

## ***Volvopluteus earlei* and *Volvariella taylorii*: new fungi for Western Australia (Basidiomycota: Agaricales: Pluteaceae)**

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### Abstract

Bougher, N.L. & Barrett, M.D. *Volvopluteus earlei* and *Volvariella taylorii*: new fungi for Western Australia (Basidiomycota: Agaricales: Pluteaceae). *Nuytsia* 34: 187–201 (2023). Re-examination of vouchered specimens of pink-spored, volvate, agaricoid fungi held at the Western Australian Herbarium (PERTH) has revealed the first recorded occurrences in Western Australia of *Volvopluteus earlei* (Murrill) Vizzini, Contu & Justo and *Volvariella taylorii* (Berk. & Broome) Singer, and affirmed the regional prevalence of *Volvopluteus gloiocephalus* (DC.) Vizzini, Contu & Justo. The identities of the two *Volvopluteus* Vizzini, Contu & Justo taxa were confirmed with molecular data relative to extra-Australian samples validated in previous studies. *Volvopluteus earlei* had not been previously reported from Australia. Evidently *Volvopluteus earlei* and *Volvariella taylorii* are less common in Australia than the larger and more often encountered Common Rosegill, *Volvopluteus gloiocephalus*.

### Introduction

Australian members of the mushroom family Pluteaceae mostly have not been critically re-evaluated since the advent of the DNA revolution in the 1990s, nor in light of the family-wide phylogeny-based revisions of the family by Justo *et al.* (2011a, b) and others. Only a few Australian samples of *Melanoleuca* Pat. and *Pluteus* Fr. have been included in published studies (Bougher & Barrett 2020; Holec *et al.* 2018; Ševčíková *et al.* 2021). The present study considers three species in the genera *Volvopluteus* Vizzini, Contu & Justo and *Volvariella* Speg. with putative global distributions. These are mushroom-like fungi with a thick veil that envelops the egg-like immature stage and later forms a cup-like volva at the base of the stipe, free pinkish lamellae, and a salmon-coloured spore deposit. All such fungi were traditionally placed under *Volvariella* until molecular phylogenetic studies necessitated the segregation of some species into a newly elevated genus *Volvopluteus* (Justo *et al.* 2011a, b). *Volvariella* and *Volvopluteus* are well distinguished morphologically. Species of *Volvopluteus* have large spores and a gelatinised pileus whereas species of *Volvariella* mostly have shorter spores and a non-gelatinised pileus. Both genera are placed in the family Pluteaceae, in the order Agaricales (e.g. Kalichman *et al.* 2020), although some studies support the placement of *Volvariella* outside the Pluteoid clade (e.g. Justo *et al.* 2011b).

At the onset of the present study, pink-spored, volvate fungi collected in Western Australia (WA) held

as vouchered specimens at the Western Australian Herbarium (PERTH) were: *Volvariella bombycina* (Schaeff.) Singer (1 collection), *Volvariella* cf. *media* (Schumach.) Singer (3), *Volvariella* sp. (6), and *Volvopluteus gloiocephalus* (DC.) Vizzini, Contu & Justo (24) (Table 1). We re-examined these collections (all from southern WA) with a view to reviewing their identity. An additional collection of a volvarioid fungus (from northern WA) was also studied prior to its lodgement and incorporation at PERTH.

## Materials and methods

### Morphology

Morphological attributes of vouchered specimens of pink-spored, volvate fungi collected in WA and held at PERTH were studied (Table 1). The micromorphology of hand-sectioned dried material rehydrated in 3% KOH was examined by light microscopy using a Nikon 80i compound microscope. For spores, the abbreviation  $x$  refers to the mean length  $\times$  mean width,  $Q$  refers to the length/width ratio and  $Qx$  refers to the mean ratio measured from  $n$  basidiospores, observed from  $p$  collections. Colour notations in descriptions are from Kornerup and Wanscher (1978) by plate and row number.

### DNA isolation, PCR amplification and DNA sequencing

The Internal Transcribed Spacer (ITS) and Large Subunit (nLSU) of nuclear ribosomal DNA were sequenced for selected samples using the methodology described in Bougher and Barrett (2020). DNA sequences were compiled with relevant GenBank accessions included in previous studies (Table 2), and aligned using the MAFFT algorithm, implemented in Geneious Prime® v. 2021.2.2 (Kearse *et al.* 2012). Phylogenetic trees were reconstructed from the concatenated ITS+nLSU alignment using RAxML algorithm (Stamatakis 2014), using a rapid bootstrapping and search algorithm with 100 bootstrap replicates and a GTR+GAMMA nucleotide substitution model, as embedded in Geneious Prime v2021.2.2 (<https://www.geneious.com>; Kearse *et al.* 2012). The nucleotide substitution model was chosen using jModeltest 2 (Darriba *et al.* 2012).

**Table 1.** Morphological identities of pink-spored, volvate fungi collected in Western Australia and held prior to this study at the Western Australian Herbarium (PERTH). PERTH collections identified prior to this study as *Volvariella gloiocephalus* are not included in this table. \*Collections cited as *Volvariella speciosa* (Fr.) Singer by Hilton (1982). \*\*Voucher specimen recently lodged at PERTH.

| PERTH      | Coll. date | Location           | Prior identification                | Reviewed identification           |
|------------|------------|--------------------|-------------------------------------|-----------------------------------|
| 00750425   | 3/08/1964  | Kings Park, Perth  | <i>Volvariella</i> sp.*             | <i>Volvopluteus gloiocephalus</i> |
| 00752983   | 1/07/1967  | Condingup          | <i>Volvariella</i> sp.              | <i>Amanita</i> sp.                |
| 00759392   | 18/07/1971 | Subiaco, Perth     | <i>Volvariella bombycina</i>        | <i>Volvopluteus gloiocephalus</i> |
| 00761427   | Aug. 1971  | Esperance          | <i>Volvariella</i> sp.*             | <i>Volvopluteus gloiocephalus</i> |
| 00767212   | 18/06/1973 | Esperance          | <i>Volvariella</i> sp.              | <i>Volvopluteus gloiocephalus</i> |
| 07572689   | 22/02/1998 | City Beach, Perth  | <i>Volvariella</i> ? sp.            | <i>Volvopluteus earlei</i>        |
| 07680279   | 2/03/2005  | Scarborough, Perth | <i>Volvariella</i> cf. <i>media</i> | <i>Volvopluteus earlei</i>        |
| 07680287   | 6/03/2005  | Scarborough, Perth | <i>Volvariella</i> cf. <i>media</i> | <i>Volvopluteus earlei</i>        |
| 07680295   | 8/03/2005  | Scarborough, Perth | <i>Volvariella</i> cf. <i>media</i> | <i>Volvopluteus earlei</i>        |
| 09519432** | 14/02/2011 | Broome             | <i>Volvariella</i> sp.              | <i>Volvopluteus earlei</i>        |
| 07552440   | 15/06/1995 | Kellerberrin       | <i>Volvariella</i> sp.              | <i>Volvariella taylorii</i>       |

