

Two new species of sequestrate *Agaricus* (section *Minores*) from Australia

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Received: 22 October 2012 / Revised: 2 December 2012 / Accepted: 7 December 2012 / Published online: 29 December 2012
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Abstract The novel species *Agaricus lamelliperditus* and *A. colpeteii* are described and illustrated, and affinities to other taxa determined by analysis of ITS sequence data. Both taxa resemble several other recently described Australian sequestrate *Agaricus* species, in particular *A. pachydermus*, *A. wariatodes* and *A. chartaceus*, which all have a powdery hymenophore from very early stages of basidiome formation. Both novel species have affinities to section *Minores*.

Keywords Arid habitats · Truffle-like · Cryptic diversity

Introduction

Agaricus is a large and important genus with over 300 species, of which more than 15 are sequestrate (Vellinga 2004; Geml 2004; Kirk et al. 2008; Moreno et al. 2010; Lebel and Syme 2012). Taxa are traditionally grouped into eight sections, and species differentiated based on reactions of basidiomata to air or various chemical reagents as well as subtle differences in morphology (Singer 1986; Mitchell and Bresinsky 1999; Kerrigan et al. 2006). The results of various phylogenetic analyses of species from temperate areas have caused some rearrangement of taxa; however, sections within the genus remain supported as distinct clades (Mitchell and Bresinsky 1999; Challen et al. 2003; Geml et al. 2004; Kerrigan et al. 2006). Zhao et al. (2011) found that with the incorporation of a large number of tropical taxa (86) into analyses, almost two-thirds of the species (56) could not be easily accommodated in traditional sections of *Agaricus*. However, Zhao et al. (2011) suggested

no major changes to the classification of temperate species, with the inclusion of tropical species, into eight traditional sections; though they did recognize seven strongly supported exclusively tropical clades.

Species in sections *Minores* and *Arvenses* can be found in temperate and tropical regions (Zhao et al. 2011), but also seem to be well-adapted to harsh environments (Geml et al. 2007, 2008; Lebel and Syme 2012). Both of these sections are characterized by anise or almond odours, yellow staining reactions of tissues, and in agaricoid taxa cap margins seldom strongly inrolled and rings typically pendant. Yellow staining of tissues also occurs in other sections such as *Xanthodermatei*; however, odours are lacking or different. Significant diversification of sequestrate forms worldwide has occurred in sections *Minores* and *Arvenses*, with the majority of taxa found in temperate or arid regions (Lebel and Syme 2012; Gube 2009; Geml 2004). Most of these taxa have been suggested to have affinities here as they exhibit yellow staining reactions of tissues and anise or almond odours.

Lebel and Syme (2012) described five new Australian sequestrate species, clarified nomenclatural issues, and, using molecular data, clarified relationships to sections within *Agaricus*. However, recent serendipitous collections from a remote arid area of Victoria and unidentified material from northern Western Australia were brought to our attention, which closely resembled several of the recently revised species. In this paper, two novel species are described and illustrated, and affinities to other taxa determined by analysis of ITS sequence data.

Materials and methods

Taxon sampling The region of nuclear rDNA data used for analyses included ITS1–5.8S–ITS2. Sequences representing

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a broad selection of species (36) within *Agaricus* sections *Minores* and *Arvenses* (including tropical taxa, Zhao et al. 2011), and representatives of sections *Agaricus* (5), *Sanguinolenti* (4), and *Xanthodermataei* (3) were retrieved from GenBank. Thirteen novel sequences representing five Australian species (four sequestrate and one agaricoid) were generated for this study. All collections for which new sequences were produced, with GenBank accession numbers, are listed in Table 1.

Nucleic acid preparation, amplification and sequencing - Genomic DNA was isolated with the EZNA forensic kit following the manufacturer's protocol. The targeted regions were amplified from purified DNA with standard fungal primer pairs ITS1F/ITS4 (White et al. 1990; Gardes and Bruns 1993). PCR protocols followed standard methods (Lebel and Tonkin 2007; Lebel and Syme 2012). Purified DNA was directly sequenced by MACROGEN (Korea) with primer pairs ITS1/ITS4. Assembly and manual editing of sequences were performed with Sequencher 4.3 (Gene Codes). Sequences were then transferred to BioEdit v7.1.3 (Hall 2011) for alignment. Alignments were automated with ClustalX v.2.0 (Thompson et al. 1997) and MUSCLE (Edgar 2004) for comparison and the alignment then manually edited. Alignment available in TreeBase (<http://purl.org/phylo/treebase/phylovs/study/TB2:S13635>).

Phylogenetic analysis Outgroups are represented by *Clarkeinda trachodes* and *Chlorophyllum agaricoides* and *Chl. brunneum*. Phylogenetic analyses were performed with Bayesian likelihood with the Metropolis coupled Markov chain Monte Carlo (MCMC) search algorithm implemented in MrBayes 3.1.2 (Ronquist and Huelsenbeck 2003). Gaps in alignments were treated as missing data. The resulting

consensus tree was visualised using FigTree (Rambaut 2009) and is presented with posterior probabilities.

