Fungi available to and consumed by translocated Gilbert's potoroos: Preliminary assessments at three translocation sites

Neale Bougher, Tony Friend and Louisa Bell



Gilbert's Potoroo (*Potorous gilbertii*) is Australia's most critically endangered mammal.



Potoroos recently have been translocated onto Bald Island off W. Australia's south coast.



At their only known natural refuge - Two Peoples Bay Nature Reserve - the potoroos feed almost exclusively on native truffle fungi all year round. Are similar fungal food resources available elsewhere and accessed by translocated potoroos?



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Abstract

Fungi, mainly truffle-like fungi, are predominant in the diet of the 'critically endangered' Gilbert's Potoroo (*Potorous gilbertii*) at Two Peoples Bay Nature Reserve - the animal's only known natural refuge. The diversity of fungi available to potoroos and the capacity of individuals to access fungi in new areas are likely to significantly influence the breeding success and survival of potoroos. This report outlines a preliminary assessment of fungi available to and consumed by translocated Gilbert's potoroos. It is based on surveys undertaken during 2007 at three translocation sites in the south coast region of east of Albany, Western Australia. At Bald Island where potoroos had been first translocated in 2005, 40 collections of fungi including 16 species of truffles were made at the same times and locations as scats obtained from four individual potoroos. A total of 27 spore types were observed in the scats indicating that the diversity of fungi consumed by translocated potoroos resident on Bald Island for at least 1 or 2 years is comparable to that of the natural population of potoroos at Two Peoples Bay Nature Reserve. At two other sites on the mainland designated for future translocations, 9 species of truffle fruit bodies were collected, indicating that suitable fungal food resources would be available to potoroos translocated there in the future.

Background

The critically endangered mammal - Gilbert's Potoroo (Potorous gilbertii) was rediscovered in 1994 on the south coast of Western Australia east of Albany at Two Peoples Bay Nature Reserve (Sinclair et al., 1995). Previously potoroos had been recorded widely in the south coast region, but the species was presumed to be extinct since last recorded in the 1870's. Less than 40 individuals exist in the single population discovered in 1994. Gilbert's potoroos now are only known from four areas on the Mount Gardner peninsula and one in the Bishops Gully area of Two Peoples Bay Nature Reserve (Friend, 2003). Hence the species remains extremely vulnerable to extinction. In response, the Department of Environment and Conservation (DEC) has established a breeding and translocation program for Gilbert's Potoroo (Courtenay and Friend, 2004). The program aims to establish potoroos in areas where they may have occurred in the past and where predators are naturally absent or excluded by fencing. Significant aspects of potoroo biology determining translocation success and breeding success are likely to include diet and interaction with other organisms, particularly fungi. Fungi, predominantly truffle-like fungi, are a significant food source for many small mammals including potoroos in Australia (Claridge et al., 1996; Claridge et al., 2007). More than 2,000 species of native truffles may occur in Australia and more than 95% of known species are endemic to Australia (Bougher & Lebel 2001). At Two Peoples Bay Nature Reserve, truffles are the major component of the Gilbert's potoroo diet (Bougher, 1998; Nguyen, 2000; Nguyen et al., 2005). The fungi interact with major potoroo habitat plants such as *Gastrolobium* and *Eucalyptus* via their mycorrhizal associations. The survival of Gilbert's potoroo, the fungi they eat, and the plants of their habitat are likely to be tightly interdependent. Fungi are likely to be a prerequisite for successful translocation and sustained survival and breeding of translocated potoroos.

The first translocation of Gilbert's potoroos was undertaken in 2005, to Bald Island a mainly granite island of 809 hectares off the south coast of Western Australia. Since their release, a series of translocated potoroos have survived, reared offspring, and consumed truffles on the island (Friend *et al.* 2005; Friend 2006). DEC is currently assessing and preparing other areas on the mainland for translocations of potoroos.

The current report outlines a preliminary assessment of fungi available to and consumed by translocated potoroos. It is based on surveys undertaken during 2007 at three translocation sites – Bald Island, Ryedene, and Waychinicup National Park (see Map). At Bald Island scats from briefly trapped potoroos were obtained at the same time as fungi fruit bodies from vegetation near the traps. This enabled direct comparisons to be undertaken between fungi fruit bodies and spore types in the scats of trapped potoroos. At Ryedene and Waychinicup, potoroos had not yet been translocated, and so it was possible to assess the fungi present before the release of potoroos, with the intention to track the consumption of fungi by potoroos translocated there later. The 14 hectare site at Ryedene was already fenced, and potoroos were to be introduced there several months after the current study. The larger translocation site at the Waychinicup National Park was not yet fenced, and introduction of potoroos there has been scheduled for 2009 or later.

Locations of study sites

Bald = Bald Island; Way = Waychinicup National Park; Ry = Ryedene.



Methods

Three potoroo translocation sites were studied during 2007: Bald Island 13-16 August, Ryedene 20 & 22 June, and Waychinicup NP 21 June (see Map). Field sampling for fungi fruit bodies was not structured spatially except at Bald Island, where sampling was focused on vegetation within 100m of traps set out to capture potoroos in order to compare fungi fruit bodies sampled with spore types in the scats of trapped potoroos. Hand-held rakes were used to find truffle fungi by raking in the leaf litter and in the soil to a maximum depth of about 10cm. Epigeous fungi fruiting above ground were also sampled. Morphological attributes of the fresh fruit bodies were recorded, and then specimens were forcibly air-dried at 45° C. Permanent vouchers of the fungi are lodged at the Western Australian Herbarium (PERTH).

At Bald Island, small cages were set up during daytime to lure and capture potoroos foraging at night. Cages were revisited early the following morning and captured potoroos were released immediately after a brief examination. Scats deposited in the cages were placed into vials of alcohol for later examination.

Preserved fruit bodies and scats later were examined in detail using a compound microscope using a 100x objective. Other relevant reference specimens of fungi at the Western Australian Herbarium were also examined to help determine the identity of the fungi.

Results

1. Bald Island

a. Fruit bodies

Forty collections of fungi were made at Bald Island during this study. Sixteen species were represented among 28 collections of truffle fungi from Bald Island. A further 12 species of epigeous fungi were collected. Many more species of epigeous fungi were observed but not recorded or collected. Of the truffles, all were Basidiomycetes except for one – the ascomycete *Elaphomyces*. None were Glomeromycetes or Zygomycetes (the other groups of fungi that have at least some species forming truffle-like fruit bodies). 11 of the 16 species of the truffle fungi remain identified only to genus pending further collections or more detailed comparative analyses. Identifications, descriptive details, and images of the specimens are provided below (Table 1, Appendix 1).

b. Scats

Five scat samples were obtained from four individual potoroos. Female 98 was trapped twice in successive days (Table 2). Based on morphological attributes, a total of 27 spore types were observed in the scats examined (Table 3). Twenty three of the spore types could be at least tentatively assigned to fungal genera. Of those, 7 could be matched to particular fungi species. Four spore types could not be assigned to any fungal taxon. 11 spore types matched spores of fruit bodies collected in the field, while 16 did not match any fungus collected as fruit bodies at Bald Island during this study (Tables 1, 3).