**Table 2.** Accessions used in the molecular phylogeny. nLSU sequences contiguous with ITS are indicated in brackets.

| Taxon   | Voucher origin                   | Voucher/ strain               | ITS GenBank | nLSU GenBank | Reference                                 |
|---|----------------------------------|-------------------------------|-------------|--------------|---|
| <i>Volvopluteus earlei</i>                                      | Pakistan                         | LAH35715                      | MW362280    |              | Kahn <i>et al.</i> (2022)                 |
| <i>Volvopluteus earlei</i>                                      | Turkey: Bursa                    | OKA-TR656                     | MW033396    | MW029827     | Kaygusuz <i>et al.</i> (2021)             |
| <i>Volvopluteus earlei</i>                                      | Turkey: Izmir                    | OKA-TR655                     | MW033395    | MW029826     | Kaygusuz <i>et al.</i> (2021)             |
| <i>Volvopluteus earlei</i>                                      | Turkey: Mersin                   | OKA-TR654                     | MW033394    | MW029825     | Kaygusuz <i>et al.</i> (2021)             |
| <i>Volvopluteus earlei</i>                                      | Democratic Republic of the Congo | Mamet7                        | HM562205    |              | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i> (as <i>Volvariella cookei</i> )      | Italy: Sardinia                  | TO AV133                      | HM246496    | HM246480     | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i>                                      | Spain                            | MA22816                       | HM562204    | HM562253     | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i>                                      | Italy: Lucca                     | ALV17724                      | MK204987    |              | Giannoni <i>et al.</i> (2018)             |
| <i>Volvopluteus earlei</i> (as <i>Volvariella media</i> )       | Italy: Sardinia                  | TO HG2001                     | HM246498    |              | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i>                                      | Italy: Sardinia                  | TO AV134                      | HM246497    | HM246482     | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i>                                      | Italy: Lucca                     | ALV17726                      | MK204989    |              | Giannoni <i>et al.</i> (2018)             |
| <i>Volvopluteus earlei</i>                                      | Italy: Lucca                     | ALV17725                      | MK204988    |              | Giannoni <i>et al.</i> (2018)             |
| <i>Volvopluteus earlei</i> (as <i>Volvariella acystidiata</i> ) | Italy: Sardinia                  | TO HG1973                     | HM246499    | HM246481     | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus earlei</i>                                      | China                            | XJ1379                        | MW811446    |              | J. Xu, unpublished                        |
| <i>Volvopluteus earlei</i>                                      | China                            | XJ1378                        | MW811445    |              | J. Xu, unpublished                        |
| <i>Volvopluteus earlei</i>                                      | Japan: Niigata                   | Kasuya B3518, TNS:F-70427     | MH021868    |              | Kasuya <i>et al.</i> (2018)               |
| <i>Volvopluteus earlei</i>                                      | Japan: Chiba                     | Kasuya B3561, TNS:F-70428     | MH021869    |              | Kasuya <i>et al.</i> (2018)               |
| <i>Volvopluteus</i> sp.   | China: Xinjiang                  | JPL-2019, GDGM73195           | MK944281    | MN056508     | Chuan-Hua, L. <i>et al.</i> , unpublished |
| <i>Volvopluteus</i> sp.   | China: Xinjiang                  | JPL-2019, GDGM74751           | MK944280    | MN056509     | Chuan-Hua, L. <i>et al.</i> , unpublished |
| <i>Volvopluteus asiaticus</i>                                   | Japan                            | TNSF15191; from TYPE material | HM562206    |              | Justo <i>et al.</i> (2011a)               |
| <i>Volvopluteus asiaticus</i>                                   | China                            | HD2022                        | OP599925    |              | Y. Yang, unpublished                      |

| <b>Taxon</b>                      | <b>Voucher origin</b>     | <b>Voucher/ strain</b>                             | <b>ITS GenBank</b>                   | <b>nLSU GenBank</b> | <b>Reference</b>                                  |
|-----------------------------------|---------------------------|--|--------------------------------------|---------------------|---|
| <i>Volvopluteus asiaticus</i>     | China                     | HD2021   | OP597804                             |                     | Y. Yang, unpublished                              |
| <i>Volvopluteus michiganensis</i> | Russia                    | LE 312006  | MK729542                             |                     | Krom <i>et al.</i> (2019)                         |
| <i>Volvopluteus michiganensis</i> | Russia: Krasnoyarsky Kray | LE311991   | MK049912                             |                     | Krom <i>et al.</i> (2019)                         |
| <i>Volvopluteus michiganensis</i> | China                     | HMJAU-CR106  | MW242669                             |                     | Rao <i>et al.</i> (2021)                          |
| <i>Volvopluteus michiganensis</i> | USA: Michigan             | A.H. Smith 32-590 (MICH 11761); from TYPE material | HM562195 (duplicate record NR_11987) |                     | Justo <i>et al.</i> (2011a)                       |
| <i>Volvopluteus michiganensis</i> | Canada: AB, Helena Lakes  | UBC F-32158  | MF954699                             |                     | Berbee, M.L. <i>et al.</i> , unpublished          |
| <i>Volvopluteus</i> sp.           | Iran: Tehran              | Ghobad-Nejhad 4321                                 | MT535739                             | MT554328            | Ghobad-Nejhad <i>et al.</i> (2020)                |
| <i>Volvopluteus michiganensis</i> | Russia                    | LE311989   | MK729541                             |                     | Krom <i>et al.</i> (2019)                         |
| <i>Volvopluteus gloiocephalus</i> | Denmark                   | DM1042   | MT644914                             |                     | Leerhoei, F. & Langkjaer, E.M., unpublished       |
| <i>Volvopluteus gloiocephalus</i> | Russia                    | 5173   | MH930216                             |                     | Vaishlya, O.B. <i>et al.</i> , unpublished        |
| <i>Volvopluteus gloiocephalus</i> | Egypt                     | EGDA-Volv1   | MW916608                             |                     | El-Gharabawy, H.M. & El-Fallal, A.A., unpublished |
| <i>Volvopluteus gloiocephalus</i> | Turkey: Hatay             | OKA-TR666  | MW033406                             | MW029837            | Kaygusuz <i>et al.</i> (2021)                     |
| <i>Volvopluteus gloiocephalus</i> | Turkey: Aydin             | OKA-TR665  | MW033405                             | MW029836            | Kaygusuz <i>et al.</i> (2021)                     |
| <i>Volvopluteus gloiocephalus</i> | Turkey: Bursa             | OKA-TR664  | MW033404                             | MW029835            | Kaygusuz <i>et al.</i> (2021)                     |
| <i>Volvopluteus gloiocephalus</i> | Turkey: Afyonkarahisar    | OKA-TR663  | MW033403                             | MW029834            | Kaygusuz <i>et al.</i> (2021)                     |
| <i>Volvopluteus gloiocephalus</i> | New Zealand               | PDD_103792   | MN738645                             | MN738593            | Cooper, J.A., unpublished                         |
| <i>Volvopluteus gloiocephalus</i> | Algeria: M'Sila forest    | CM042  | KP826741                             |                     | Benazza, M. <i>et al.</i> , unpublished           |
| <i>Volvopluteus gloiocephalus</i> | USA: Arizona              | Mushroom Observer 420468                           | MW633065                             |                     | Clements, T.A., unpublished                       |
| <i>Volvopluteus gloiocephalus</i> | Spain                     | LOU18247   | HM562209                             |                     | Justo <i>et al.</i> (2011a)                       |