Morphology Macroscopic characters were described directly from fresh material or based on examination of, and field notes on, dried collections. Colours are described in general terms. Fresh material was dried in a food dehydrator at 35 °C for 12 h. Habitat, associated plant communities and fruiting season are based on field notes. Chemical reactions of tissues were not tested. Hand-cut sections of fresh and dried material were mounted either in a 5 % aqueous solution of KOH, then stained with ammoniacal Congo red, or directly stained with Melzer's reagent. Measurements were made at $\times 400$ or $\times 1,000$ with a calibrated ocular micrometer. Spore and other dimensions are given as length range \times width range. For spores, the mean \pm standard deviation is also provided for length \times width ($n=15$ /collection). The length: width ratio (Q) of individual spores is presented as the range of Q values. Spore measurements do not include the hilar appendage. Dimensions of basidia are given as length range \times width range ($n=8-12$ /collection), and do not include the sterigmata. All illustrations are based on the type collection unless otherwise stated. Informal taxon names are standardized according to Barker (2005) and names of herbaria are abbreviated according to Thiers (<http://sweetgum.nybg.org/ih/continuously updated>).

Results

The ITS dataset, including the 5.8S gene, comprised 93 sequences of 791 characters long. Several of the major clades in sections *Arvenses* and *Minores* identified by Zhao et al. (2011) were recovered, although sampling was not as complete.

Table 1 Novel sequences produced for this study with Genbank accession numbers

Taxon	Herbarium accession no.	Collection no.	Geographic location	GenBank ITS no.
<i>Agaricus chartaceus</i>	MEL2231690		Coolgardie, WA	JX984570
<i>Agaricus colpeteii</i> TYPE	MEL2358009	TL2424	Mildura, VIC	JX984565
<i>Agaricus colpeteii</i>	MEL2358036	TL2384	Mildura, VIC	JX984566
<i>Agaricus colpeteii</i>	MEL2358044	TL2375	Mildura, VIC	JX984564
<i>Agaricus lamelliperditus</i>	PERTH	MDB F12/11	Kununurra, WA	JX984558
<i>Agaricus lamelliperditus</i> TYPE	MEL2363446	MDB F61/96	West Kimberly, WA	JX984559
<i>Agaricus lamelliperditus</i>	MEL2323501	P.Grey 107	SW of Bourke, NSW	JX984560
<i>Agaricus lamelliperditus</i>	MEL2323503	P.Grey 109	SW of Bourke, NSW	JX984561
<i>Agaricus lamelliperditus</i>	DNA	SD101	Simpson Desert, NT	JX984562
<i>Agaricus lamelliperditus</i>	MEL2363444	MDB F55/06	North Kimberly, WA	JX984563
	PERTH			
<i>Agaricus wariatodes</i>	MEL2339254	Trappe28631	Lockhart, NSW	JX984556
<i>Agaricus wariatodes</i>	MEL2339253	Trappe28634	Riverina, NSW	JX984557
<i>Agaricus sp.</i>	MEL2358037	TL2383	Mildura, VIC	JX984567

Support for deeper branches within sect. *Minores* is not that strong, so the emphasis is less on relationships between clades. Both novel species belong in a broadly interpreted section

Minores. *Agaricus lamelliperditus* T.Label & M.D. Barrett sp. nov. is part of a strongly supported clade (PP=1) of sequestrate taxa, with *A. wariatodes* (Grgur.) T.Label and *A. chartaceus*