The occurrence of spore types across the various scats was variable. Seven spore types were observed in all scats examined – *Gymnomyces* sp. nov. 2 (spore type 1); *Zelleromyces daucinus* (4); *Hysterangium* cf. *affine* (11); *Hysterangium* sp. cystidioid (12); *Austrogautieria* sp. (13); *Protoglossum* sp. (large spores) (15); and *Pogiesperma* sp (18). All of those fungi species were also observed as fruit bodies except for *Austrogautieria* and *Protoglossum*. A total of 8 spore types were common to all 4 individual potoroos trapped (female potoroo 98 had spore type 3 *Gymnomyces/Cystangium* in vial 1 but not vial 2).

Nine spore types were observed only once – an unknown ascomycete (spore type 5), two *Protoglossum* species (7, 20), a *Glomus* sp. (14), *Quadrispora tubercularis* (17), two *Gymnomyces* species (22, 27), an unknown (24), and *Hysterangium inflatum* (25). None of those fungi were observed as fruit bodies.

c. Comparison between fruit bodies and scats

Spores of 11 of the 16 species of truffle fungi collected as fruit bodies at Bald Island were observed in potoroo scats (Tables 1, 3). No spores of any of the epigeous fungi collected or any other epigeous fungi were observed in scats. Spores of 5 of the truffle fungi collected as fruit bodies were observed in all scats examined – *Gymnomyces* sp. nov. 2 (spore type 1), *Hysterangium* cf. *affine* (11), *Hysterangium* sp. cystidioid (12), *Pogeisperma* sp. (18), and *Zelleromyces daucinus* (4). Unexpectedly, spores of *Mesophellia brevispora* were not observed in scats, even though there was evidence in the field of its fruit bodies recently consumed by animals.

In some cases, fruit bodies of a particular species collected nearby to where a particular potoroo was trapped did occur as spores in the scats of that individual (Table 2). For example *Zelleromyces daucinus* and *Pogiesperma* sp. were found near to where female potoroo 98 was trapped, and were also present as spores in the scats of that potoroo. However this was not the case for many of the fungi. For example 6 species of truffle fungi were collected near to where female 118 was trapped, but only two of the fungi (both *Hysterangium* species) were observed in its scats. In some cases

spores of a fungus collected as fruit bodies were observed in scats from an individual other than from the nearest trapped potoroo, e.g. *Cystangium sessile* was found fruiting near female 118 but its spores were found in three other different individuals – females 98 and 100 (see Tables 2, 3).

2. <u>Ryedene fruit bodies</u>

Thirty three collections of fungi were made at Ryedene during this study. Five species were represented among 10 collections of truffle fungi from Ryedene. A further 23 species of epigeous fungi were collected. Many more species of epigeous fungi were observed but not recorded. Of the truffles, all were Basidiomycetes except for one - the ascomycete *Hydnoplicata convoluta*. None were Glomeromycetes or Zygomycetes (the other groups of fungi that have at least some species forming truffle-like fruit bodies). 4 of the 5 species of the truffle fungi remain identified only to genus pending further collections or more detailed comparative analyses. Identifications, descriptive details, and images of the specimens are provided below (Table 3, Appendix 3).

3. <u>Waychinicup NP fruit bodies</u>

Sixteen collections of fungi were made at Waychinicup NP during this study. Four species were represented among 5 collections of truffle fungi from Waychinicup NP. A further 11 species of epigeous fungi were collected. Epigeous fungi were not in abundance in the areas surveyed. Of the truffles, all were Basidiomycetes. None were Glomeromycetes or Zygomycetes (the other groups of fungi that have at least some species forming truffle-like fruit bodies). 3 of the 4 species of the truffle fungi remain identified only to genus pending further collections or more detailed comparative analyses. Identifications, descriptive details, and images of the specimens are provided below (Table 4, Appendix 4).

<u>**Table 1</u></u>: Summary of fungi species obtained as fruit bodies from Bald Island.** (see Appendix 1 for details and images of the truffle collections). Crosses indicate number of scat samples (out of 5) in which each species was present.</u>

Species	Herbarium Code	Hypogeous (Truffle)	Confirmed in Potoroo Scats	
		Or Epigeous	(see also TABLE 3 & Appendix 2)	
Chondrogaster sp.	BOUGHER 367; 390	Truffle	+++	
Cystangium seminudum	BOUGHER 385; 386	Truffle	++	
Descomyces sp. nov.	BOUGHER 372; 373	Truffle	-	
Elaphomyces sp. nov.	BOUGHER 387	Truffle	+++	
Gymnomyces boranupensis	BOUGHER 375; 376	Truffle	+++	
Gymnomyces sp. nov. 1	BOUGHER 369	Truffle	-	
Gymnomyces sp. nov. 2	BOUGHER 370	Truffle	+++++	
<i>Hysterangium</i> cf. <i>affine</i> = sp. white messy	BOUGHER 363; 378; 389	Truffle	+++++	
Hysterangium sp. cystidioid	BOUGHER 379; 380; 392	Truffle	+++++	
Hysterangium sp. pink, thick peridium	BOUGHER 371; 377	Truffle	-	
Mesophellia brevispora	BOUGHER 368; 381	Truffle	-	
Pogisperma sp.	BOUGHER 362; 384	Truffle	+++++	
Protoglossum sp.	BOUGHER 365; 366	Truffle	++	
Pseudohysterangium sp.	BOUGHER 374	Truffle	+++	
Trappea sp.	BOUGHER 388	Truffle	-	
Zelleromyces daucinus	BOUGHER 382	Truffle	+++++	
Amanita xanthocephala	BOUGHER 391	Epigeous	-	
Anthracophyllum archeri	BOUGHER 399	Epigeous	-	
Camarophyllus sp.	BOUGHER 401	Epigeous	-	
Entoloma sp.	BOUGHER 397	Epigeous	-	
Geastrum sp.	BOUGHER 395	Epigeous	-	
Geastrum sp.	BOUGHER 398	Epigeous	-	
Inocybe sp.	BOUGHER 364	Epigeous	-	
Laccaria sp.	BOUGHER393	Epigeous	-	
Lycoperdon cf. perlatum	BOUGHER 396	Epigeous	-	
Pycnoporus coccineus	BOUGHER 394	Epigeous	-	
Rhodophyllus/ Entoloma sp.	BOUGHER 400	Epigeous	-	
Russula clelandii	BOUGHER 383	Epigeous	-	

<u>Table 2</u>: Individual potoroos on Bald Island trapped 14-15 August 2007, and from which scats were examined.

* indicates spores of the fungus were observed in scats obtained from the particular potoroo on same day (see Table 3).