| <b>Taxon</b>   | <b>Voucher origin</b>         | <b>Voucher/ strain</b>                       | <b>ITS GenBank</b> | <b>nLSU GenBank</b> | <b>Reference</b>                           |
|--|-------------------------------|--|--------------------|---------------------|--|
| <i>Volvopluteus gloiocephalus</i>                              | Spain                         | LOU13710                                     | HM562208           |                     | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i>                              | Portugal                      | LOU18619                                     | HM562207           |                     | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i>                              | Spain                         | AJ239  | HM562202           | HM562252            | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i> (as <i>V. gloiocephala</i> ) | Italy: Sardinia               | TO AV135                                     | HM246490           | HM246476            | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i>                              | USA: California               | PBM2272                                      | HM562203           |                     | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i>                              | New Zealand                   | JAC10832                                     | MN738634           | MN738571            | Cooper, J.A., unpublished                  |
| <i>Volvopluteus gloiocephalus</i>                              | Canada: BC, New Westminster   | UBC F-32227                                  | MF954700           |                     | Berbee, M.L. <i>et al.</i> , unpublished   |
| <i>Volvopluteus gloiocephalus</i>                              | USA: California               | MO479337                                     | OP470065           |                     | Schwartz, J. <i>et al.</i> , unpublished   |
| <i>Volvariella gloiocephalus</i> (as <i>V. gloiocephala</i> )  | Italy: Sardinia               | TO AV136                                     | HM246495           | HM246478            | Justo <i>et al.</i> (2011a)                |
| <i>Volvopluteus gloiocephalus</i> (as <i>Vp. sp.</i> )         | Iran: Zanjan                  | 12   | MK785230           |                     | Ammarellou, A. & Alvarado, P., unpublished |
| <i>Volvariella gloiocephala</i>                                | USA: California               | AFTOL-ID 890, PBM2272                        | DQ494701           | AY745710            | Matheny <i>et al.</i> (2006)               |
| <i>Volvopluteus gloiocephalus</i>                              | Australia: WA, Wembley Downs  | <i>N.L. Bougher</i> NLB 1579, PERTH 08945233 | <b>OP809559</b>    | <b>(OP809559)</b>   | <b>This study</b>                          |
| <i>Volvopluteus earlei</i>                                     | Australia: WA, near Broome    | <i>M.D. Barrett</i> F86/11, PERTH 09519432   | <b>OP809558</b>    | <b>OP808462</b>     | <b>This study</b>                          |
| <i>Volvopluteus earlei</i>                                     | Australia: NT, near Katherine | <i>M.D. Barrett</i> F14/10, DNA D0290038     | <b>OP809557</b>    | <b>OP808461</b>     | <b>This study</b>                          |
| <i>Pluteus romellii</i>  | Australia: WA, Kings Park     | <i>N.L. Bougher</i> NLB 1554, PERTH 08944962 | MT537114           | (MT537114)          | Bougher & Barrett (2020)                   |
| <i>Pluteus terricola</i>                                       | New Zealand                   | JAC14632, PDD:106511                         | MN738653           | MN738580            | Cooper, J.A., unpublished                  |
| <i>Pluteus multiformis</i>                                     | Spain                         | X472   | MN250225           |                     | Ševčíková <i>et al.</i> (2021)             |

## Results

### PERTH collections labelled as *Volvopluteus gloiocephalus*

All 24 PERTH collections labelled as *Volvopluteus gloiocephalus* were confirmed as correctly identified.

### PERTH collections labelled other than as *Volvopluteus gloiocephalus*

Four of the five collections from the 1960s and 1970s that had been referred to *Volvariella* were found to match *Volvopluteus gloiocephalus* (Table 1) by virtue of their large spores, cystidia forms, and a highly gelatinised pileipellis. One of these collections (PERTH 00759392, from Subiaco in 1971) had been previously identified as *V. bombycina*, a lignicolous (wood-inhabiting) species with a non-gelatinised pileipellis. This material, which consists of a single, immature specimen that lacks spores, shows no evidence of a lignicolous habit and has a broad ixocutis. Both of these features are indicative of *Volvopluteus gloiocephalus*. The fifth collection (PERTH 00752983, from Condingup in 1967) has smooth, hyaline spores, and velar tissue with some inflated cells, features typical of *Amanita* Pers.

A collection from City Beach in 1998 labelled as *Volvariella*? sp. and three collections from Scarborough in 2005 previously identified as *Volvariella* cf. *media* are referable to *Volvopluteus earlei* (Murrill) Vizzini, Contu & Justo (Table 1) on account of their small white basidiomata, large spores, variably-shaped cystidia, and gelatinised pileipellis. A collection from Broome in 2011 was also morphologically confirmed as *V. earlei* and subsequently lodged (PERTH 09519432).

A collection from near Kellerberrin in 1995 (PERTH 07552440) previously identified as *Volvariella* sp. morphologically conforms with *Volvariella taylorii* (Berk. & Broome) Singer based on features such as its small, grey, radially fibrillose pileus, grey volva, ellipsoid-ovoid to oblong spores, cystidia forms, and non-gelatinised pileipellis.

### Molecular identities of *Volvopluteus* species

The RAxML analysis of concatenated ribosomal sequences from ITS and nLSU confirmed the identities of the two Australian *Volvopluteus* species (Figure 1). The two tropical Australian collections (from Broome, WA and the Northern Territory) were strongly supported as belonging to a clade containing all samples of *V. earlei* from other continents. The single southern Australian accession of *V. gloiocephalus* was strongly supported as belonging to a clade containing all other *V. gloiocephalus* from other continents. Both species are thus confirmed to be present in Australia, and belong to the same ribosomal lineages as present on multiple continents. The morphological species concepts of both species of *Volvopluteus* applied here are thus confirmed consistent with their global distributions.

