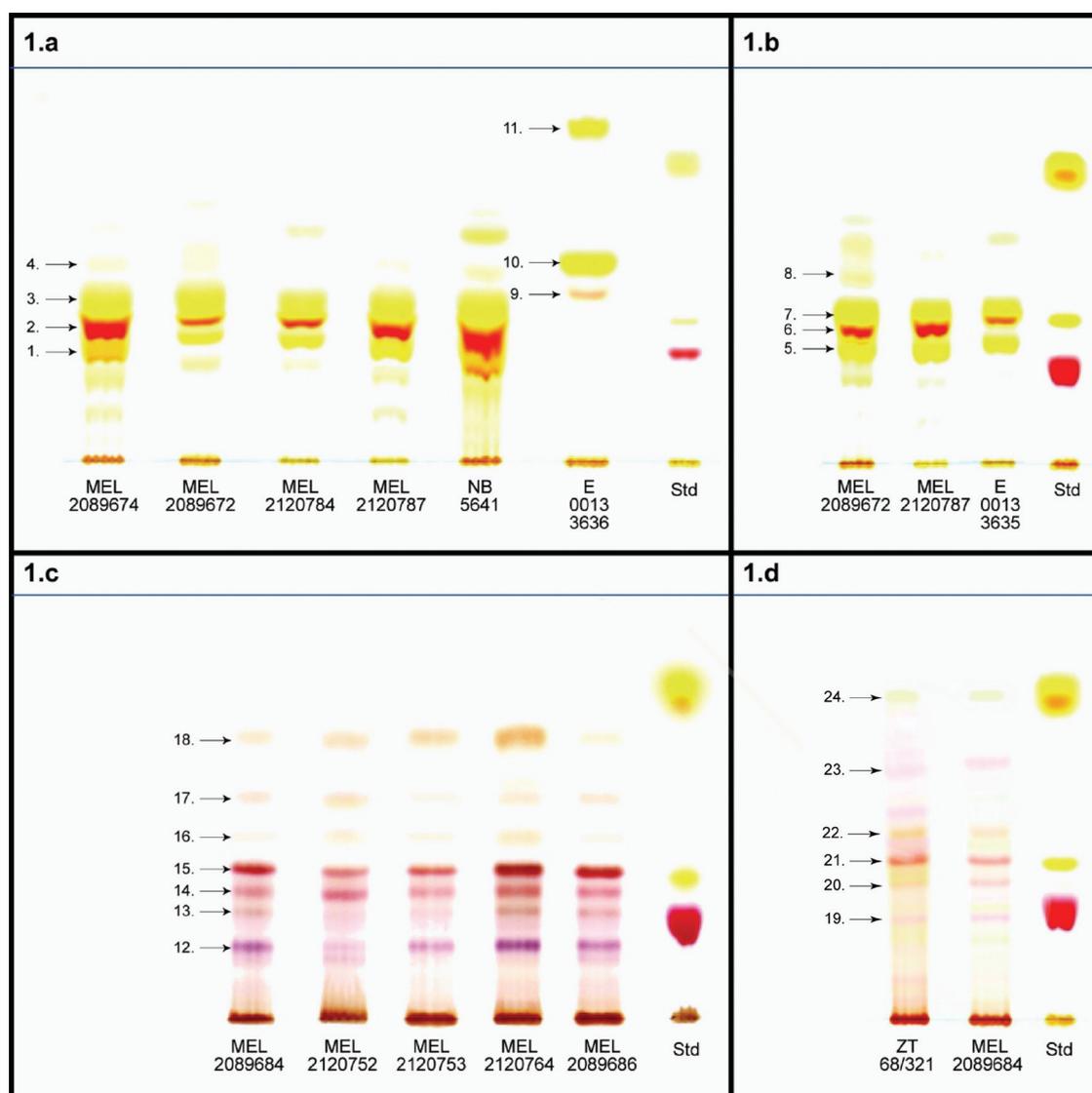
The ornamentation of spores was examined using Scanning Electron Microscopy (SEM). Air-dried spores taken from lamellae fragments of mature basidiomes were settled onto adhesive carbon tabs (Proscitech, Thuringowa, Qld) on the upper surface of 12 mm diameter stubs. Stubs were coated with gold using an Edwards S150B sputter coater (Dynavac, Bayswater, Victoria) and examined using a Philips XL30 FEG Field Emission Scanning Electron Microscope operated at 5.0 kV.