Scat Sample	Date	Trap Line	Potoroo	Date Translocated	Truffle fruit bodies collected nearby	Notes
					v	
1	14/8/07	Trap line F100-N6 trap 4	Female 98	9/8/05	BOU363 Hysterangium cf. affine *	Potoroo with a one month old pouch young
2	15/8/07	Trap line F100-N6 trap 5	Female 98	9/8/05	BOU379 Hysterangium sp. cystidioid BOU380 Hysterangium sp. cystidioid BOU382 Zelleromyces daucinus * BOU383 Russula clelandii BOU384 Pogiesperma sp. *	Same potoroo as previous
3	15/8/07	Trap line F100-N4 trap 4	Female 100	7/12/05	BOU362 Pogiesperma sp. * BOU381 Mesophellia brevispora	Most southerly site sampled. Large <i>Gastrolobium</i> and abundant eucalypts occuring along exposed granite strips near the edge of the island
4	15/8/07	Trap line F100-N4 trap 5	Male 114	28/8/06	BOU362 Pogiesperma sp. * BOU381 Mesophellia brevispora	Same site as previous
5	15/8/07	Summit Trap line trap 1	Female 118	Island-born	BOU385 Cystangium seminudum BOU386 Cystangium seminudum BOU387 Elaphomyces sp. nov. BOU388 Trappea sp. BOU389 Hysterangium cf. affine * BOU390 Chondrogaster sp. BOU391 Amanita xanthocephala BOU392 Hysterangium sp. cystidioid *	Steep track up the side of the Bald Island's central summit.

<u>Table 3</u>: Types and identities of fungi spores in scats of Gilbert's Potoroo translocated to Bald Island collected during the current study from from animals trapped 14-15 August 2007. See Appendix 2 for images of the spore types. * indicates fruit bodies collected at Bald Island during this study (see Table 1).

	SPORE TYPE	IDENTITY	COLOUR & AMYLOIDY (IN MELZERS)	SIZE	MATCHING FRUIT BODIES	S C A T 1	S C A T 2	S C A T 3	S C A T 4	S C A T 5
1.	Faintly amyloid, subglobose, smaller than others, isolated short warts, prominent hilar appendix up to 2 μm long	Gymnomyces sp. nov. 2 *	amyloid	7.3 - 8.1 x 7.1 - 5.7 μm	BOU370	+	+	+	+	+
2.	Globose, broken reticulum	Gymnomyces boranupensis *	amyloid	9.9 µm diam.	BOU375, 376	+	+	-	-	+
3.	Globose, large isolated pegs, isolated - not densely spaced	Gymnomyces/Cystangium	amyloid	6.1 - 8.4 μm	None	+	-	+	+	+
4.	Globose, very strong reticulum with tall ridges	Zelleromyces daucinus *	amyloid	7.7 – 8.8 µm	BOU382	+	+	+	+	+
5.	Large broad fusoid spore with truncate projecting appendage at both ends.	Unknown Ascomycete	pale	?	None	+	-	-	-	-
6.	Fusoid spores with inflating bubbly persporium	Chondrogaster sp. *	pale brown	9.4 – 12 x 4.2 - 5.7 μm	BOU367, 390	+	-	+	+	-
7.	Cortinarioid: ellipsoid, assymetrical in side view, finely ornamented, smaller than others	Protoglossum or Cortinarius	bright brown	7.9 – 8.8 x 5.2 – 5.5 μm	None	+	-	-	-	-
8.	Globose, densely spinose	Cystangium seminudum *	amyloid	7.2 – 9.3 μm	BOU385, 386	+	-	+	-	-
9.	Dextrinoid fusoid smooth, thick-walled . Some assymetrical.	Pseudohysterangium /Hysterogaster *	dextrinoid (brown)	11 – 13.1 x 4.4 - 5 μm	BOU374	+	+	+	-	-
10.	Globose densely warted/spinose	Elaphomyces sp. nov. *	dark blackish olive	12.2 – 14.9 µm	BOU387	+	-	+	+	-
11.	Fusoid, hyaline without perispore, apiclulus not conspicuous	Hysterangium cf. affine *	hyaline	8.3 x 3.6 μm	BOU363, 378, 389	+	+	+	+	+
12.	Fusoid, wrinkling persporium, hyaline	Hysterangium sp. cystidioid *	hyaline	14.2 x 4.4 µm	BOU379, 380, 392	+	+	+	+	+
13.	Fusoid, longitudinally ridged	Austrogautieria sp.	golden yellowish or greenish	12.5 – 14.9 x 5.5 – 7.4 µm	none	+	+	+	+	+
14.	Very large globose spores and attachment hyphae seen	Glomus sp	bright yellow	?	none	+	-	-	-	-
15.	Large, very dark brown broad ovoid to turbinate, cortinarioid, verrucose	<i>Protoglossum</i> sp. (like a large- spored P. atratum)	dark brown	15 – 17.1 x 12.7 – 13.9 μm	none	+	+	+	+	+
16.	Ellipsoid, verrucose or broken reticulum, non-inflating perisporium	Protoglossum sp. *	bright brown	12.7 – 14.1 x 7.7 – 9.6 µm	BOU365, 366	+	+	-	-	-
17.	Asymmetric in side view cortinarioid spores adhering in clusters of four. (only one cluster seen, spores not mature)	Quadrispora tubercularis	bright brown	?	None	+	-	-	-	-
18.	Small cylindric, inconspicuous hilar appendix. The most abundant spore type in scat samples.	Pogiesperma sp. *	hyaline	$5.4 - 6.9 \ x \ 2.9 - 3.6 \ \mu m$	BOU362, 384	+	+	+	+	+
19.	Subglobose non-amyloid, pale yellowish in Melzers, coarsely reticulate, tall ridges	Unknown (possible <i>Octavianina</i> sp.)	pale yellowish	?	none	+	+	+	-	-
20.	Slender ellipsoid, flattened on adaxial side, coarsely verrucose with broad rounded warts including on the non-mucronate apex, apiculus claw-like.	Protoglossum sp.	dark orange-brown	13.9 – 17.4 x 8.3 – 10.2 μm	none	-	-	+	-	-
21.	Perfectly broad ellipsoid, surface rugulose-wrinkled, sometimes partial braod reticulum, overlying perisporium or gelatinous material?, thin- walled, no hilar appendix visible.	Hydnoplicata convoluta	hyaline to pale yellowish	11.2 – 13.3 x 8.0 – 9.6 μm	none	-	-	+	+	+
22.	Globose, large, strong reticulum, ridges up to 1.5 μm tall (one only seen)	Gymnomyces sp.?	amyloid very dark purple	18.9 µm diam.	none	-	I	-	+	-
23.	Globose, coarsely warted, pegs up to $1.2 \mu m$ tall	Unknown	bright golden	6.6 - 16.2 μm diam.	none	+	-	-	+	-
24.	Globose, large spores strongly reticulate, pegs up to 1.6 μm tall	Unknown	dull brown		none	-	I	-	-	+
25.	Fusoid, with inflating wing-like truncate perisporium	Hysterangium inflatum	hyaline	$10.5 - 11.8 \text{ x } 3.3 - 6.7 \mu\text{m}$	none	-	-	-	-	+
26.	Globose, strong but broken reticulum, tall ridges up to 2.5 μm (1 only)	Gymnomyces sp.?	Amyloid deep purple	8.7 μm diam.	none	-	-	-	+	+
27.	Globose, large spores, strong but broken reticulum, tall ridges up to 2 μm Only one spore seen.	Gymnomyces sp.?	Amyloid deep purple	12.2 – 14.7 μm diam.	none	-	-	-	-	+

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<u>**Table 3:**</u> Summary of fungi obtained as fruit bodies from Ryedene 20-22 June 2007 (see Appendix 3 for images and more details of the truffle collections)