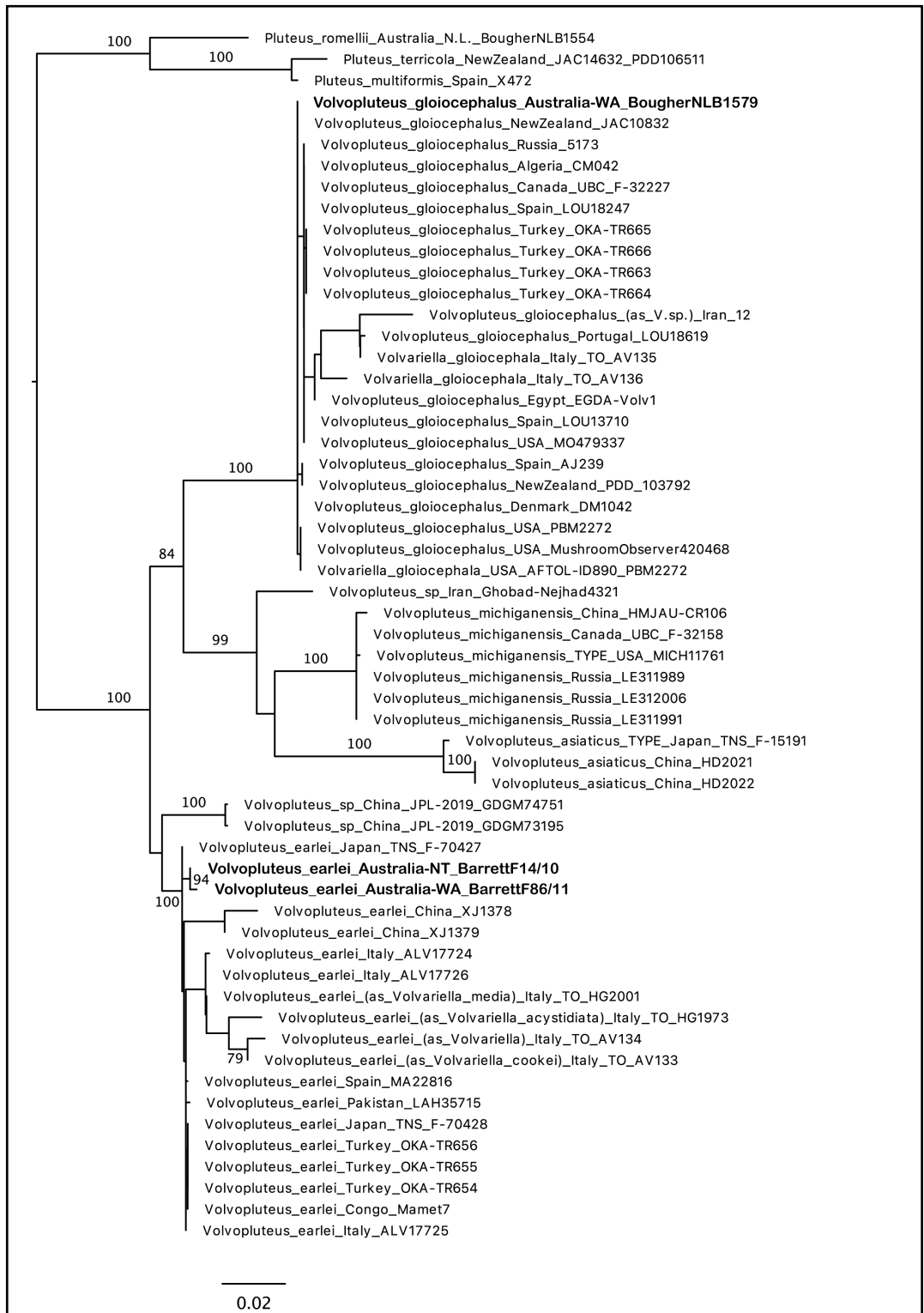


Figure 1. Phylogenetic placement of Australian *Volvopluteus* samples (indicated in bold) amongst globally representative samples. RAxML tree with support values from 100 bootstrap replicates.

## Taxonomy

**Volvopluteus gloiocephalus** (DC.) Vizzini, Contu & Justo, in Justo, A., Vizzini, A., Minnis, A.M., Menolli, N. Jr., Capelari, M., Rodriguez, O., Malysheva, E., Contu, M., Ghignone, S. & Hibbett, D.S., *Fungal Biology* 115(1): 15 (2011).

≡ *Agaricus gloiocephalus* DC., *Fl. franç.* 5/6: 52 (1815).

[*Volvariella speciosa* auct. non (Fr.) Singer: Bougher & Syme (1998: 226).]

*Pileus* 30–110 mm wide, ovoid at unexpanded stage, conico-convex when young, soon developing an umbo and expanding to flat, uniformly greyish beige (2A2–3A2) except darker brownish (5C5) at the umbo, thick-viscid or more often thick-glutinous (not very sticky), drying shiny-smooth; margin incurved and entire, then plane and eroded when old, entire and translucent-striate close to margin. *Lamellae* free, up to 16 mm deep, crowded, white in button stage, soon beige (2A2), then pink and finally brownish pink (6D6); edge minutely fringed, very eroded with age; 1 or 2 tiers of lamellulae. *Stipe* 35–120 × 5–22 mm, cylindrical or tapering upwards, solid or partially hollow, dry, white tinged yellowish or brownish (4B4), with long striate, silky fibrils, and sometimes with minute white floccules near base of stem. *Volva* usually buried in soil, saccate, white, with a free limb up to 1 mm thick, no veil remnants on cap. *Context* white, sometimes dulling with age and watery when old. *Odour* not distinctive. *Taste* none, indistinct. (Figure 2A)

*Basidiospores* pink-brown (near 7D7) in deposit, pale yellow-brown in water or KOH, (10.5) 12–17.5 × 7.5–10 μm,  $x = 14.9 \times 8.7 \mu$ ,  $Q = 1.55\text{--}1.93$ ,  $Q_x = 1.72$  ( $n = 50$ ,  $p = 1$ ), not dextrinoid or amyloid, ellipsoid, smooth, thick-walled (up to 1 μm), germ pore absent. *Basidia* 42–56 × 12–16 μm, cylindro-clavate to clavate, 4-spored, sterigmata up to 5 μm long. *Cheilocystidia* 32–70 × 10–25 μm, mainly fusoid-ventricose, some more swollen or clavate, with or without a narrow apical extension (2–3 × up to 18 μm). *Pleurocystidia* 45–60 × 20–40 μm, clavate, ventricose, some with short rostrum up to 5 μm long, scattered. *Pileipellis* with a broad outer layer of dissolved, hyaline hyphae embedded in a gelatinous matrix; hyphae in lower pellis with intact walls 6–10 μm broad, smooth, hyaline. *Clamp connections* absent. (Figure 3A–C)

*Selected specimens* [28 PERTH collections examined]. WESTERN AUSTRALIA: Colin Road, Wembley Downs, emerging in leafy mulched garden bed, 21 Aug. 2017, *N.L. Bougher* NLB 1579 (PERTH08945233; ITS/LSU sequence GenBank OP809559); Woodlake Retirement Home, Woodlake Retreat, Kingsley, Perth, in garden bed with woodchips and rich mulch, 15 Aug. 2009, *N.L. Bougher & M. Bougher* BOU 00571 (PERTH 08473927); Wellington Street, Albany, in composted hay, 19 May 1993, *K. Syme* KS 642/93 (PERTH 05506301).