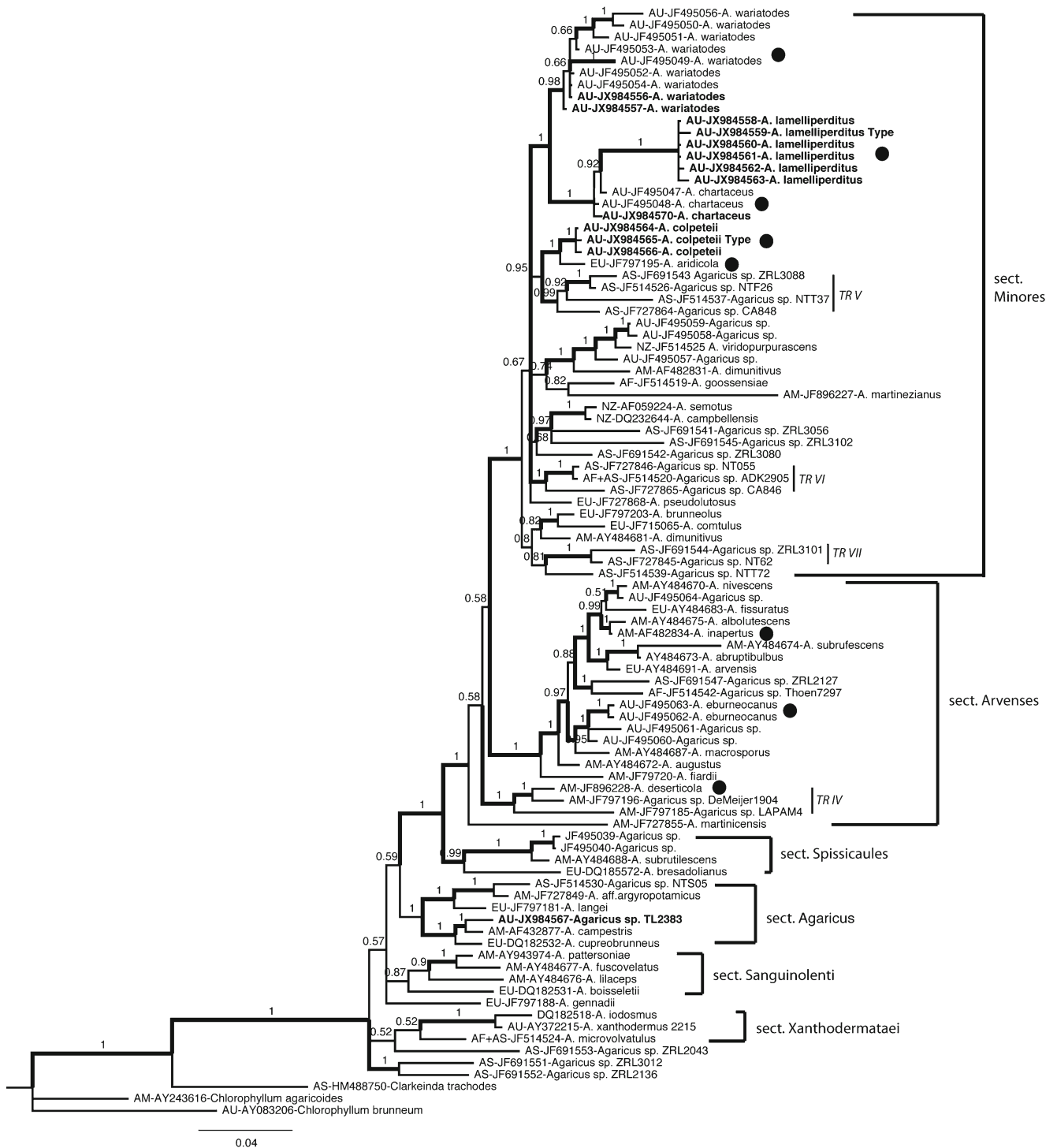


Fig. 1 Phylogenetic tree focused on *Agaricus* sections *Minores* and *Arvenses* based on a Bayesian analysis of ITS sequences. Outgroup taxa are represented by *Clarkeinda trachodes*, *Chlorophyllum agaricoides* and *Chl. brunneum*. Posterior probabilities $\geq 95\%$ are indicated by thicker lines. Novel sequences indicated by bold names, and

sequestrate taxa by a black dot. Branch labels: continent (Africa AF, Americas AM, Asia AS, Europe EU, Australia AU, New Zealand NZ); GenBank accession; and species name where known or collection no. Representative sections within *Agaricus* are indicated on the right hand side

T.Lebel as sister taxon (Fig. 1). *Agaricus colpeteii* T.Lebel sp. nov. is part of a strongly supported clade (PP=0.95), matching ‘TR V’ of Zhao et al. (2011), that includes four undescribed species from Thailand and the northern hemisphere sequestrate, *A. aridicola*, as sister taxon (Fig. 1). A sequence from an epigeal agaricoid species of *Agaricus* (TL2383) collected from one of the same sites, at the same time as some of the *A. colpeteii* material, is in a completely different section (sect. *Agaricus*).

The other sequestrate taxa included in the analysis, *A. inapertus* and *A. deserticola* from the northern hemisphere and *A. eburneocanus* from Australia, belong in separate strongly supported clades in sect. *Arvenses*.

Taxonomy

Key to sequestrate *Agaricus* species worldwide (novel species bold; non-Australian species roman)