Identit	y of Fungus	Herbarium Code	Hypogeous (Truffle) Or
			Truffle
Gymnomyces	<i>sp</i> .	BOUGHER 326	Truffle
Hydnoplicata	convoluta	BOUGHER 317; 333	Truffle
Hysterangium	<i>sp</i> .	BOUGHER 291; 325; 329	Truffle
Pogiesperma	sp. A	BOUGHER 324; 327; 328	Truffle
Pogisperma	sp. B	BOUGHER 292	Truffle
Aleurina	ferruginea	BOUGHER 00316	Epigeous
Amanita	xanthocephala	BOUGHER 00286	Epigeous
Amanita	umbrinella	BOUGHER 00314	Epigeous
Austropaxillus	muelleri	BOUGHER 00320	Epigeous
Cortinarius	cystidiocatenata	BOUGHER 00280; 285	Epigeous
Cortinarius	sp.	BOUGHER 00321	Epigeous
Cortinarius	<i>sp</i> .	BOUGHER 00322	Epigeous
Cymatoderma	cf. elegans	BOUGHER 00323	Epigeous
Galerina	<i>sp</i> .	BOUGHER 00315	Epigeous
Inocybe	<i>sp</i> .	BOUGHER 00284	Epigeous
Inocybe	sp.	BOUGHER 00288	Epigeous
Inocybe	sp.	BOUGHER 00319	Epigeous
Inocybe	sp.	BOUGHER 00331	Epigeous
Laccaria	sp.	BOUGHER 00283	Epigeous
Licehnomphalina	umbellifera	BOUGHER 00289	Epigeous
Mycena	sp.	BOUGHER 00281	Epigeous
Ramaria	<i>sp</i> .	BOUGHER 00282	Epigeous
Ramaria	capitata var. ochraceosalmonicolor	BOUGHER 00287	Epigeous
Ramaria	versatilis	BOUGHER 00290	Epigeous
Russula	sp.	BOUGHER 00313	Epigeous
Russula	neerimea	BOUGHER 00330	Epigeous
Russula	clelandi	BOUGHER 00332	Epigeous
Sistotrema	<i>sp</i> .	BOUGHER 00318	Epigeous

<u>Table 4</u>: Summary of fungi obtained as fruit bodies from Waychinicup National Park 21 June 2007 (see Appendix 4 for images and more details of the truffle collections)

Identity	of Fungus	Herbarium Code	Hypogeous (Truffle) Or Enigeous
Hysterangium	sp. A	BOUGHER 00304	Truffle
Hysterangium	sp. B	BOUGHER 00305	Truffle
Pseudohysterangium	sp.	BOUGHER 00306	Truffle
Zelleromyces	daucinus	BOUGHER 00299; 303	Truffle
Clavulina	cinerea	BOUGHER 00302	Epigeous
Craterellus	sp.	BOUGHER 00296	Epigeous
Fistulina	mollis	BOUGHER 00301	Epigeous
Galerina	sp.	BOUGHER 00294	Epigeous
Inocybe	sp.	BOUGHER 00293	Epigeous
Lactarius	eucalypti	BOUGHER 00297	Epigeous
Mycena	carmeliana	BOUGHER 00295	Epigeous
Mycena	sp.	BOUGHER 00307	Epigeous
Omphalotus	nidiformis	BOUGHER 00300	Epigeous
Russula	neerimea	BOUGHER 00298	Epigeous
Tubaria	serrulata	BOUGHER 00308	Epigeous

Discussion

The findings of this study demonstrate that a diverse range of truffle and other fungi occur in sites targeted so far for translocation of Gilbert's potoroo. At the two sites studied on the mainland designated for future translocations, 9 species of truffle fruit bodies were collected, indicating that suitable fungal food resources would be available to potoroos translocated there in the future. A total of 27 spore types were observed in scats examined from Bald Island and this indicates that potoroos established on the island for at least one or two years since translocation are consuming many species of truffle fungi. This includes individual potoroos that are successfully breeding, e.g. an individual with pouch young that has been resident on the island for two years (Female 98), and a potoroo that was born on the island (Female 118).

The diversity of spore types in scats recorded in the current study indicates that the diversity of fungi consumed by translocated potoroos on Bald Island is comparable to that of the original population at Two Peoples Bay Nature Reserve. About 25 spore types were found in scats collected during the period December 1994 to March 1998 in the first study of fungi consumed by natural populations of Gilbert's potoroos at Two People's Bay (Bougher, 1998). Nguyen *et al.* (2005) reported up to 44 fungal spore types in scats collected June – September 2000 (Nguyen *et al.*, 2005). Because all studies of spores in scats of Gilbert's Potoroo so far have had a different intensity and duration of sampling effort, these figures are not directly comparable, but at least indicate that the translocated potoroos are likely to be accessing a similar diversity of fungi to that consumed by natural populations.

The occurrence of spore types across the various scats was variable. The results of scat analysis show that different individuals were consuming different sets of fungi on Bald Island at the same point in time. Only 8 of the 27 spore types were common to scats of all four individual potoroos examined on the island. Some of these differences may be a reflection of the limited sampling undertaken in this study, and it is not known how differences in foraging patterns and preferences between individuals may vary over time.

In many cases, the spores observed in scats of a particular potoroo were not matched to any of the fruit bodies collected during this study. The limited sampling of this study undoubtedly did not reveal all the truffle fungi fruiting in a given area. The potoroos were finding fungi species that we did not find. Conversely, in many cases fruit bodies of a particular truffle species collected nearby to where a particular potoroo was trapped did not corresponded to any of the spores in the scats of that individual. This suggests that individual potoroos do not necessarily access all the truffle fungi species fruiting in the animal's area at any one point in time. For example, spores of *Mesophellia* were not observed in scats even though discarded fruit body shells and intact fruit bodies were observed in the field near a trapped animal (Male 114). Perhaps at the time of sampling *Mesophellia* fruit bodies were being consumed by animals other than this particular potoroo, either by other potoroos or other species such as quokkas.

In similarity with other studies on mycophagy by potoroos, many of the truffle fungi recorded in the current study are endemic to Australia but known to be widespread throughout the continent (Bougher & Lebel 2001). *Mesophellia, Castoreum and Hysterangium* have often been found to be the most abundant truffle fungi consumed by Gilbert's potoroo at Two Peoples Bay Nature Reserve (Bougher, 1998, Nguyen *et al.*, 2005) and other potoroids (Claridge *et al.*, 1993; Claridge *et al.*, 2007). In the current study, this was also the case for *Hysterangium* but not so for *Mesophellia* or *Castoreum*. Different truffle species fruit at different times of the year (Claridge *et al.*, 1993) and perhaps the latter two fungi become more abundant at other times of the year. In the current study, *Pogiesperma* species were among the most abundant spore type in scats. Bald Island is long-unburnt

and has developed a thick litter layer in many parts of the island. Fleshy truffles such as *Hysterangium* and *Pogiesperma* may be particularly favoured in such litter conditions as they are probably more prone to desiccation than tougher species such as *Mesophellia* and *Castoreum*.