### Pigment chemistry

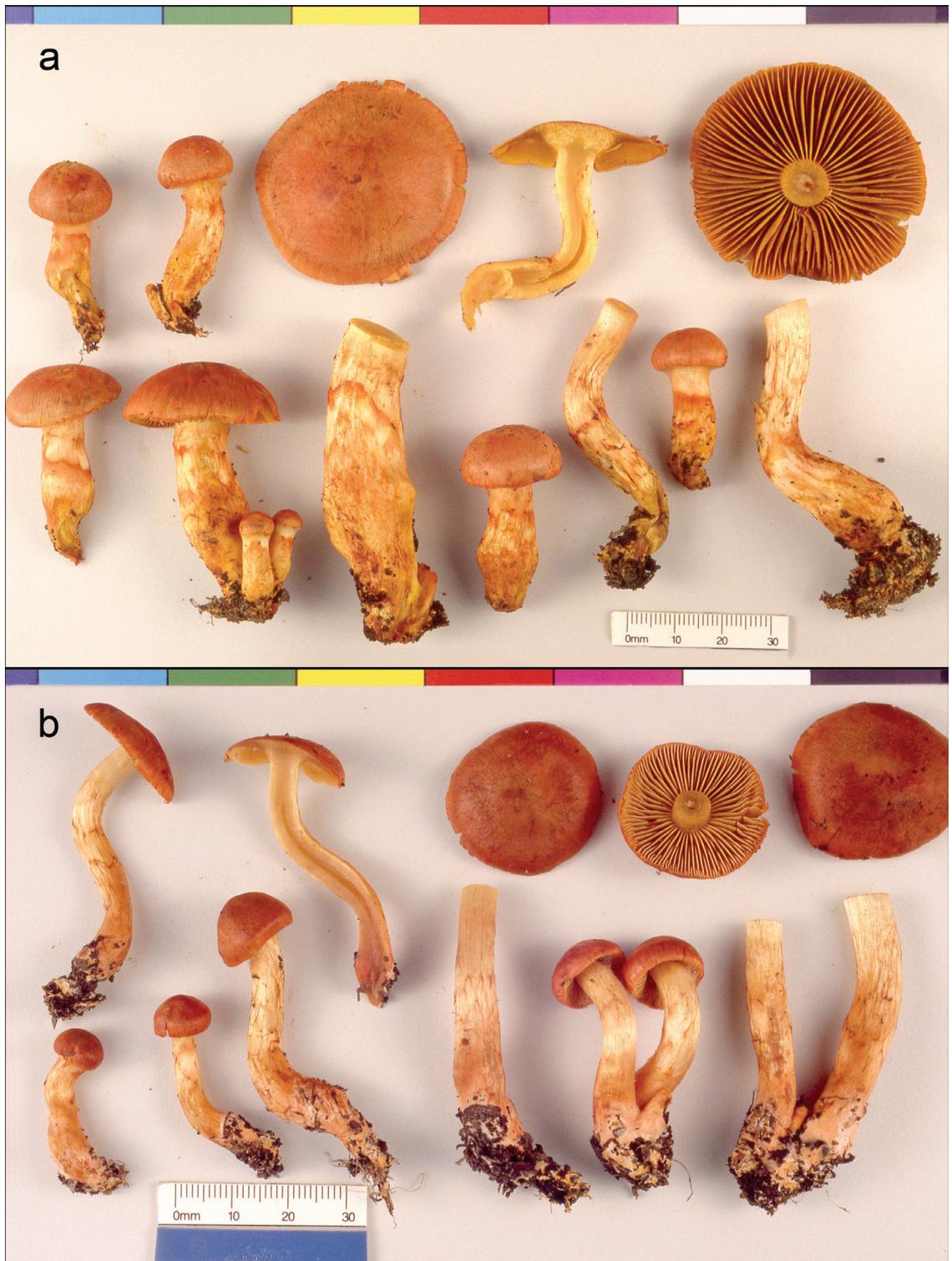
Methanolic pigment extracts were evaporated to dryness under gaseous nitrogen. Extract residues were reconstituted with a minimal amount of dichloromethane. Reconstituted extracts were subjected to thin-layer chromatography (TLC) on POLYGRAM SIL G/UV254 pre-coated plastic, 10  $\times$  20 cm plates with a silica layer thickness of 0.25 mm. A standard extract of *C. persplendidus* Gasparini (synonym *Dermocybe splendida* E.Horak), a species whose major pigments have been identified (Elsworth *et al.* 1999; Gill & Smedel 2000), was included on each plate. TLC plates were developed at room temperature

in a standard TLC chamber. The mobile phase solvent consisted of toluene:ethyl formate:formic acid at a ratio of 50:49:1, prepared in 500 ml batches and stored at room temperature. Saturation of the chamber was achieved by adding 50 ml of eluent to the chamber at least half an hour prior to the development of a plate. The  $R_f$  factor (Relative to Front) for each pigment band was calculated by dividing the distance moved by the band (measured from the middle of the band) by the distance moved by the solvent front.