- 1a Basidiomata with a strong smell of almonds, marzipan, cucumber, curry or anise.....2
- 1b Basidiomata without odour or with a different odour.....6
- 2a Basidiomata large, pileus 30–150 mm diam. and stipe 10–30 mm diam. × 15–150 mm long.....3
- 2b Basidiomata smaller, pileus 10–50 mm diam. and stipe 4–11 mm diam. × 10–40 mm long.....4
- 3a Stipe with a prominent veil, which ruptures to form rings and/or a sheathing pseudovolva; typically found in deep, sandy soils and dune systems.....
.....*Agaricus aridicola*
- 3b Stipe with a distinct veil, which typically does not rupture and a pseudovolva absent; found under conifers in mountains of western USA.....
.....*Agaricus inapertus*
- 4a. Hymenophore initially pale yellow becoming greyish tan but no darker; smell of curry
.....*Agaricus chartaceus*
- 4b. Hymenophore initially pale coloured, becoming dark olivaceous brown to greyish black; smell of marzipan or almonds and cucumber.....
.....5
- 5a. Pileus thin, remaining fleshy; spores globose to subglobose; basidia 4-spored.....
.....*Agaricus wariatodes*
- 5b. Pileus thin, becoming dry and papery; spores subglobose to ellipsoid; basidia none seen (even in immature specimens).....***Agaricus colpeteii* sp. nov.**
- 6a. Basidiomata large, pileus generally >40 mm diam. (up to 120 mm).....7
- 6b. Basidiomata small, pileus generally <25 mm diam. (up to 30 mm).....10
- 7a. Hymenophoral trama always staining reddish-orange and stipe-context reddening.....
.....*Agaricus erythrosarx*
- 7b. Hymenophoral trama and stipe-context not changing colour, or sometimes staining yellowish or orange-yellow or pinkish.....8
- 8a. Pileus thin, papery when dry; hymenophore pale yellow to tan or umber yellow, not darkening to brown or purple
.....***Agaricus lamelliperditus* sp. nov.**
- 8b. Pileus thick, fleshy; hymenophore pale brown becoming dark brown to purple brown or blackish at maturity.....9
- 9a. Spores 8–10 × 6–7.5 μm, subglobose to ellipsoid; semi-arid areas of southern Australia.....
.....*Agaricus melanosporus*
- 9b. Spores 4.5–7.5 × 5.5–6.5 μm, globose to subglobose; arid and semi-arid areas of western USA.....
.....*Agaricus deserticola*
- 10a. Pileus or hymenophore staining yellow when bruised.....
.....*Agaricus inilleasperus*
- 10b. Pileus or hymenophore not staining or becoming reddish.....11
- 11a. Stipe flesh white to cream gradually becoming very pale apricot, and with yellow stains at base; spores predominantly ellipsoid or ovoid 8–13.5 (–16) × 6–8 μm.....
.....*Agaricus eburneocanus*
- 11b. Stipe flesh not changing colour; spores globose to broadly ellipsoid 6.0–8.5 × 5–7.5 μm.....12
- 12a. Hymenophore pale brown to dark purple-brown, sublammellate to elongated labyrinthine; spores globose to subglobose.....
.....*Agaricus pachydermus*
- 12b. Hymenophore pale yellow to greyish tan or umber yellow, not darkening further, loculate only when very young, soon powdery; spores subglobose to broadly ellipsoid.....
.....***Agaricus lamelliperditus* sp. nov.**

***Agaricus colpeteii* T.Lebel sp. nov.**

Mycobank no.: MB801763

Differing from *A. pachydermis* by the yellow staining reaction of tissues, strong almond/cucumber odour and powdery hymenium early in development.

Type: Australia: Victoria. Neds Corner Station, SW corner of property near State Forest, 24 Nov. 2011, T.Lebel TL2424, HOLOTYPE (MEL).

Etymology: In honour of Colleen (col-) and Peter (pete-) Barnes, keen naturalists and current managers (2011) of Neds Corner Station, where the species was discovered. Many thanks also to the various volunteers and researchers on the BushBlitz discovery team including station hand ‘Bluey’, who aided in collecting and naming this species.

Illustrations: Figs. 2, 3

Basidiomata 8–33 (–48) mm high × 10–38 (–55) mm diam., mostly subglobose but sometimes globose, pyriform or elongated; apex generally convex, smooth or with some minor folds and indentations. *Pileus* white to pale cream or pale tan, bruising pale yellow when fresh, drying to pale greyish or silvery white (no colour changes apparent), smooth, faintly striate fibrous or with broad, appressed, barely visible concolourous scales, thin and papery, cracking easily when dry; margin remaining fused to stipe or breaking in small patches around stipe as sporocarp dries. *Context* 0.5–1.0 mm thick, 0.5–2.5 mm in younger specimens, white to cream-coloured, not changing colour. *Hymenophore* initially loculate, pale cream to pale tan, firm, moist, becoming dark olivaceous brown then eventually dark brown to greyish black, dry and powdery. *Stipe-columella* 5–11 mm long × 3–7 mm diam., percurrent or not, central, solid, smooth, projecting beyond hymenophore, cylindrical, tapering or with slightly bulbous base, cream to

pale tan coloured; context cream to pale tan coloured, no colour change on exposure or bruising. *Odour* strongly of almond and cucumber when fresh, none when dry; *taste* slightly acid.

Spores (6.0–)7.0–8.0 × 5.4–6.2 (–6.5) μm, mean 6.9 ± 0.53 × 5.9 ± 0.35 μm, Q=1.04–1.34, subglobose to (broadly) ellipsoid, sometimes obovate, often asymmetrical, smooth, thick-walled, hyaline to very pale yellow in KOH and weakly dextrinoid when young, tan brown to dark brown in KOH when mature. *Hymenium* totally disintegrated, with no visible structure. *Basidia* none observed. *Pileipellis* a cutis 18–58 μm wide, of compact subparallel to patchily interwoven hyaline hyphae 3–5 μm diam., overlying a context 31–90 μm wide, of slightly inflated hyphae 5–8 μm diam. No clamps observed.