*Notes.* This is a common and conspicuous species in gardens, parks and farms throughout southern Australia. It is readily recognised by its large basidiomata with a slimy to sticky pileus, dull pink mature lamellae, and tall white stipe lacking a ring and with a basal cup-like volva. In WA, as elsewhere, its basidiomata may occur singly or extensively in large numbers, and are usually found in disturbed areas rich in organic matter, e.g. mulched garden beds, woodchip beds, composted hay, or areas of mowed or unkempt grass. The ITS/LSU sequence of a typical collection from the Perth region (PERTH 08945233) has 100% similarity with sequences of *V. gloiocephalus* on GenBank (Figure 1).

The above description has been adapted from that presented in Bougher and Syme (1998, as *Volvariella speciosa* (Fr.) Singer), which was based on a Western Australian collection (PERTH 05506301).



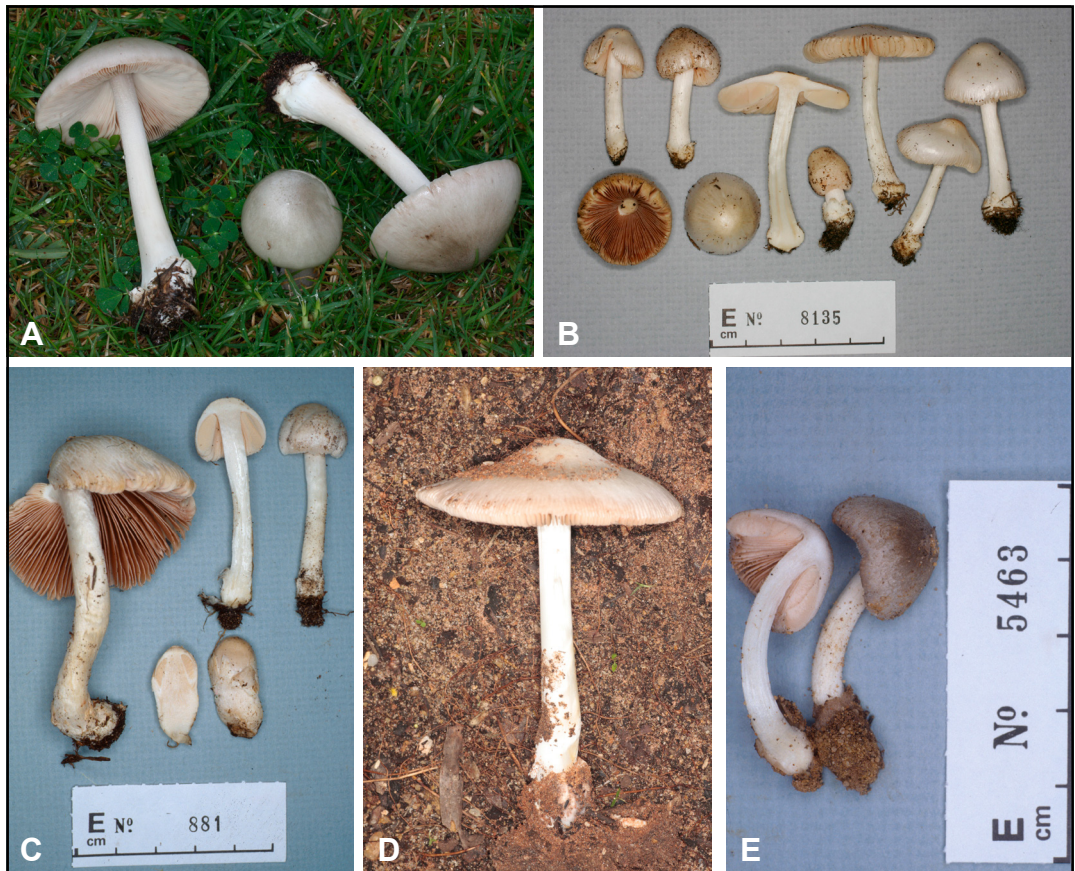


Figure 2. Basidiomata. A – *Volvopluteus gloiocephalus* (PERTH 08473927); B – *Volvopluteus earlei* (PERTH 07680287); C – *Volvopluteus earlei* (PERTH 07572689); D – *Volvopluteus earlei* (PERTH 09519432); E – *Volvariella taylorii* (PERTH 07552440). Scale bars = 2 cm.

***Volvopluteus earlei*** (Murrill) Vizzini, Contu & Justo, in Justo, A., Vizzini, A., Minnis, A.M., Menolli, N. Jr., Capelari, M., Rodriguez, O., Malysheva, E., Contu, M., Ghignone, S. & Hibbett, D.S., *Fungal Biology* 115(1): 15 (2011).

≡ *Volvariopsis earlei* Murrill, *Mycologia* 3(6): 282 (1911).

*Pileus* 20–50 (80) mm wide, initially hemispherical or conic, expanding to bluntly conic-campanulate with or without a low umbo, finally plane or undulate, white to cream to almost snow-white, dulling with age then becoming slightly darker or pale yellowish biscuit in centre (near to but paler than 4A–B2); margin decurved then plane, becoming thin-fleshed at the extreme margin often exposing the lamellae, may become deeply split; pileus surface viscid-sticky in wet conditions, slippery as becoming drier, shiny and minutely radially appressed silky fibrillose in dry conditions. *Lamellae* free, densely crowded to subcrowded, ventricose, white when very young then cream becoming pale apricot-pink (4A2), then darker pink (6A2–6A3), then dark dull brown-pink (7C5, 7C6) at maturity, up to 5 mm deep; edge smooth, concolorous and entire, lamellulae in one series. *Stipe* 40–70 (100) × 5–10 mm, white, cylindric or tapering slightly towards apex, base rounded to clavate; surface dry, shiny, smooth to the eye but longitudinally appressed silky fibrillose (under lens); annulus absent. *Volva* 21–32 × 15–25 mm, sheathing, white, membranous, saccate with a narrow free rim, often less