It is well known in literature on thin-layer chromatography (e.g. Kidd *et al.* 1985) that the  $R_f$  of a given band may vary from one run to another. Taking this into account in a taxonomic study utilising TLC with numerous collections of Australian *Cortinarius*, Jones (2003) carried out a series of preliminary TLC runs in which samples of unknown extracts were run in various combinations. Extracts showing similar profiles were then run together on the same plate. Samples of putative taxa in the present study were thus eventually



**Figure 1.** Thin layer chromatography plates for pigment extracts. (a and b) *Cortinarius austrocinnabarinus* and a European collection of *C. cinnabarinus* (E 133636), (d and e) *C. cramesinus*, including the type (ZT 68/321) from New Zealand. The blue line indicates the solvent front. Std denotes the standard extract from *C. persplendidus*.



**Figure 2.** Basidiomes. (a) *Cortinarius austrocinnabarinus* (MEL 2089672), (b) *C. cramesinus* (MEL 2089686). Scale bar = 30 mm.

run together on individual plates, with the exception of the voucher *Watling 19352* (from Gill & Giménez 1988) and the type of *Dermocybe cramesina* for which the small amount of extract available precluded numerous runs.  $R_f$  values quoted in the text refer to the particular plate being discussed. Each distinct band on each TLC plate is numbered in bold. Results of chromatographic analyses are presented separately for each TLC plate. Some lanes have been deleted for illustrative purposes.

## Results

### Pigment chemistry

Two distinct patterns were observed (Fig. 1). In the first pattern (Fig. 1a) there was an orange band [**1**] (yellowish in profiles from more dilute extracts) at  $R_f = 0.27$ – $0.30$ , which abutted a red band [**2**] at  $R_f = 0.32$ – $0.33$ , which abutted a strong and broad yellow band [**3**] at  $R_f = 0.40$ . A faint pinkish yellow band [**4**] at  $R_f = 0.49$  is barely visible in all profiles except MEL 2120784. Weaker bands included a faint yellow band, visible just above the baseline in the profiles from more concentrated extracts, as well as some faster migrating yellow bands. An extract from the voucher *Watling 19352* (E 133635), cited by Gill and Giménez (1988) in their studies of austrocorticin, was run on a separate plate (Fig. 1b), along with MEL 2120787 and MEL 2089672. The profiles had an orange/yellowish band [**5**] at  $R_f = 0.27$ – $0.30$ , a red band [**6**] at  $R_f = 0.32$ – $0.33$ , and a yellow band [**7**] at  $R_f = 0.40$ . The profile of MEL 2089672 also had a pinkish yellow band [**8**] at  $R_f = 0.49$ . The major bands [**5,6,7,8**] of the samples run on the second plate (fig. 1b) corresponded very well to the major bands [**1,2,3,4**] in Figure 1a.

In the profile of a Danish collection (E 133636) of *C. cinnabarinus* (Fig. 1a) included in the study, the three visible bands were a pink band [**9**] at  $R_f = 0.42$  below a strong and broad yellow band [**10**] at  $R_f = 0.51$ , and a fast migrating yellow band [**11**] at  $R_f = 0.85$ . These bands did not match up to bands present in the first pattern described above, nor with profiles of the second pattern described below.

In the second pattern (Fig. 1c), all the main bands from the first pattern were absent. Figure 1c shows a slower migrating purple band [**12**] at  $R_f = 0.17$ , with

three evenly spaced brownish red bands [**13, 14, 15**] at  $R_f = 0.25, 0.29$  and  $0.35$  above. Band [**13**] is indistinct for the more dilute profiles of MEL 2120752 and MEL 2120753. In addition there were three greyish brown bands [**16, 17, 18**] at  $R_f = 0.43, 0.52$  and  $0.66$ , appearing at various levels of density over the five profiles. The overall pigment pattern was almost identical across the five collections.

A relatively dilute extract from the type of *D. cramesina* from New Zealand (ZT 68/321) was compared on a separate plate (Fig. 1d) against a similarly dilute extract from one of the collections (MEL 2089684) that had the second pattern. The profile of the *D. cramesina* type showed a number of points of similarity to MEL 2089684, sharing a weak purplish band [**19**] at  $R_f = 0.23$ . A brownish pink band [**20**] at  $R_f = 0.31$  is weak in ZT 68/321 but distinct in the profile of MEL 2089684, below a brownish red band [**21**] at  $R_f = 0.36$ . With minor differences in colour and intensity, these three bands [**19, 20, 21**] are considered to correspond to, respectively, the lower purple band [**12**] and the upper two reddish brown bands [**14, 15**] in Figure 1c. Immediately above the group of major bands in Figure 1d there was a greyish brown band [**22**] at  $R_f = 0.44$ . Both profiles share a fast migrating weak pinkish mauve band [**23**] at  $R_f = 0.60$ ; in ZT 68/321 this band blurs upwards, and is preceded by a mauve band. A fast migrating, extremely faint yellow band [**24**] at  $R_f = 0.76$  was also shared by both profiles.