Distribution: Emergent to epigeal, solitary or in small groups in sandy loam or clay soil, scattered in open spaces between saltbushes and small herbaceous plants in mixed chenopod shrubland on saltpan floodplain or mixed Black

Fig. 2 Basidiomata of *Agaricus colpeteii*. A. TL2375 (photo M. Cheng); B. Holotype TL2424 (photo T. Lebel)

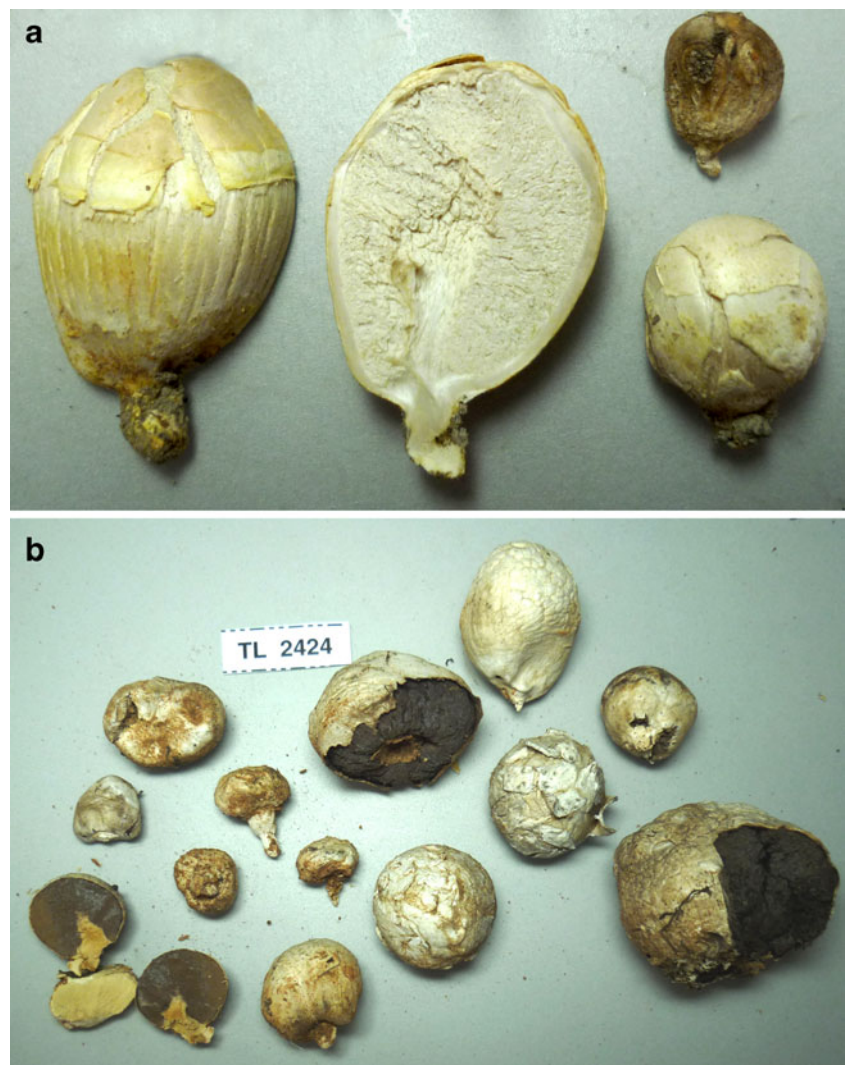
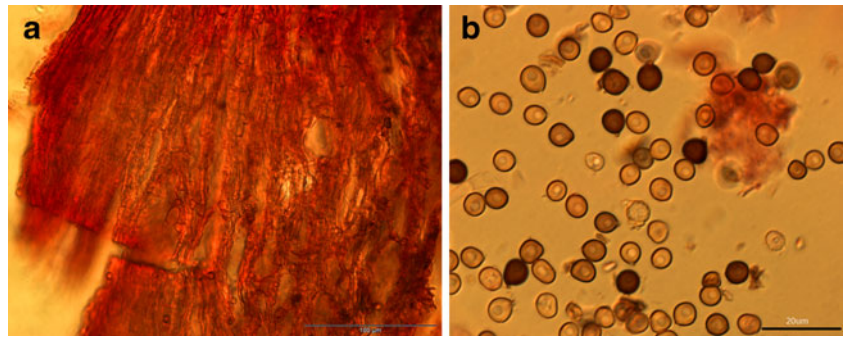


Fig. 3 Microscopic features of *Agaricus colpeteii* (Holotype). **a** Pileipellis and context hyphae; **b** basidiospores. (**a, b** stained in congo red and 3 % KOH)



Box forest and chenopod flats, rarely found under canopy of shrubs. Found in association with *Mareiana* spp, *Enchytraea* sp, and *Eucalyptus largiflorens*.