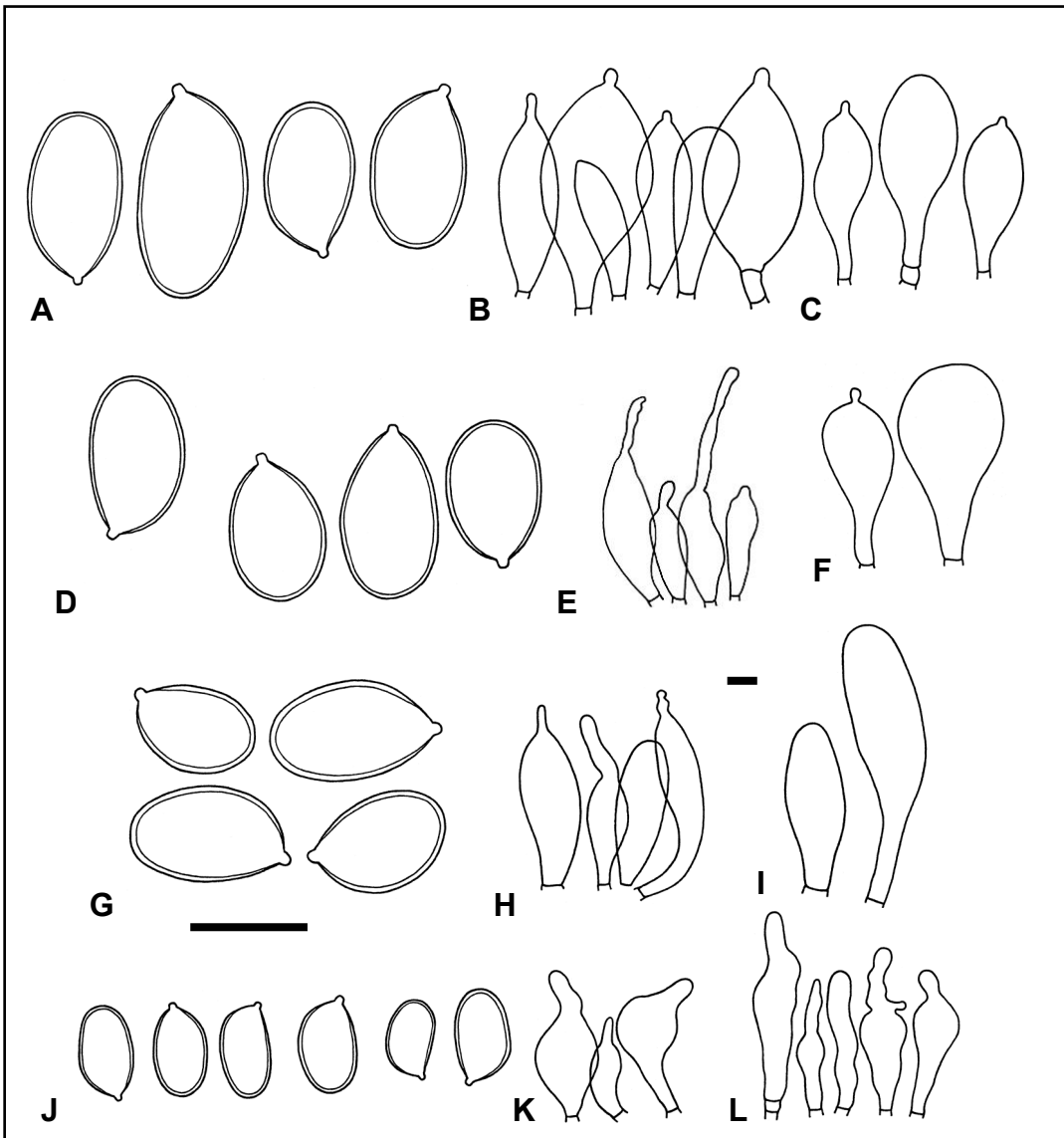


Figure 3. Micromorphology. A–C, *Volvopluteus gloiocephalus* (PERTH 08473927): A – spores; B – cheilocystidia; C – pleurocystidia. D–F, *Volvopluteus earlei* (PERTH 07572689): D – spores; E – cheilocystidia; F – pleurocystidia. G–I, *Volvopluteus earlei* (PERTH 09519432): G – spores; H – cheilocystidia; I – pleurocystidia. J–L, *Volvariella taylorii* (PERTH 07552440): J – spores; K – cheilocystidia; L – pleurocystidia. Scale bars = 10  $\mu\text{m}$  (longer bar spores, shorter bar cystidia).

conspicuous in mature specimens as the rim may become appressed to the stipe with age. *Context* white in pileus and stipe, stipe solid eventually becoming densely longitudinally fibrous/stuffed or developing a thin hollow centre with age, not staining or bruising. *Odour* fabaceous (fresh beans), or cooked fresh sweet corn, or not noticeable. (Figure 2B–D)

*Basidiospores* dark orangish pink or brownish pink or deep salmon pink-brown (near 6-7C–D7–7E7) in deposit, pale yellowish in water or KOH, PERTH 07680287:  $12\text{--}15.5 \times 7.3\text{--}9.5 \mu\text{m}$ ,  $x = 13.5 \times 8.4 \mu\text{m}$ ,  $Q = 1.47\text{--}1.92$ ,  $Qx = 1.63$  ( $n = 50$ ,  $p = 2$ ), PERTH 09519432:  $10.5\text{--}14.7 \times 6.8\text{--}8.9 \mu\text{m}$ ,  $x = 11.8 \times 7.5 \mu\text{m}$ ,  $Q = 1.46\text{--}1.71$ ,  $Qx = 1.58$  ( $n = 27$ ,  $p = 1$ ), ellipsoid, smooth, thick-walled, apiculus

prominent. *Basidia* 35–51 × 10–14 µm, clavate to cylindro-clavate, hyaline, mostly 4-sterigmate, rarely 2-sterigmate. *Cheilocystidia* 30–75 × 11–24 µm, variable in shape – clavate, ellipsoid, fusoid, ventricose or lageniform, with or without apical extension up to 12 µm long (rarely up to 45 µm long), smooth, thin-walled, hyaline, often abundant and crowded. *Pleurocystidia* 31–85 × 15–38 µm, clavate to cylindro-clavate or vesiculose, sometimes with an apical extension, smooth, thin-walled, sometimes thickened in upper part, hyaline, inconsistently present (scattered singly, rare or absent). *Pileipellis* a narrow ixocutis of gelatinised hyphal remnants, overlying a cutis of non-gelatinised hyaline hyphae 3–7 µm wide. *Caulocystidia* none observed. *Clamp connections* absent. (Figure 3D–I)

*Specimens examined.* WESTERN AUSTRALIA: North end of Cable Beach, Broome, in pile of dirt and mulch used for garden landscaping, 14 Feb. 2011, *M.D. Barrett* F86/11 (PERTH 09519432; ITS sequence GenBank OP809558; LSU sequence GenBank OP808462); Yaltara Park, City Beach, Perth, on lawn/grass, 22 Feb. 1998, *N.L. Bougher & M.E. Bougher s.n.* (PERTH 07572689); Drabble-Cobb Park, Scarborough, Perth, scattered on and embedded in grass, 2 Mar. 2005, *N.L. Bougher, M. Bougher & R. Bougher* E 8134 (PERTH 07680279); *loc. id.*, 6 Mar. 2005, *N.L. Bougher, M. Bougher & R. Bougher* E 8135 (PERTH 07680287); *loc. id.*, 8 Mar. 2005, *N.L. Bougher, M. Bougher & R. Bougher* E 8136 (PERTH 07680295).