### Taxonomy

The two distinct chromatographic patterns correlated well with differences in macro- and micro-morphological characters of the collections examined. Two taxa, *C. austrocinnabarinus* and *C. cramesinus*, are recognised.

#### 1. *Cortinarius austrocinnabarinus* R.H.Jones & T.W.May sp. nov.

*Cortinarius cinnabarinus* sensu Australian authors; *Dermocybe cramesina* sensu Fuhrer (1993; 2001; 2005); *D. sp. Watling 19352* sensu Gill (1995).

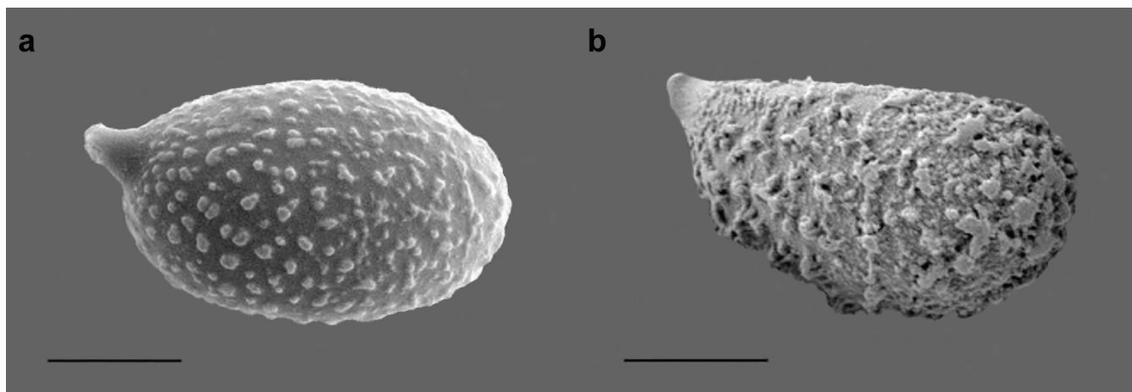
A *C. cinnabarinus* *sporis parvioribus*, 5.5–7.5(–8.5) × 4–6  $\mu\text{m}$ , *pilei infirme hygrophani et austrocorticino praesenti differt*.

**Type: AUSTRALIA: VICTORIA.** Kinglake NP, NW of intersection Melba H'way with Kinglake–Toolangi Rd, 30.vi.1985, T.W. May B365 & B.A. Fuhrer (holotype: MEL 2120784).

*Pileus* 17–55(–85) mm diam., hemispherical or conico-convex when young, becoming convex to plano-convex or subumbonate, often eventually broadly undulating towards edge, rarely upturned; deep orange red to red (8–9)(A–C)(7–8), becoming pastel red 7(A–B)5 on outer pileus towards maturity, sometimes the outer pileus colour fades at maturity leaving disc area darker, and pileus surface colour slightly patchy; surface dry; coarsely felty fibrillose, especially when young, more radially so on outer pileus; reddish orange universal veil remnants forming radial to subradial aggregations becoming sparse towards maturity, sometimes forming minute appressed scales, sometimes with veil remnants remaining attached to margin; weakly hygrophanous, not translucent-striate; margin sometimes exceeding lamellae, slightly inflexed when young, becoming straight, rarely recurved, entire to minutely eroded or rimose at maturity. *Lamellae* sinuate or occasionally adnexed; to 12 mm deep; moderately crowded; initially yellowish orange 5(A–B)(7–8), becoming brownish orange to reddish orange (6–7)(B–C)(7–8); edge entire, rarely irregularly denticulate or finely eroded, concolourous or occasionally blotchy, discolourous to darker red, especially at maturity. *Stipe* 34–87 mm long, 5.5–17 mm diam. at middle, to 20 mm diam. at widest part, cylindrical or attenuated upwards, often subfusoid and rapidly attenuated downwards

towards base; upper stipe light yellow to pale orange (4–6)A4, lower stipe appearing more reddish orange or yellowish red (7–8)A(7–8), often in zones of colour; surface dry; longitudinally fibrillose; sometimes rusty brown arachnoid cortina remnants on upper stipe of young specimens; orange red veil remnants forming fine to moderately coarse longitudinal aggregations, occasionally forming distinct subdowny bands, or a dense subdowny covering over large areas of the stipe surface of young specimens, otherwise aggregations becoming more dense on mid stipe, forming distinct bands or zones, often becoming more coarse on lower stipe; base with fine yellowish or pinkish orange downy covering, grading into coarser basal mycelium with bright orange rhizomorphs. *Context* in pileus, to 4 mm deep; pallid yellowish brown in disc area, more orange brown towards edge; in stipe, pallid pale yellowish orange to yellowish brown. *Odour* weakly spicy or chemical. *Chemical reactions* 3% KOH on pileus, rapid, strong, deep purple; on stipe, rapid, strong, purple; lamellae tissue in KOH solution leaching faint yellow to yellowish brown pigment. (Fig. 2a)