Other specimens examined: AUSTRALIA: Victoria. Neds Corner Station, SE corner of property, along southern boundary track, approx. 4.5 km from corner with Graves Track, 22 Nov. 2011, *T.Lebel TL2405*; SW corner of property on northern boundary of exclusion paddock, 29 Nov. 2011, *T.Lebel, M.Cheng, P.Simper, K.Harman TL2384*; near Snake Lagoon track, approx. 3.7 km from homestead, 30 Nov. 2011, *T.Lebel & 'Bluey' TL2375*. Fig. 4

Notes: *Agaricus colpeteii* resembles several other recently described truffle-like *Agaricus* species, in particular *A. pachydermus*. *Agaricus colpeteii* differs in the initially loculate hymenophore becoming powdery at young stages versus sublamellate to elongated labyrinthine, yellow staining

of pileus and context, the almond/cucumber odour, the less 'agaricoid' basidiome shape, and weakly dextrinoid reaction of the spores.

Basidiomata of *A. colpeteii* are generally found at the soil surface, or barely embedded, in chenopod vegetation floodplains. Analysis of molecular data confirms placement of *A. colpeteii* in a strongly supported section *Minores*, but in a different clade than the one containing *A. wariatodes*, *A. chartaceus* and *A. lamelliperditus*.

***Agaricus lamelliperditus* T.Lebel & M.D. Barrett sp. nov.**
Mycobank no.: MB801764

Differs from *A. wariatodes* in the hymenium remaining pale brown, the apparent lack of yellow staining of tissues and weakly spicy odour.

Type: Australia: Western Australia. West Kimberley Region, 300 m NW of Beverley Springs Station Homestead

Fig. 4 Distribution of five arid zone Australian sequestrate *Agaricus* species. *A. chartaceus*: black square; *A. colpeteii*: black circle; *A. lamelliperditus*: black rectangle; *A. pachydermus*: star; *A. wariatodes*: open circle with diagonal line

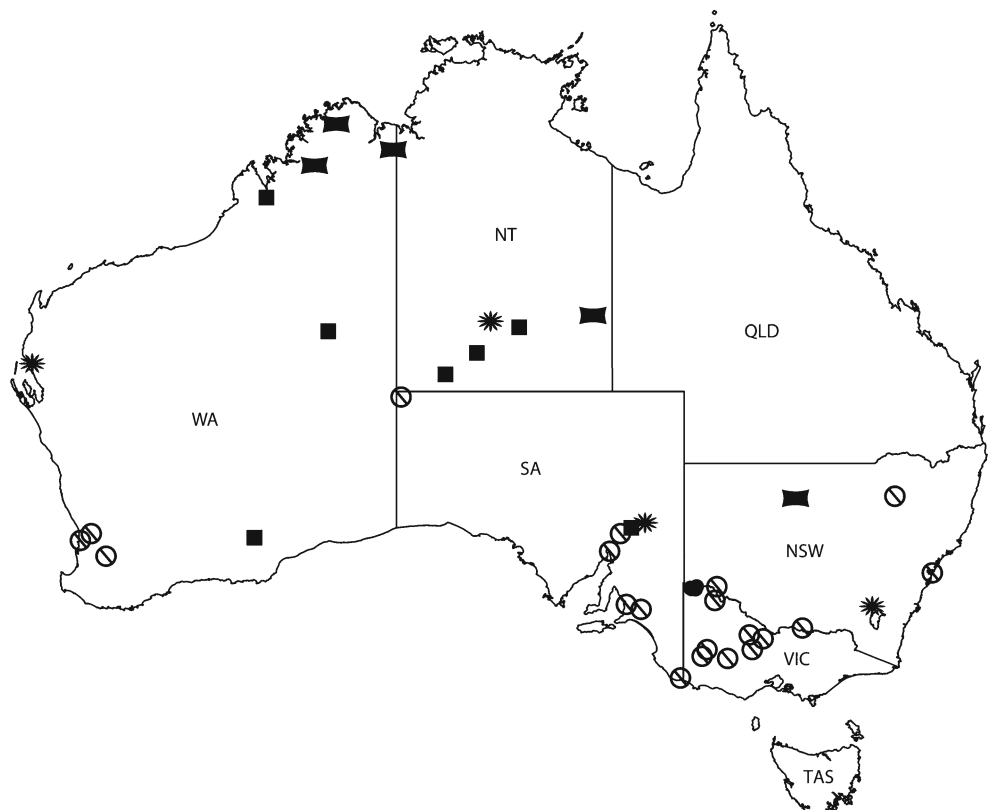
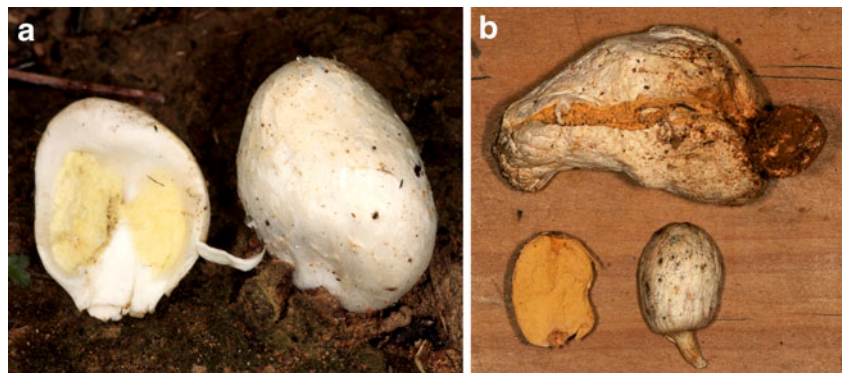


Fig. 5 Basidiomata of *Agaricus lamelliperditus*. **a** MDB F12/11; **b** MDB F55/06 (photos M.D. Barrett)



(now Charnley River Station), 12 Jan. 1996, *M.D. Barrett* F61/96 HOLOTYPE (MEL).