*Notes.* The specimens described here from WA conform well to *V. earlei* (e.g. as described by Justo & Castro 2010) by virtue of the following morphological features: small, white basidiomata with pilei usually < 50 mm wide, large spores (> 10 µm long), variably-shaped hymenial cystidia sometimes with an apical mucro or longer finger-like appendage, and a gelatinised pileipellis. All the WA collections examined have at least some lageniform cheilocystidia, and some rostrate pleurocystidia occur in the southern WA collections. *Volvopluteus asiaticus* Justo & Minnis has predominantly lageniform cheilocystidia and mostly rostrate pleurocystidia (Justo *et al.* 2011b). However, that species differs from *V. earlei* by having larger basidiomata (pileus > 5 cm diam.). The collection from Broome (F86/11; PERTH 09519432) conforms to sequences (ITS 99% similarity) of *V. earlei* on GenBank and belongs to the same phylogenetic lineage as representatives of that species from other continents (Figure 1).

***Volvariella taylorii*** (Berk. & Broome) Singer [as ‘*taylori*’], *Lilloa* 22: 401 (1951) [1949].

≡ *Agaricus taylorii* Berk. & Broome, *Ann. Mag. Nat. Hist.*, Ser. 2 13: 398 (1854).

*Pileus* 16 mm wide, convex, margin decurved; surface dry and radially appressed silky fibrillose, dark grey (near 5D2) at centre grading paler grey towards margin (near 5B3). *Lamellae* free, crowded, not ventricose, 2.5 mm deep; pale pink (near 5A3–6A2), edge smooth, concolorous and entire. *Stipe* 30 × 4 mm; cylindrical, base slightly swollen; surface dry, minutely longitudinally appressed silky fibrillose (under lens); entirely white. *Volva* grey (near 5D2), membranous, saccate with a narrow free ragged rim. *Context* white in pileus and stipe, stipe solid, not staining or bruising. *Odour* not distinctive. (Figure 2E)

*Basidiospores* reddish brown (near 6D6) in deposit, pale yellowish in water or KOH, 7–8.3 × 4.2–5 µm,  $x = 7.6 \times 4.6 \mu\text{m}$ ,  $Q = 1.52\text{--}1.84$ ,  $Qx = 1.66$  ( $n = 27$ ,  $p = 1$ ), ellipsoid or ovoid (broadest towards base) to oblong, smooth, thick-walled, apiculus prominent. *Basidia* 21–36 × 8–10 µm, slender clavate, hyaline, 4-sterigmate. *Cheilocystidia* 31–53 × 9–20 µm, ventricose with blunt mucro < 5 µm long or an apical appendage up to 15 × 9 µm, smooth, thin-walled, hyaline or glassy, scattered singly or in small groups. *Pleurocystidia* 40–70 × 10–20 µm, clavate to sublageniform, mostly with a finger-like or sometimes branched apical appendage up to 10–20 × 5–7 µm, smooth, thin-walled, hyaline or glassy,

scattered singly. *Pileipellis* a non-gelatinised cutis of hyphae 6–40 µm wide giving rise to emerging mostly undifferentiated or sometimes tapering hyphal-like end-cells, larger inner hyphae often have constricted septa and some end-cells swollen up to 60 µm wide. *Caulocystidia* none observed. *Clamp connections* absent. (Figure 3J–L)

*Specimens examined.* WESTERN AUSTRALIA: Hilltop site between Site 17A & B, Higginson Rd, 16 km along Bencubbin-Kellerberrin Rd, N of Kellerberrin, single specimen on soil under *Allocasuarina campestris* and *Acacia acuminata*, 15 June 1995, B. Dunstan & I.C. Tommerup s.n. (PERTH 07552440).

*Notes.* This collection from WA's Avon Wheatbelt bioregion conforms with *V. taylorii* as described from other regions of the world (e.g. Pradeep *et al.* 1998 – India; Seok *et al.* 2002 – South Korea). Conforming features include its small, grey, radially fibrillose pileus, grey volva, terrestrial habit, ellipsoid-ovoid to oblong spores 7–8.3 × 4.2–5 µm, hymenial cystidia with an apical appendage (sometimes branched), and non-gelatinised pileipellis with emerging undifferentiated end cells. The identification of this collection as *V. taylorii*, herein based on morphology, remains to be confirmed with molecular data.

### Discussion

It is unsurprising that most of the PERTH collections of pink-spored, volvate fungi are of *Volvopluteus gloiocephalus* as this is a very common and conspicuous species aptly known locally as the 'Common Rosegill'. It is also unsurprising that one of the collections originally identified as *Volvariella* is a species of *Amanita*. Many *Amanita* species have a conspicuous, cup-like volva and a few have pink gills, e.g. *A. carneiphylla* O.K. Mill. In addition to having a universal veil, most *Amanita* species produce a white or cream spore deposit. Therefore they are readily distinguishable from species of *Volvariella* and *Volvopluteus*, which lack a universal veil and have a pinkish spore deposit. One PERTH collection of *Volvopluteus gloiocephalus* had been originally identified as *Volvariella bombycina*, a large, lignicolous species predominant in the northern hemisphere and unconfirmed in Australia. It is unlikely to occur in WA unless introduced.

Some morphological variation is evident among the Western Australian collections of *Volvopluteus earlei*. For example, the mean spore size of the southern collections (e.g. 13.5 × 8.4 µm for PERTH 07680287) is greater than that of a northern collection (PERTH 09519432: 11.8 × 7.5 µm). Also, among and within the Western Australian collections of *V. earlei* the hymenial cystidia vary in shape and in the presence and length of an apical extension. These features, and the presence and abundance of pleurocystidia, were observed in some cases to be highly variable among and within individual specimens. Similar variability in spores and cystidia has been reported for *V. earlei* outside Australia. For example, mean spore size has been quoted as 13.53 × 8.38 µm for Italian collections (Giannoni *et al.* 2018), and 12.5 × 7 µm for Japanese collections (Kasuya *et al.* 2018). Collections lacking any pleurocystidia have been reported from Spain (Justo & Castro 2010), and some from Italy have non-rostrate cheilocystidia or lack hymenial cystidia (e.g. *V. earlei* f. *acystidiatus* (N.C. Pathak) Vizzini & Contu: Justo *et al.* 2011b).