*Basidiospores* [285/12] 5.5–7.5(–8.5) × 4–6 μm, mean 6.27–7.36 × 4.67–5.45 μm (grand mean 6.74 × 4.93 μm), Q = (1.09–)1.25–1.56(–1.78), mean Q = 1.22–1.52 (grand mean 1.37), broadly ellipsoid to ellipsoid, occasionally elongate, amygdaliform or broadly amygdaliform; yellow-brown; ornamentation finely to moderately verrucose, the verrucae sparse to moderately dense, under SEM verrucae isolated and relatively rounded. Basidia 27–37 × 6.5–7.5(–9) μm, clavate, four or rarely two-spored, sterigmata to 4 μm long. *Cheilocystidia*



**Figure 3.** Scanning electron micrographs of spores. (a) *Cortinarius austrocinnabarinus* (MEL 2089672), (b) *C. cramesinus* (ZT 68/321, holotype). Scale bar = 2 μm.

and *Pleurocystidia* not present. *Lamellar trama* regular. *Pileipellis* a cutis or trichoderm consisting of repent, and often also some loosely interwoven and ascending, cylindrical, encrusted hyphae 4–8 µm diam., with yellow intracellular pigment (in KOH releasing bright purple pigment); with a differentiated subcellular subpellis, consisting of broader, short hyphae to 32 µm diam., with amorphous bright orange intercellular pigment masses (in KOH wine red). *Clamp connections* present in all tissues. (Figs 3a, 4a)

**Specimens examined:** AUSTRALIA: TASMANIA. Southwest Tasmania NP, The Wedge Forest Reserve, 15.iv.2000, G. Gates & D. Ratkowsky (R.H. Jones 104) (MEL 2314611); VICTORIA. Kinglake NP, Island Ck Picnic Area, Stringybark Tk near junction of Andrews Hill West Tk, 27.vii.1999, R.H. Jones 80 & N. Polikarpowski (MEL 2314610); Kinglake NP, Island Ck Picnic Area, Stringybark Tk near junction of Andrews Hill West Tk, 7.vii.2000, R.H. Jones 154 & K. Beattie (MEL 2314613); Narbethong–Marysville Road, 3.5 km NE Narbethong, Old Coach Rd turnoff, 25.vi.2000, R.H. Jones 150 (MEL 2089672); Kinglake Junction, 2.vi.1984, B.A. Fuhrer 899 (MEL 2052786); Kinglake NP, NW of intersection Melba H'way with Kinglake–Toolangi Rd, 19.vi.1986, T.W. May B384 & B.A. Fuhrer (MEL 2192221; duplicate is *Wat.[ling]* 19352 in E 133635, as 'Kinglake, coll. M. Gill [sic], T. May B384, July [sic.] 1986'); same locality, 22.vi.1986, B.A. Fuhrer 1125 (MEL 2053344); Buninyong, Union Jack Reserve, 21.vii.2000, R.H. Jones 167 & B. Andrews (MEL 2089674); 21.vii.2000, R.H. Jones 168 & B. Andrews, (MEL 2314614); Wombat SF, Blakeville–Bunding Rd, 500 m from Daylesford–Ballan Rd, 100 m N Blakeville–Bunding Rd, 9.vii.2000, R.H. Jones 155 & Field Naturalists Club of Ballarat (MEL 2089673); WESTERN AUSTRALIA. Dwellingup, Plavins Forest Block, off Murray River Rd, 9.vii.1996, N.L. Bougher E5641 (PERTH, ex CSIRO–Wembley); Walpole, Lochart Forest Block, W of Thompson Rd, 5.vi.2000, K. Syme 1055/00 (MEL 2120787; PERTH).

**Additional material examined:** *Cortinarius cinnabarinus*. DENMARK. Copenhagen, 22.ix.1980, collector illegible (Roy Watling 13716) (E 133636).

**Habitat:** Gregarious or often caespitose on soil in *Eucalyptus* dominated forest, often including *Leptospermum* spp.

**Coloured illustrations:** The following cited illustrations are likely to represent *Cortinarius austrocinnabarinus*: Cole *et al.* (1978: pl. 2, as *C. cinnabarinus* [photo is by Fuhrer]), Fuhrer (1985: p. 33, as *C. aff. cinnabarinus*; 1993: p. 33, as *D. cramesina* [see errata slip]; 2001: p. 33, as *D. cramesina*; 2005: p. 61, as

*D. cramesina*), Gill (1995: p. 2, as *Dermocybe* sp. *Watling* 19352 [photo is by Fuhrer, but not a photograph of this particular collection]) and McCann (2003: p. 21, as *D. cramesina*). In the illustration of *C. cinnabarinus* in Willis (1963: pl. 9) the pileus is a little too red, and the uniform deep reddish orange colour of the stipe is also not consistent with *C. austrocinnabarinus*. Voucher material specific to particular illustrations is not available to confirm identification, but note that several Fuhrer collections at MEL originally determined as *C. cinnabarinus* or *D. cramesina* are listed above under 'Specimens examined'.