Etymology: ‘lost lamellae’ in reference to the lack of structure of the hymenophore which is powdery from early stages of sporocarp formation, lamelli = lamellae, perditus = lost (L).

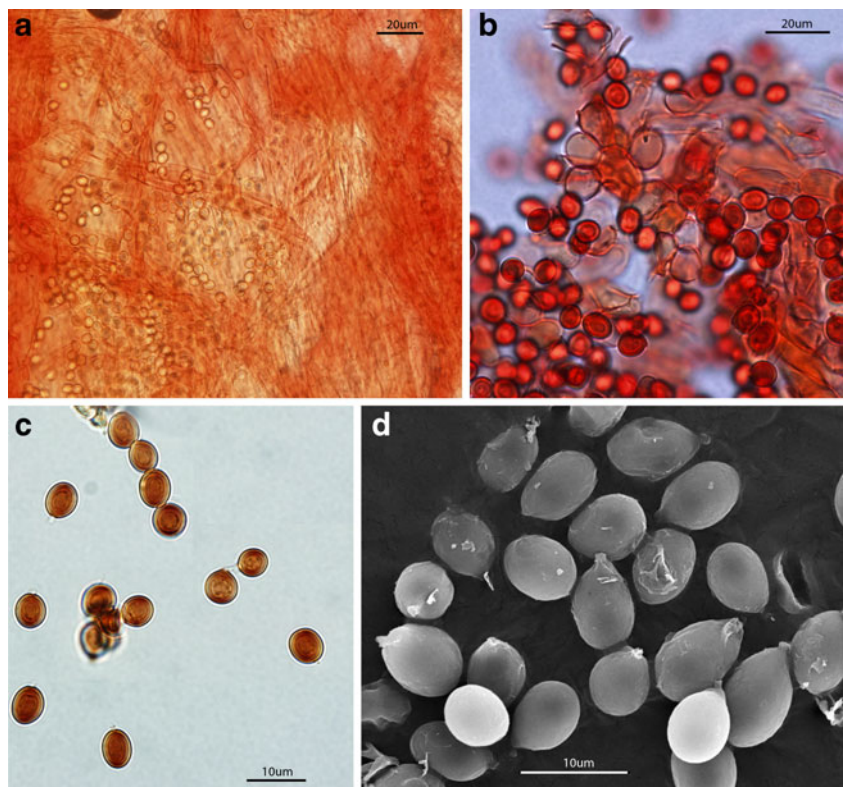
Illustrations: Figs. 5, 6

Basidiomata 12–40 mm high × 10–30 mm diam., irregularly subglobose to elongate, lobed or not; apex convex to plano-convex or sometimes indented slightly. *Pileus* white to pale cream or pale tan, silky smooth, thin, papery when dry, sometimes finely cracked, drying slightly wrinkled; margin remaining fused to stipe. *Context* 0.5–3.0 mm thick, 1–3 mm in younger specimens, white to cream coloured, not changing colour. *Hymenophore* pale yellowish becoming

greyish-tan or umber yellow but not darkening to brown or purple, initially minutely loculate but becoming powdery from very young stages. *Stipe-columella* 16–42 × 2–12 mm diam., percurrent or not, central, solid, fibrous projecting beyond hymenophore, cylindrical or with slightly bulbous base, cream coloured to pale greyish cream; context cream coloured, no colour change. *Odour* weak but not distinctive fresh, dried slightly spicy; *taste* not recorded.

Spores (6.5–)7.0–8.5 × 5.0–6.5(–7.5) μm, mean 7.2 × 6.1 μm, Q=1.0–1.3, globose, subglobose to broadly ellipsoid, mostly symmetric, smooth, thick-walled, hyaline to very pale yellow in KOH, weakly dextrinoid. *Basidia* 7–16(–18) × 8–12(–14) μm, clavate to broadly clavate; sterigmata mostly two or three, rarely four. *Pileipellis* a cutis 18–45 μm wide, of compact subparallel to patchily interwoven

Fig. 6 *Agaricus lamelliperditus* (Holotype). **a** Pileipellis and context hyphae; **b** basidia and basidiospores; **c** basidiospores; **d** SEM of basidiospores. (**a**, **b** stained in congo red and 3 % KOH; **c** Melzers reagent)



hyaline hyphae 3–5 µm diam., overlying a context 36–110 µm wide, of slightly inflated hyphae 5–8 µm diam. No clamps observed.