In difference to other studies so far (e.g. Bougher, 1998; Nguyen *et al.*, 2005) no spores of epigeous fungi in scats of Gilbert's Potoroo were observed in this study, with the unconfirmed exception of spore type 7 (Table 3, Appendix 2). This may be attributable to the time of sampling on Bald Island which was later in the year than the main fruiting time for epigeous fungi in the area.

Far more truffle fungi and other fungi species are likely to occur at the translocation sites than the species obtained in this preliminary survey. Some of the most widespread and common species of truffles known in the region were not found fruiting during this study. For example although the spores of *Austrogautieria* were observed in 3 out of 5 scat samples at Bald Island no fruit bodies were found there. The limitations of sampling in this current study were compounded by being undertaken towards or beyond the end of the likely main fruiting season for fungi in the region, particularly the August sampling at Bald Island.

As in previous studies (e.g. Bougher, 1998; Nguyen *et al.*, 2005) the identification of the fungi consumed by potoroos remains problematic. e.g. in the current study only 5 of the truffle fungi were assigned species names, and 11 of the 16 species remain identified only to genus pending further collections or more detailed comparative analyses. Some of the fungi may be un-named and undescribed and new species to Science. Further collecting of fungi and scats over multiple seasons and years, and matching of spores in scats to fungi collected as fruit bodies is required to improve identification. The current study, in which many (11 out of 16) species of truffle fungi collected as fruit bodies were present in the scats, shows that collecting fruit bodies near trapped animals at the same time is a useful strategy for potentially helping to identify the fungi in the future.

This study has shown that a diversity of fungi are being accessed by translocated potoroos resident on Bald Island for at least 1 or 2 years. Some other questions and issues in relation to translocation of potoroos include:

- How quickly do translocated potoroos begin to access diverse truffles after they are released?
- The seasonality of fungi in the potoroo diet. At Two Peoples Bay Nature Reserve, fungi are consumed by potoroos all year round (Bougher, 1998; Nguyen, 2000; Nguyen *et al.*, 2005), but it is not known if other sites with different vegetation types have suitable fungi available all year round.
- The sustainability of translocated potoroo populations: Could successful potoroo translocations foster populations of animals that over-exploit the fungi?
- What are the mycorrhizal host plants of truffle fungi in translocation sites? All truffle fungi, including the truffle fungi obtained in this survey, are presumed to be ectomycorrhizal, i.e. they have and depend upon mutually beneficial partnerships with plants. In order to gain a better understanding of the ecology of potoroo habitats data about the identity, abundance, and health of putative ectomycorrhizal host plants at translocation sites needs to be assessed in relation to fungi survey data.
- Multiple surveys over multiple years need to be undertaken to adequately assess the diversity and abundance of fungi at translocation sites.
- A better capacity to identify fungi and to track patterns in consumption of fungi by potoroos would be facilitated by creation and coordination of an illustrated database. This would enable correlation of data such as for individual animals, spores in scats, fungi fruit bodies, and mycorrhizal plant associations.

Acknowledgements

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Appendices

Appendix 1: Truffle fungi species obtained as fruit bodies from Bald Island during the current study. Also see summary Table 1.

Details

Chondrogaster sp. BOUGHER 367; 390

Macro Morphology

Characteristic Features: (i) elastic mycelium inbetween the fruit bodies, (ii) outer peridium of elastic hyphae; middle peridium 0.3-0.4mm thick of mycorrhizal roots and entangled hyphae; inner peridium thin whitish giving rise to some thin short veins; (iii) gleba solid spore brown packages with intervening whitish sterile material more or less radially arranged; (iv) abundant rhizomorphs at base.

Fruitbodies: 6-10mm diam.; globose; base attached to abundant elastic and sand-encased rhizomorphic whitish mycelium. No sterile base or columella evident. Peridium: Outer layer very thin and variable in integrity, of whitish parallel, elastic hyphae that form the chewing gum as seen in the photo of the split specimen. Middle layer thicker (up to 0.3mm wide) a dense entanglement of similar hyphae as in the outer peridium and dull brownish mycorrhizal roots. Inner layer (or just the trama?) very thin, white compact, giving rise to some white veins that penetrate into and submerge into the gelatinised gleba. Macroscopically the peridium appears to the eye as smooth, whitish, matted with much adhering soil. Gleba: solid, gelatinised but not liquified; brown (near 6F6) with thin, short (0.1mm) whitish intrusions emminating from the peridium and in a radial pattern (not perfectly) inbetween sinuous and irregular-shaped spore packages. Some veins appear to become brown and gelatinised, so as to become semi-translucent and harder to discern. The locules are variable in colour- many are pale yellow-brown esp. near centre, while others are darker brown and more gelatinised esp. near outer gleba. There is no evidence of gleba becoming powdery in these specimens. The veins are solid, become gelatinised, and are not of gummy separable hyphae.

BOUGHER 00390 cm

Images

Micro Morphology

Spores ellipsoid-fusoid, with inflating bubble-like inflations, pale brown in KOH. Hilar appendix inconspicuous.

Comments

There is no columella, but the tramal plates are quite thick and therefore conspicuous, often in a radial arrangement. This species has gummy mycelium in the peridium and embedded and adhering to soil and roots. The peridium dries dull brown (compared for example with BOU379 etc which dry pale kahki green).

The brown gleba suggests this is not Hysterangium. However aggregation of multiple basidiomes into a tightly bound common matrix as typical of Chondrogaster was not seen.

Differs from Gummivena in that: (i) spore mass doesn't become powdery; (ii) not strictly 3-layered peridium (inner layer maybe just trama); (iii) veins in gleba are not of gummy tissue.

Also differs from Gummivena by:

(a) lacks a columella

- (b) has a white peridium versus smooth brown peridium in *Gummivena*.
- (c) lacks a thick brown woody rhizomorph but has many whitish rhizomorphs and elastic mycelium binding soil.
- Different spores broad fusoid with bubbly inflating perisporium versus (d) Gummivena basidiospores: 10-12 x (4-) 5-5.5 µm, narrowly ellipsoid to subfusiform, the length-width ratios 2-2.4 (-2.7), smooth in youth, at maturity ornamented with minute (< 0.5 μ m tall or broad) lines and dots or an occasional swelling up to 0.5 μ m tall and 2 μ m broad, the sterigmal attachment 1.5 (-2) µm broad, prominent or inconspicuous.

Cystangium seminudum BOUGHER 385; 386

Macro Morphology

Characteristic Features: (i) pure white to becoming ivory peridium; (ii) white loculate gleba; (iii) small sterile basal pad in one specimen.

Micro Morphology

Spores globose, deeply amyloid, densely spinose, spines up to 2 μ m tall, isolated (no reticulum), 7.5-10.5 μ m diam. Pellis cellular, with overlying entangled epithelium of trichodermial hyphae emerging sometimes very abundant, scarce.

Comments

The two-layered peridium with an overlying entangled epithelium and underlying cellular subpellis, globose spores densely covered with spines up to 2 μ m tall suggests *Cystangium seminudum*. These collections have similar spores to *Cystangium balpineum* but differ from that species by lacking any rosy colours of the peridium. *Cystangium sessile* also has similar spores but has a sublamellate or more often loculate gleba, strongly developed stipecolumella, and the spines on the spores are not crowded.