**Notes:** Among Australian species of *Cortinarius* subgenus *Dermocybe* and other *Dermocybe*-like members of *Cortinarius* (see Jones 2003), *Cortinarius austrocinnabarinus* is characterised by the strong orange-red colour to the pileus, that often fades to pastel orange-red on the outer pileus at maturity, and the relatively small spores that are broadly ellipsoid to ellipsoid. The habit is often caespitose, and there are characteristically distinct bands or zones of coarse, orange-red veil remnants on the mid to lower stipe. Young basidiomes can be relatively squat, with the unexpanded pileus seated on a short, broad stipe. A unique feature associated with this taxon is the brownish orange colouration remaining on fingertips after the handling of basidiomes.

In addition to the distinct differences between pigment profiles for *C. austrocinnabarinus* and *C. cramesinus* discussed above, *C. austrocinnabarinus* can be distinguished from *C. cramesinus* by the pilei of *C. austrocinnabarinus* often developing a washed-out orange colour by maturity, whereas the orange colour in pilei of *C. cramesinus* is less intense at maturity, and is usually restricted to the margin of an often browner coloured pileus. Dried material of *C. austrocinnabarinus* retains a strong orange colouration in the pileus, whereas dried material of *C. cramesinus* is a dark vinaceous brown without orange tints to the pileus. The orange red veil remnants of *C. austrocinnabarinus* usually form coarser longitudinal and band-like aggregations on the stipe than do those of *C. cramesinus*, which has a pinkish basal mycelium (remaining visible in dried material) rather than the orange red mycelium of *C. austrocinnabarinus*. Spores of *C. cramesinus* are about the same length as those

of *C. austrocinnabarinus*, but are slightly narrower, and hence tend to be more often elongate than ellipsoid. Under SEM the verrucae of *C. austrocinnabarinus* are relatively rounded and isolated, whereas verrucae of *C. cramesinus* are more irregular in shape, with some interconnections (Fig. 3).

The identity of material from which austrocorticin and related pigments were isolated by Gill and Giménez (1988) has been confirmed as *C. austrocinnabarinus* by examination of the voucher material cited therein (collected from the type locality). The principal anthraquinones of *C. austrocinnabarinus* are austrocorticin, austrocorticinic acid and austrocorticone (Gill 1995). These compounds are the only naturally occurring quinones of the emodin/endocrocin type that have a two carbon side chain at the position C3. They are unique among anthraquinones from *Cortinarius* in having a propionate-triggered octaketide assembly (Gill & Giménez 1988).

The pileus colour of *Cortinarius austrocinnabarinus* is similar to the dark cinnabar-red to scarlet colouration of the European *C. cinnabarinus*, and the two species also share the presence of a subcellular subpellis (Høiland 1983). However, the latter species differs in the hygrophanous pileus and the larger spores up to 10 µm long (Høiland 1983), and in containing different major pigments, such as cinnarubin and fallacinol (Keller 1982), reflected in the different pigment profile (Fig. 1a). The North American counterpart of *C. cinnabarinus* is *C. californicus*, which differs from *C. austrocinnabarinus* by the reddish brown or dark reddish orange pileus that is hygrophanous, the longer spores, (7.4–)8–9.5(–11)

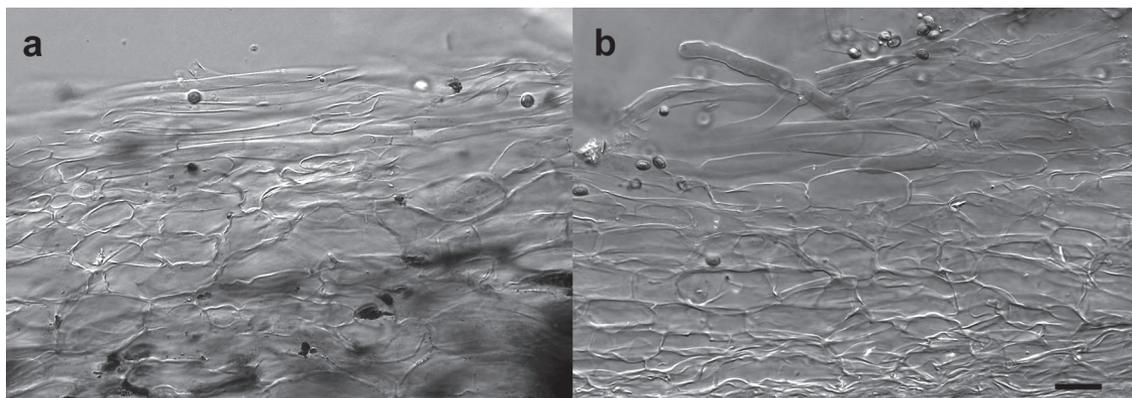
µm, the association with conifers (Ammirati 1989), and in having cinnarubin as its major pigment (Keller & Ammirati 1983). *Cortinarius hesleri* Ammirati & A.H.Sm. *nom. prov.*, a name used for a collection from North America similar to *C. californicus*, but associated with broad-leaved trees (Phillips 1991), also has cinnarubin as the major pigment (Keller & Ammirati 1983).