*Volvopluteus earlei* had not been previously recorded in Australia; however, it has been documented from many other parts of the world (see Justo & Castro 2010) including humid areas such as the Congo, Cuba, Mexico, and southern USA, and parts of Mediterranean Europe such as Italy and Spain. It is also known from some temperate areas such as Japan (Kasuya *et al.* 2018). Its widespread occurrence predominantly in the northern hemisphere and lack of previous records in Australia suggest that it

may have been introduced recently into southern WA from elsewhere. In the Perth region it is only currently known from two, artificially-watered, grassed urban parks. Its basidiomata are often quite conspicuous at these sites and appear to emerge after an episode of warm, humid weather early in the year (February, March), much earlier than the main, local fungal fruiting season (usually June–August). The status of tropical Australian specimens of *V. earlei* is less certain. The collection from near Broome in WA was found in a pile of dirt and mulch used for landscaping. Similarly, a collection from near Katherine in the Northern Territory (DNA D0290038, see Figure 1) was fruiting on a mix of dirt and native grasses from a grader spill beside a highway. Both are anthropogenically disturbed sites, and spores could have been introduced to the sites. An additional, un-vouchered observation (M. Barrett, Dec. 1995) in grader spill near the northern edge of Beverley Springs Station (now Charnley River Station), in a very remote area of the Kimberley Region, was very unlikely to have been transported to the site, although the habitat was certainly disturbed. Introduction of genotypes with horticulture is likely, but *V. earlei* may also have been indigenous in Northern Australia prior to European arrival. Either way, the species evidently is encouraged by anthropogenic disturbance. Even though there have been few records of it to date, *V. earlei* may be widespread or at least becoming widespread in Australia. The first regional records of *V. earlei* also have been made in recent years in some other parts of the world. Justo and Castro (2010) reported on the first record in Spain and suggested that it may be a tropical species alien to Europe. Spanish and Italian collections of *V. earlei* were collected during late spring and summer in artificially irrigated gardens.

PERTH 07552440 is the first record of *Volvariella taylorii* in WA. Another record, recently listed by Bougher and Barrett (2020) as '*Volvopluteus* sp. (small fruit bodies with non-viscid grey pileus)' from sandy soil at Bold Park, Perth may also be *V. taylorii* but its identity cannot be confirmed as no specimen was vouchered (photographs show a small grey pileus and grey volva). Leonard (2015) in referring to a Queensland record of *V. taylorii* (as *V. pusilla* var. *taylori*) wrote: 'there are references in the Australian literature but do not appear to be any collections in Australian Herbaria'; however, there is at least one preserved Australian collection identified as *V. taylorii*, originating from Blackbutt, Queensland in 1989 (BRI AQ0646466). Based on the limited records so far, it seems that *V. taylorii* may be expected to occur over wide parts of Australia but perhaps it has been sparsely reported because it is small and/or it may fruit less frequently or abundantly than some other pink-spored, volvate fungi such as *Volvopluteus gloiocephalus*.

Four species of pink-spored, volvate fungi are currently known to occur in WA (Table 3) although there are likely to be others. For example, in northern WA, at least three unidentified *Volvariella* species are known (M.D. Barrett, unpublished data). A census of the larger fungi of WA (Hilton 1982) includes three species of *Volvariella*: *V. bombycina*, *V. speciosa* and *V. cynopotamia* (Berk.) Singer. The inclusion of *V. bombycina* cannot be assessed because the associated voucher (UWA 1219) cannot be located. The census cites UWA 1566, 1065 and 819 as collections of *V. speciosa* (stating that specimens so far collected intergrade with *V. speciosa* var. *gloiocephala* (DC.) Singer). Two of these vouchers (UWA 1566, PERTH 00761427; UWA 819, PERTH 00750425) represent *Volvopluteus gloiocephalus*; the third (UWA 1065) cannot be located. *Volvariella cynopotamia* was described by Berkeley (1881: 389, as *Agaricus cynopotamia* Berk.) who examined scanty materials of a single collection from 'Swan River' sent to him at The Royal Botanic Gardens, Kew in 1849 by James Drummond. That collection has pink, subglobose spores  $5.5\text{--}8 \times 4.7\text{--}6.5 \mu\text{m}$  (Pegler 1965). No subsequent collections attributable to this species have been reported from WA, but there is one from Queensland in 2010 (BRI AQ0793965). The epithet is more accurately spelt *V. cynopotamia*, as its etymology is presumably based on 'Swan River'.

**Table 3.** Some comparative distinguishing features of the four species of pink-spored, volvate agaricoid fungi currently known to occur in Western Australia.

| Character              | <i>Volvopluteus gloiocephalus</i>        | <i>Volvopluteus earlei</i>            | <i>Volvariella taylorii</i>           | <i>Volvariella cygnopotamia</i> |
|------------------------|--|---------------------------------------|---------------------------------------|---------------------------------|
| Pileus width           | mostly > 50 mm                           | < 50 mm                               | < 50 mm                               | < 50 mm                         |
| Pileus surface         | viscid, slowly dry                       | viscid, rapidly dry                   | dry                                   | dry                             |
| Volva                  | white                                    | white                                 | grey                                  | white? *                        |
| Spores                 | > 10 µm; ellipsoid                       | > 10 µm; ellipsoid                    | < 10 µm; ellipsoid-ovoid-oblong       | < 10 µm; subglobose             |
| Pileipellis            | gelatinised ixocutis                     | gelatinised ixocutis                  | non-gelatinised cutis                 | non-gelatinised cutis           |
| WA distribution        | southern                                 | southern & northern                   | southern                              | southern                        |
| WA frequency & habitat | common: gardens, woodchips, mulch, lawns | uncommon: grass, garden woodchip heap | uncommon: one location to date - soil | rare: ground                    |

\*The protologue for *Volvariella cygnopotamia* by Berkeley (1881, as '*Agaricus cycnopotamia*') quotes the following: 'Volva composed of intricate threads, with a central dark patch which appears to contain spiral vessels, but the material is so scanty that it is impossible to speak with certainty.'

### Acknowledgements

Curators at the Western Australian and Northern Territory herbaria are thanked for their support accessing and processing specimens used in this study. Graham Brown and David Liddle, and Kings Park Science are thanked for logistical and material support during collection expeditions during which the tropical specimens were obtained. We thank Hana Ševčíková (referee) for her constructive comments on this paper.

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