Descomyces sp. nov. BOUGHER 372; 373 Macro Morphology

Characteristic Features: (i) white thin smooth peridium without yellow fibrils; (ii) loculate brown gleba; (iii) white basal pad; (iv) limoniform ornamented spores; (v) peridium predominantly hyphal but some inflated elements seen..

Micro Morphology

Spores broad ellipsoid to sublimoniform spores with subreticulate ornamentation, ornaments up to 2 μ m tall, tightly enveloped by a non-inflating perisporium, apex smooth not or barely mucronate emerging; hilar appendix broad entire truncate not protruding; (12.7) 13.3 – 15.3 x (7.5) 8.0 – 9.9 μ m. Many aborted or distorted spores, and occasional giant spores present. Basidia consistently 4-spored, cylindro-clavate 31-43 x 6.3-7.7 μ m. Clamps present. Peridiopellis predominately hyphal but some scattered inflated elements present.

Comments

This is an undescribed species of *Decscomyces* characterized by the broad ellipsoid to sublimoniform spores with a subreticulate ornamentation, 4-spored basidia, and predominantly hyphal peridiopellis. *Timgrovea ferruginea* also has quadrisporic basidia, and similar spores, but its spores are more citriniform with a prominent mucro, and have a more complete reticulum.

This new species has similar spores to *D. giachini* but that species has a cuplike hilar appendix on many of its spores, larger spores, and bisporic basidia.

D. albellus has a similar appearance in the field and a hyphal peridium but has larger more mucronate spores, bisporic basidia, and golden hyphae in the pellis. *D. angustispora* is another *Descomyces* with quadrisporic basidia, but differs by having a bright chestnut gleba, and larger, ellipso-fusoid spores.

Elaphomyces sp. nov. BOUGHER 387 Macro Morphology

Characteristic Features: (i) fruitbodies deeply embedded in a dense clod of soil, red mycelium and red mycorrhizal roots; (ii) outer peridium black, thin but tough-leathery surrounded by soil/mycelium mass; (iii) gleba pale ash grey becoming slate grey and powdery. Maybe related to a species designated as H1563 from Tasmania in 1990, or at least similar to that one?

Fruit bodies: 5-20mm diam.; globose to phaseoliform; deeply embedded within



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a dense matrix; up to 40mm wide. Peridium: outer peridium a thin (less than 0.5mm) but tough-leathery layer, black in cross-section and with a black surface that appears smooth to the eye, but undulating/cratered under high power lens and mat or slightly felty (not shiny). Inner peridium a thick (about 1mm) whitish layer of solid but spongy/rubbery material. Outer peridium immediately covered by the external matrix but this can be cleanly removed from the peridium by lifting/scraping away with a scalpel. Matrix: Consists of densely packed dry soil, roots and mycelium. The mycelium is bright red (near 9B8, but more fire engine red than that) and is rhizomorphic in parts and loosely envelopes short roots to form mycorrhiza. Under microscope the hyphae are pale but have abundant coarse re granules on the walls. The matrix follows the shape of the embedded fruitbody i.e. can be globose. Gleba: At first solid pale ash grey rubbery, becoming darker slate grey (near 11F2) from centre outwards, then powdery (masses of globose spores can be seen under lens), and some thin white threads in the gleba (not many, and more or less oriented radially but not attached to the peridium.

Micro Morphology

Spores globose, very dark brown in water, black to dark olive brown in KOH, densely covered by crowded pegs up to 1.2 μ m tall flat-topped, bases isolated but some are irregular /shortly reticulate, neatly topped with a perisporium, 9.4 -12.4 μ m diam. Young spores in clusters, appear angular, subtended by septate, hyaline spiral hyphae 3-9 μ m broad. Black outer peridium with hyaline smooth thick-walled black hyphae in a tight parallel arrangement, and red-encrusted hyphae, no clamps seen. No asci observed.

Comments

This is a new species of *Elaphomyces* to be included in a world monograph of the genus currently in preparation. Description and images of this collection forwarded to monograph authors in USA in October 2007.

Gymnomyces sp. nov. 1 BOUGHER 369 Macro Morphology

Characteristic Features: (i) very thin smooth white peridium bruising dull tan; (ii) gleba loculate with dominant radial orientation, becoming sublamellate at the base of the fruit bodies; (iii) no latex seen; (iv) large size; one fruit body 40mm x 20mm tall.

Micro Morphology

Spores globose, dark amyloid, broken reticulum with ridges up to 1 μ m tall, plage not conspicuous, 7.7 -10.7 μ m. Basidia quadrisporic, 40-52 x 9-11 μ m. Cystidia scattered, not protruding hymenium, cylindrical (mainly) to slender clavate and oil filled when mature, clear and with apical mucro when young, 33-45 x 5-6.5 μ m. Pellis hyphal, entangled septate hyaline hyphae 5-7.5 μ m broad, compacted into a cutis at surface, no emerging hyphae, no oeliferous hyphae, sphaerocyst nests in pellis trama.

Comments

Rusty brown discolouration of the otherwise white lamellate lower gleba and peridium is distinctive. Not considered as *Macowanites (Russula)* because the columella development is not strong or consistent.

It has similar colouration and colour change to *G. pallidus* but does not have the pileicystidia or a turf of surface pellis hyphae of that species. BOU369 has predominantly cylindrical hymenial cystidia whereas *G. pallidus* has more ventricose cystidia.

Gymnomyces sp. nov. 2 BOUGHER 370

Macro Morphology

Characteristic Features: (i) shiny smooth yellowish (potato-like) peridium; (ii) cream loculate gleba.

Micro Morphology

Spores amyloid, globose to broad ellipsoid, coarsely warted (isolated warts), 6.6-8 x 6.8-8.8 μ m. Peridium highly hyphae up to 10 μ m broad in gelatinised layer. Subpellis hyphal. Hymenial cystidia abundant-crowded, lageniform,



BOUGHER 00369





ventricose-rostrate, subulate, 40-65 μ m. long, base 4.5-7.0 μ m., neck 2.5-3.3 μ m., septate, protruding up to 40 μ m from hymenium. Basidia 4-spored, 30 x 8.5 μ m.

Comments

This seems to be a unique undescribed species characterised by: (a) extremely large and abundant crowded, hymenial cystidia, (b) broad, gelatinised peridium which macroscopically appears smooth and yellowish like a potato.



Characteristic Features: (i) cream loculate gleba (no latex observed); (ii) thin, smooth cream peridium; (iii) columella absent; (iv) micro-peridium a cutis (no inflated elements present). Same species as BOU 376.

Characteristic Features: (i) cream loculate gleba (no latex observed); (ii) thin, smooth cream peridium; (iii) columella absent; (iv) micro-peridium a cutis (no inflated elements present). Same species as BOU 375.

Micro Morphology

Spores globose, strongly amyloid, broken reticulum, ridges conspicuous in profile, 6.6-7.4 μ m diam. Cystidia scattered, infrequent, mainly stubby fusoid-ventricose-rostrate. Peridium a hyphal, broad layer with abundant oeliferous hyphae including with ventricose ends in some. Basidia quadrisporic.