Distribution: Hypogeous to emergent, solitary or in small groups in red-loam, sandy loam or clay soil in disturbed sites beside roads or in a partly cleared paddock in grassy *Eucalyptus* woodland, *Eucalyptus/Melaleuca* woodland or open mulga woodland. In different habitats found in association with *Eucalyptus tetradonta*, *Sorghum* sp., *Chrysopsis setifolius*, *Acacia* spp., and Chenopodiaceae.

Other specimens examined: AUSTRALIA: Western Australia. North Kimberley region, at new Theda Station Homestead airstrip, 15 Jan. 2006, *M.D. Barrett & R.L. Barrett F55/06* [PERTH, MEL]; South of Kununurra, near old meatworks at end of Packsaddle Plains Rd, 10 Jan 2011, *M.D. Barrett F12/11* [PERTH]. New South Wales. Gundabooka N Park, (SW of Bourke), 1 June 2000, *P. Grey 109* [MEL2323503part A]; Gundabooka N Park, (SW of Bourke), 1 June 2000, *P. Grey 107* [MEL2323501]. Northern Territory. Northern Simpson Desert-Hay River Region, Mt Tietkens, Aust. Geog. Scientific Expedition, 11 July 2007, *E. Davison SD101 & SD95* [DNA]. Fig. 4.

Notes: *Agaricus lamelliperditus* resembles *A. wariatodes*, *A. chartaceus* and *A. colpeteii* both macro- and microscopically, however the smell is weakly ‘spicy’ rather than of curry, marzipan, or cucumber and the distribution is more northern and central Australian than most of the other species (Fig. 4). The hymenophore is powdery from a very early stage and does not darken, which also differentiates it from *A. pachydermus* and *A. colpeteii*. Analysis of molecular data, show the affinities of this species lie with sect. *Minores*, in a strongly supported clade with *A. wariatodes* and *A. chartaceus*, sister to *A. chartaceus*.

Discussion

The sequestrate fruit body form has arisen independently from agaricoid forms at least seven times within *Agaricus*, with the highest diversification (8 species) occurring in sections *Minores* and *Arvenses* (Lebel and Syme 2012; Gube 2009; Geml 2004). Zhao et al. (2011) suggested that the northern hemisphere sequestrate species *A. deserticola* and *A. aridicola*, were more closely related to various tropical taxa than to sections *Arvenses* or *Minores*. However, given short branch lengths and low bootstrap support on some clades, a broader interpretation of their results could be to include their tropical clades V, VI & VII within *Minores*. This would mean that *A. aridicola* would be retained within a broad section *Minores*. On the other hand *Agaricus deserticola* is in a strongly supported clade ‘TR IV’, with two Brazilian species, close to a strongly supported sect. *Arvenses* clade (Zhao et al. 2011). More sampling of taxa across tropical and temperate Australia

and south east Asia will aid in further identifying major clades within the genus.

Of the nine Australian sequestrate species, *A. eburneocanus* has affinities to sect. *Arvenses*, and *A. colpeteii*, *A. chartaceus*, *A. lamelliperditus*, and *A. wariatodes* to a broadly defined sect. *Minores* (Fig. 1). No sequence data are currently available for *A. pachydermus*; however, the lack of odour and yellow staining of tissues would tend to exclude this species from either sect. *Arvenses* or *Minores*. Although *A. colpeteii* closely resembles all three species in the *A. wariatodes* clade, its affinities are with a completely different group of tropical species from Thailand (Fig. 1).

Differentiation of species is difficult within agaricoid taxa; with the reduction or loss of morphological features in sequestrate taxa, the use of molecular data is essential to help tease out affinities to major clades within *Agaricus*, and indicate potential species boundaries (Geml et al. 2004; Kerrigan et al. 2006; Lebel and Syme 2012). Variation within the *A. wariatodes* clade may indicate cryptic species diversity, particularly when the very broad distribution of some of the other Australian arid region species is considered (Fig. 4). However, the powdery hymenophore (at maturity), emergent habit, and papery pileus (easily torn or lacerated) or rupturing veil of many of the arid zone sequestrate *Agaricus* species worldwide, is likely to allow wind dispersal of spores over long distances. Also, the habitats in which these species occur are often of low lying shrubs and grasses, small pockets of low growing, shrubby trees, and lots of open space. Broad geographic distributions for species are therefore possible.

With the current taxon sampling, the *Agaricus wariatodes/A. lamelliperditus/A. chartaceus* clade contains only sequestrate taxa. Clades with only sequestrate species are not common in lineages within the Agaricaceae (Gube 2009; Lebel and Syme 2012; Vellinga 2004). The single agaricoid collection (TL2383) made from the same location as *A. colpeteii*, belongs to a completely different section within *Agaricus* (Fig. 1). However, it seems likely that with the inclusion of further taxa from the same arid habitats and collection sites that agaricoid sister taxa will be found.

Acknowledgments Thank you to the Australian Biological Resources Study for funding some of the fieldwork (Bushblitz 2011, Neds Corner), and the RBG Melbourne for supporting molecular work. Staff of PERTH and MEL are acknowledged for providing access to specimens and aid in locating collection data. Reviewers of the manuscript are acknowledged for their input.

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