## 2. *Cortinarius cramesinus* (E.Horak) R.H.Jones & T.W.May, *comb. nov.*

BASIONYM: *Dermocybe cramesina* E.Horak *Sydowia* 40: 87 (1988).

In consideration of the perceived immaturity of the single collection on which the protologue is based, the following description is derived from characters of Australian collections, unless otherwise indicated.

*Pileus* to 50 mm diam., hemispherical or obtusely conical when young, becoming convex to plano-convex, occasionally with slightly undulating edge at maturity; orange red to brownish red 8(B–D)(6–7), sometimes more orange 7B7, edge often brighter orange (7–8)A(5–7); surface dry; felty fibrillose when young, becoming more radially fibrillose, often with fine radial aggregations of veil remnants, sometimes with minute appressed scales on mature specimens; rarely subhygrophanous, not translucent-striate; margin straight or occasionally inflexed, entire or occasionally finely rimose at maturity. *Lamellae* sinuate to narrowly adnate; to 5 mm deep; brownish orange (5–6)(B–C)(6–8); edge entire, rarely finely eroded towards distal part. *Stipe* to 80 mm long, to 8 mm diam. at middle, to 11 mm diam. at widest part, cylindrical or slightly attenuated



**Figure 4.** Pileipellis in cross section, mounted in 3% KOH. (a) *Cortinarius austrocinnabarinus* (MEL 2089673), (b) *C. cramesinus* (MEL 2089684). Scale bar = 20 µm.

upwards, often with finely subbulbous base; pale or light orange (5–6)A(3–4) upper stipe, to orange red (7–8)A(5–7) or sometimes more vivid (8–9)A(7–8) on lower stipe; surface dry; finely longitudinally fibrillose; cortina remnants sometimes forming an indistinct rusty brown band near stipe apex of younger specimens, not apparent on more mature specimens; orange red or brownish red veil remnants forming fine longitudinal to sublongitudinal aggregations, sometimes forming an indistinct band on upper to mid stipe, rarely with less distinct bands below, aggregations usually becoming denser and more arachnoid on lower stipe; basal area usually with pale pink or orange pink downy covering; basal mycelium orange pink. *Context* in pileus, to 2 mm deep, pallid yellow brown; in stipe, pallid yellow brown. *Chemical reactions* 3% KOH on pileus, rapid, strong, blackish purple; on stipe, rapid, strong, reddish purple; lamellae tissue in KOH solution leaching faint light brown pigment. (Fig. 2b)

*Basidiospores* [120/6]  $5.5\text{--}8.5\text{--}(10) \times 3.5\text{--}5.5 \mu\text{m}$ , mean  $5.80\text{--}8.15 \times 3.90\text{--}4.90 \mu\text{m}$  (grand mean  $7.36 \times 4.53 \mu\text{m}$ ),  $Q = (1.38\text{--})1.44\text{--}1.80\text{--}(2.00)$ , mean =  $1.49\text{--}1.68$  (grand mean 1.62) [in the type,  $6\text{--}7.5 \times 4\text{--}5 \mu\text{m}$ ,  $Q = 1.44\text{--}1.56\text{--}(1.67) \mu\text{m}$ , mean =  $6.7 \times 4.5$ , mean  $Q = 1.49$ ], ellipsoid to elongate, narrowly amygdaliform to amygdaliform; yellow-brown; ornamentation finely verrucose, verrucae moderately dense to dense, under SEM the verrucae are irregular in shape and show some interconnections. *Basidia*  $27\text{--}32 \times 6\text{--}7.5 \mu\text{m}$ , narrowly clavate, four-spored, sterigmata to  $5 \mu\text{m}$  long. *Cheilocystidia* and *Pleurocystidia* not present. *Lamellar trama* regular. *Pileipellis* a cutis or trichoderm consisting of repent or ascending (and then often in bundles), cylindrical, non-encrusted hyphae  $6\text{--}14 \mu\text{m}$  diam., with pinkish grey or pale orange intracellular pigment (in KOH greyish purple to strongly vinaceous or reddish purple); with a differentiated subcellular subpellis, consisting of broad, short hyphae, to  $31 \mu\text{m}$  diam., pale orange in mass or with orange intracellular pigment globules (in KOH purple). *Clamp connections* present in all tissues. (Figs 3b, 4b)