Comments

This species lacks the orange to brick red patches on the peridium (as occur in *G. westresii*). It has globose spores with a broken reticulum. However, the reticulum is coarser and more resembles that of *G. glarea. G. westreii* spores are broad ellipsoid, not globose. Resembles *Gymnomyces pallidus* which also has oeliferous and cystidioid elements in pellis and fusoid-ventricose cystidia, but that species has larger spores.

Hysterangium cf. *affine* = *H*. sp. white messy BOUGHER363; 378; 389 Macro Morphology

Characteristic Features: (i) dull greenish gleba, gelatinised; (ii) peridium 1mm thick, becoming pinkish when cut, surface cream (not changing), smooth, with adhering roots. (iii) associated with a thick white mycelial mass with adhering sand and roots. (iv) Abundant fruit bodies.

Micro Morphology

Spores narrow fusoid, smooth, without loosening perisporium or barely loosening perisporium, 10-11 x $3.4 - 4.2 \mu m$, with or without projecting hilar appendix.

Comments

On Bald Island, this fungus produces more fruit bodies in single clusters than the other truffle species observed there so far. The white peridium has abundant roots often coated with white mycelium. The peridum dulls upon handling.



BOUGHER 00370

cm





Hysterangium sp. cystidioid BOUGHER 379; 380; 392 Macro Morphology

Characteristic Features: (i) pale yellowish-greenish (near 5C3) not loculate, gelatinised gleba; (ii) small basal pad; (iii) peridium 0.5mm thick, surface smooth whitish; (iv) base attached to elastic white mycelium (fruit bodies do not seem to be embedded in the mycelium).

Micro Morphology

Spores fusioid, perisporium loosening. Cystidia, and clamp connections observed.

Comments

The peridium dries pale kahki green (compared for example with BOU367 & 390 which dry dull brown). Cystidia are not a common structure among *Hysterangium* species.



Characteristic Features: (i) smooth, dull pink, thick peridium (pink *in situ*, before any handling) without any adhering roots or soil; (ii) dull greenish gelatinised gleba.

Micro Morphology

Spores fusoid, perhaps minutely densely vertuculose. Perisporium adhering tightly. Hilar appendage truncate, not projecting.

Comments

This species is characterized by its thick, smooth peridium which lacks adhering roots or soil and is pink in the field before handling and dull pink upon handling. The peridium of this species is distinctive as it tends to clearly split and separate from the gleba when handled. Macroscopically this species is similar to other species with pink or red peridia such as *H. salmonaceum*, which differs by having narrower spores with a loose perisporium.

Mesophellia brevispora BOUGHER 368; 381 Macro Morphology

Characteristic Features: (i) pale cream solid central core, no veins or pockets in core; (ii) spore mass powdery, dull greyish-greenish (near 3E2-4E2) with only a few scattered inconspicuous trabeculae and those not particularly radially oriented nor extending to the outer peridium; (iii) outer peridium a thin but brittle layer in section, whitish inner surface with embedded blackish mycorrhizal roots, and with a dry layer of roots and soil densely packed about 1mm thick on outside surface; (iv) core not easily dislodged from spore mass, and without any distinct cutis around it.

Micro Morphology

Spores: ellipsoid to subfusoid with conspicuous but short basal collar, smooth, thin-walled, dull greenish-grey in KOH. 7.5-9.6 x 4.1-4.9 μ m

Oil globules floating free in abundance

Gleba Hyphae: 6.0- 9.5 μ m broad. No clamps seen. Smooth, with walls up to 2 μ m thick. ; glebal core solid; locules absent in gleba core; occasional globose cell 16.9 μ m diameter; oeliferous hyphae present; some bundles of parallel tightly packed; endocutis thin, papery hyphae are present, and could be interpreted as 'veins'. Hyphae of trabeculae: 4 to 5 μ m broad, smooth, thin-walled, tightly packed parallel, partially agglutinated.

Comments

The following suggests these collections are *M. brevispora*: (i) spores mainly under 9 μ m long; (ii) gleba core easily detached, attached by abundant fine trabeculae; (iii) gleba not loculate. However the collections from Bald Island do not have a yellowish-grey spore mass (they are dull greyish-greenish near 3E2-4E2), or an inconspicuous cup on the spores, or obvious open pockets in the gleba core. In having greenish, spore mass and lacking pockets in gleba core it also closely matches *M. clelandi* which is known only from eastern Australia.

Two potoroos were trapped near this location on same day. I noticed a trail of *Mesophellia* shells along a track and followed it to a dry patch of soil which turned out to have a good cluster of fruit bodies (BOU 381). This is the same species, from same approximate site in 2005 - identified by the current author as *M. brevispora* (E8198).

Pogiesperma sp. BOUGHER 362; 384 Macro Morphology

Characteristic Features: (i) dull pink minutely loculate gleba; (ii) thin, onelayered, whitish, smooth peridium dulling slowly after handling; (ii) no sterile tissue (base or columella).

Micro Morphology

Spores fusoid to ovoid, 6-7 x 2.5-3 µm, smooth, some transversely septate?.





Comments

This species occurs throughout southern Western Australia, typified by its dull pink gleba and thin, smooth white peridium. The name *Pogiesperma* is not yet validly published as a genus because fungi with morphology considered to represent *Pogiesperma* have been found to include a polyphyletic assemblage of fungi.

Protoglossum sp. BOUGHER 365; 366

Macro Morphology

Characteristic Features: (i) thick gelatinised peridium (glutinous not viscid); (ii) small short white button-like stipe at base: (iii) young basidiomes dull purplish, colour soon fading.

Micro Morphology

Spores ellipsoid, some slightly adaxially flattened, 16-19.5 x 9.4-12.7 μ m coarsely verrucose with loose hyaline perisporium, very few spores adhering in tetrads or pairs. Short smooth mucro evident in some spoes. Peridium a broad gelatinous matrix with sinuous loosely intertwined narrow clamped hyaline hyphae. Subpellis of broad, brown, encrusted hyphae (not polygonal) arranged parallel to the surface.

Comments

Resembles *Quadrispora pyriformis* in having large coarsely verrucose spores, some adaxially flattened, and a broad gelatinised peridium. But differs from it by having very few spores in tetrads (only one per several microscope views), ellipsoid rather than ovoid spores, and a prominent hilar appendix. The shape of the basidiomes with a small white or pale basal pad and thick gelatinised peridium is similar to the form of *Protoglossum luteum*, which has orange rather than purplish tinges and broad ellipsoid to subglobose spores. Compared with other named Australian species, the large size of the spores and coarse ornamentation are closest to *Thaxterogaster leucocephalus*. But that species has a pale peridium and smaller spores 12.5-14.5 x 8-11 microns (Beaton *et al.* 1985). *Protoglossum violaceum* occurs in WA. It has a violet peridium that fades with age but the peridium is narrower, and its spores are less coarsely ornamented than BOUGHER 365 and 366.

Pseudohysterangium / Hysterogaster BOUGHER 374 Macro Morphology

Characteristic Features: (i) white thin smooth peridium; (ii) white minutely loculate gleba.

A single specimen only.

Micro Morphology

Spores hyaline in KOH, dextrinoid, thick-walled, minutely densely vertuculose, tightly adhering perisporium, truncate hilar appendix not protruding, 11-12 x 4.6-5.1 μ m. Trama highly gelatinised, dextrinoid, no clamps, some inflated sphaeropedunculate elements in trama. Peridium a broad layer of polygonal cells.

Comments



BOUGHER 00384

BOUGHER 00366

BOUGHER 00374

cm



<u>Appendix 2</u>: Fungi spore types in scats of Gilbert's Potoroo translocated to Bald Island. 'Closest matching fruit bodies' & 'Observed in x of 4 individual potoroos' refers to collections at Bald Island during the current study only. Spore types shown in blue matched fruit bodies. Spores not shown to equivalent scale. See Table 3 for further details of spores.

Spore Type	Comments	Spore Type	Comments
Spore type 1	<i>Gymnomyces</i> sp. nov. 2 Closest matching fruit bodies BOU 370 Observed in 4 of 4 individual potoroos.	Spore type 2	<i>Gymnomyces boranupensis</i> Closest matching fruit bodies BOU 375; 376 Observed in 2 of 4 individual potoroos.
Spore type 3	<i>Gymnomyces</i> sp. / <i>Cystangium</i> sp. No spores from fruit bodies match. Observed in 4 of 4 individual potoroos.	Spore type 4	Zelleromyces daucinus Closest matching fruit bodies BOU 382 Observed in 4 of 4 individual potoroos.
Spore type 5	Unknown Ascomycete No spores from fruit bodies match. Observed in 1 of 4 individual potoroos.	Spore type 6	<i>Chondrogaster</i> sp. Closest matching fruit bodies BOU 367; 390 Observed in 3 of 4 individual potoroos.
Spore type 7	Protoglossum / Cortinarius No spores from fruit bodies match. Observed in 1 of 4 individual potoroos.	Spore type 8	<i>Cystangium seminudm</i> Closest matching fruit bodies BOU 85; 386 Observed in 2 of 4 individual potoroos.
Spore type 9	Pseudohysterangium Hysterogaster Closest matching fruit bodies BOU 374 Observed in 3 of 4 individual potoroos.	Spore type 10	<i>Elaphomyces</i> sp. nov. Closest matching fruit bodies BOU 387 Observed in 3 of 4 individual potoroos.
Spore type 11	<i>Hysterangium</i> cf. <i>affine</i> Closest matching fruit bodies BOU 363; 378; 389 Observed in 4 of 4 individual potoroos.	Spore type 12	<i>Hysterangium</i> sp. Closest matching fruit bodies BOU 392 Observed in 4 of 4 individual potoroos.

Spore type 13	Austrogautieria sp.	Spore type 14	Glomus sp.
	No spores from fruit bodies match.	(No image available)	No spores from fruit bodies match.
	Observed in 4of 4 individual potoroos.		Observed in 1 of 4 individual potoroos.
Spore type 15	Protoglossum sp.	Spore type 16	Protoglossum sp.
	No spores from fruit bodies match.		Closest matching fruit bodies BOU 362; 384
	Observed in 4 of 4 individual potoroos.		Observed in 1 of 4 individual potoroos.
Spore type 17	Quadrispora tubercularis	Spore type 18	Pogiesperma sp.
	No spores from fruit bodies match.	0080	Closest matching fruit bodies BOU 362; 384
	Observed in 1 of 4 individual potoroos.	8	Observed in 4 of 4 individual potoroos.
Spore type 19	Unknown	Spore type 20	Protoglossum sp.
(No image available)	No spores from fruit bodies match.	Contraction of the second	No spores from fruit bodies match.
	Observed in 2 of 4 individual potoroos.		Observed in 1 of 4 individual potoroos.
Spore type 21	Hydnoplicata convoluta	Spore type 22	Gymnomyces sp. ?
1800	No spores from fruit bodies match.		No spores from fruit bodies match.
	Observed in 3 of 4 individual potoroos.		Observed in 1 of 4 individual potoroos.
Spore type 23	Unknown	Spore type 24	Unknown
200	No spores from fruit bodies match.		No spores from fruit bodies match.
Contraction of the second	Observed in 2 of 4 individual potoroos.		Observed in 1 of 4 individual potoroos.
Spore type 25	Hysterangium inflatum	Spore type 26	Gymnomyces sp. ?
-	No spores from fruit bodies match.	-	No spores from fruit bodies match.
	Observed in 1 of 4 individual potoroos.		Observed in 2 of 4 individual potoroos.
Spore type 27	Gymnomyces sp. ?		
- AF,	No spores from fruit bodies match.		
	Observed in 1 of 4 individual potoroos.		

Appendix 3: Truffle fungi species obtained as fruit bodies from Ryedene during the current

study. Also see summary listing in Table 3.



Pogiesperma sp. A BOUGHER 324; 327; 328

Macro Morphology

Characteristic Features: (i) pure white smooth, thin gleba, not bruising/staining; (ii) peridium smooth, thin, white in section; (iii) gleba pinkish, minutely loculate, no radial pattern; (iv) no sterile tissue/columella.

Micro Morphology

Spores cylindric, thin-walled, smooth or very minutely vertuculose or dimpled, some appear transversely septate, no germ pore, apiculus inconspicuous, hyaline in KOH, yellowish in Melzers, 6.6-7.7 x 2.6-3.4 μ m.

Comments

An abundant species characterized by its pinkish gleba. Probably the same species as *Pogiesperma* sp. (Bougher 362, 384) from Bald Island.

Pogiesperma sp. B BOUGHER 292

Macro Morphology

Characteristic Features: (i) pinkish, chambered gleba; (ii) exuding clear latex, sticky.

Fruitbody: up to 15mm wide, globose to ellipsoid to reniform. Peridium: thin (less thatn 0.5mm), 1 - layered white in section; surface dark dull yellowish, cream when younger, smooth, dry. Gleba: empty locules, pale pink, trama whitish, no radial pattern. Latex: clear, sticky. Rhizomorphs: none, basal mycelium inconspicuous. One specimen has white dendroid intrusions through the gleba.

Micro Morphology

Spores cylindric but stockier than for BOU324, smooth (no vertucosity visible), $5.0-6.2 \times 3.0-3.8 \mu m$.

Comments

Differs from *Pogiesperma* sp. A by having dendritic sterile intrusions in the gleba, and shorter non-vertuculose spores. Macroscopically it could be mistaken as a russuloid fungus, i.e. as *Gymnomyces* or *Cystangium*.



<u>Appendix 4</u>: Truffle fungi species obtained as fruit bodies from Waychinicup National Park

during the current study. Also see summary list in Table 4.





Zelleromyces daucinus BOUGHER 299; 303

Macro Morphology

Characteristic Features: (i) red-brown peridium; (ii) residual short emergent stipe, and radial orientation of gleba at base.

Micro Morphology

Spores globose, with coarse complete reticulum of deeply amyloid ridges up to 1.3 μ m tall, 8.3-10.3 μ m diam. Thick-walled cells comprising the peridium. Lactiferous hyphae in trama.

Comments

Epithelial peridium structure and spore type conform with *Zelleromyces daucinus*. Matches the specimens of this species from Bald Island (BOUGHER 382).