**Specimens examined: NEW ZEALAND: NELSON.** Lake Rotoiti, trail to St Arnaud Range, 30.iv.1968, E. Horak, ZT 68/321 (isotype); **AUSTRALIA: VICTORIA.** Kinglake NP, Island Ck Picnic Area, 7.vi.1999, R.H. Jones 53 (MEL 2089684); Kinglake NP, Masons Falls Picnic Area, 9.vi.2000, R.H. Jones 132 (MEL

2120764); Wombat SF, Old Farm Rd, Werribee Tk, 4.vii.1999, R.H. Jones 73 (MEL 2120752); Wombat SF, McCarthy Rd near Squires Ck turnoff, 4.vii.1999, R.H. Jones 74 (MEL 2120753); Wombat SF, Wombat–Barkstead Rd, 4 km from Barkstead, 300 m from road, 16.vi.2000, R.H. Jones 140 (MEL 2089686).

**Habitat:** In Australia gregarious on soil of *Eucalyptus* dominated forest; in New Zealand under *Nothofagus fusca* and *N. menziesii*.

**Notes:** Australian material is more robust (pileus to 50 mm diam., stipe to 65 mm long, to 8 mm diam. at middle, to 11 mm diam. at widest part) than the single collection from New Zealand (pileus to 30 mm diam., stipe to 50 mm long, to 4 mm diam.). Also, the pileus surface and lower stipe fibres are not as brightly coloured as in the New Zealand collection, in which they are 'cinnabar red or crimson red' (Horak 1988). Based on appearance of the type material obtained for inclusion in this study, it is possible that the New Zealand collection was immature, and hence smaller and with the colour more concentrated in the pileus and lower stipe. In other respects, including spore size and shape and the structure and pigmentation of the pileipellis, the Australian material conforms to the protologue.

There is sufficient similarity in pigment profiles of the type material and Australian material to suggest that it is prudent to assign the Australian collections to *C. cramesinus*. Differences between the pigment profiles of the *C. cramesina* type and the Australian collections are within the range of variation observed in other species (Jones 2003). Further collections of *C. cramesinus* from New Zealand are desirable in order to further investigate variation in basidiome colour and pigment chemistry within the species.

Analyses based on molecular data suggest that a new combination in *Cortinarius* for *Dermocybe cramesina* is necessary. In molecular analyses, most species formerly placed in *Dermocybe* are placed either in a clade containing only species of *Dermocybe* that is nested within a larger *Cortinarius* clade (Chambers *et al.* 1999; Høiland & Holst-Jensen 2000; Seidl 2000; Peintner *et al.* 2001, 2004; Garnica *et al.* 2005) or, in relatively few cases such as for *C. californicus* and *C. cinnabarinus*, are placed outside *Dermocybe sens. strict.* but remain within the broader *Cortinarius* clade (Høiland & Holst-Jensen 2000; Seidl 2000; Garnica *et al.* 2005). The relationships of New Zealand species assigned to *Dermocybe* by Horak (1988) have not yet been established from molecular data.

## Discussion

Data presented here confirm the finding of Gill (1995) and Morgan (1988) that the pigment pattern of *C. austrocinnabarinus* is quite distinct from that of *C. cinnabarinus* and *C. cramesinus*. In addition, none of the other species of Australian *Cortinarius* that were examined by Jones (2003) had similar pigment profiles to either *C. austrocinnabarinus* or *C. cramesinus*.

*Cortinarius cinnabarinus* is one of a growing number of species of ectomycorrhizal agarics originally described from Europe, now shown to have been erroneously reported from Australia (or to only occur there as exotics). For example, in *Cortinarius*, Gasparini (2001) described *Cortinarius austroviolaceus* Gasparini for Australian material previously identified as the Northern Hemisphere *C. violaceus* (L.) Gray; in *Russula* Pers., Grgurinovic (1997) considered that the collections that had been assigned by Cleland (1934) to six Northern Hemisphere species belonged in fact to independent species, newly typified on Australian material; and in *Laccaria* Berk. & Broome, May (1997) found that all collections previously assigned to *Laccaria laccata* (Scop. : Fr.) Cooke were erroneously identified, and this particular species occurred in Australia under exotic *Quercus* only. The presence of seemingly identical taxa from the Northern and Southern Hemispheres has complicated the naming of austral fungi, but it is now a reasonable assumption that austral ectomycorrhizal taxa are distinct from those of the Northern Hemisphere.

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