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Observations on the genus *Omphalotus* in Australia

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Summary. A phenotypically variable taxon with luminescence and subglobose to short elliptical spores has been described as a *Pleurotus* under several different species names in Australia. The variable levels of luminescence and general coloration of the fruiting bodies suggested that more than one taxon might be involved. Genetic analysis revealed a bifactorial matingsystem. Intercompatibility of all phenotypic variants was achieved using single spore isolates. A new combination in the genus *Omphalotus* has been proposed for this taxon.

KEY WORDS: Basidiomycetes, Agaricales, Tricholomataceae.

Introduction

Cleland (1934–35) treats *Pleurotus nidiformis* Berk. under the name *P. lampas* Berk. listing as synonyms, *P. candescens* Muell. and *P. phosporeus* Berk. and "doubtful *P. nidiformis* Berk". He provides a general description, the spore size and shape, and an account of the ecology. A wide distribution of the species in Australia is indicated by Cleland.

Macdonald & Westerman (1979) in their "Guide to the Fungi of Southeeastern Australia" illustrate a predominantly white form of the species and list the taste as mild and the smell pleasant. Young (1982) in his book "Common Australian Fungi" illustrates both the brown and white forms and is the first author to list it as poisonous. He also indicates that it is on "old *Eucalyptus* stumps or dead trunks" as well as on a variety of wood substrates and distributed throughout most of the states in Australia. Fuhrer (1985) in the "Field Companion to Australian Fungi" illustrates the brown form of the species along with a color illustration of the luminescent lamellae. He also indicates that the fungus is poisonous. Griffiths (1985) in his "Field Guide to the Larger Fungi of the Darling Scarp & South West of Western Australia" illustrates the brown form. He also gives additional information on the nature of the poisoning and indicates that one vomits but experiences no ill-effect after the initial sickness. Once again the luminescence ("phosphorescence") is described and he points out that it was first described from Western Australia in 1844 by Berkeley). Lastly, Shephard & Totterdell (1988) illustrate the white form but also describe the brown form and note that the color is quite variable.

The various authors list it from every state except the Northern Territory. It is interesting that the two rather distinctive forms are illustrated in both color photography and paintings but not mentioned as such.

Our study of the species in the field over three field seasons in Western Australia indicated that there might be two closely related species. The white form luminesced so strongly that only a few seconds to 30 seconds were necessary in the dark to clearly see the luminescent lamellae while the brown form by contrast may require about 4 to 7 minutes in the dark because of the lower level of luminescence. However, brown forms have also been sampled during the present study that strongly luminesced as described for the white form above.

It was thought that this obvious difference should be investigated. Pure cultures of several collections were made including single spore isolates of both forms in order to carry out genetic testing.

In 1844 Berkeley first described *Pleurotus nidiformis* with a cap color as "reddish brown". He subsequently described a second luminescent taxon, *P. lampas* (Berkeley 1845). This color of this taxon was described as "quite entire and pale, then deeply lobed and gradually passing through various tawny shades into deep brown or black". This would seem to be a recognition of the white form which often has a dark brown or black center. The white form also gradually darkens somewhat in age. Cleland (1916) also recognized the variability of the pileus coloration and assumed that the several names given to it were a result of its variability.

In 1848 Berkeley also described *Pleurotus phosphorus* (Berkeley 1848) from Tasmania. He indicates that the fungus forms "dense masses, or occasionally growing singly" and that it is "pale yellowish brown". He also emphasized the strong luminescence of this species: "Mr. Gunn was able to read by its light" and the fact that "it remained luminous for six days or more after being gathered". Among the three described species the differences in luminescence as well as the differences in the pigmentation of the pileus is stressed. Our current mycological observations of the species in the field as well as illustrations in contemporary mycological field guides in Australia would suggest that there might be more than one luminescent species. In addition, there is a *Pleurotus* in the *P. ostreatus* complex which occurs in the same habitats and could be mistaken for one of the luminescent taxa if microscopic analysis and the observation of the luminescence was not carried out.

Saccardo (1887) lists all three luminescent species and indicates that *P. nidiformis* is found along the Swan River (in Western Australia near Perth) and he also comments that it is related to *P. ostreatus*. However, the species referred to the genus *Pleurotus* have elliptical to long elliptical, inamyloid spores, and clavate-capitate cystidia (Vilgalys & al. 1992: Figs. 11–19). In addition, none of the species are luminescent nor are they toxic.

The objective in this study is to explore the relationship among the described luminescent Australian taxa and their phenotypic appearance and provide evidence for the placement of them in the genus *Omphalotus* rather than the genus *Pleurotus*.

Material and methods

Fresh collections were obtained and described, and the host determined. Colors were recorded using Methuen Handbook of Color (1978). The presence and intensity of the luminescence was recorded for each collection by two or more people. Spore prints were made on white bond paper. Dilutions were made from the spore print and spread on malt agar. Individual colonies were subcultured, checked for clamp connections, and placed in the Virginia Tech Culture Collection using the designation .01, .02, etc. Four collections with haploids derived from single spore isolates were analyzed including: two dark forms VT 1490 (R.N. Hilton, WA 2406) and VT 1946 (OKM 23886); two light forms VT 1948 (OKM 23908), and VT 1949 (OKM 23906).

Genetics

Haploid isolates, previously derived from single spores previously checked for clamp connections, were used in all experiments. Subsequent selfing of VT 1948 (Fig. 1) reveals a typical bifactorial mating system. Selfing of 12 single spore isolates (ssi's) of VT 1946 (not shown) yielded 3 of the 4 mating types and VT 1946.04 was the lone member of the second bifactorial pair. Mating among ssi's was then carried out confronting selected ssi's from collections representing different phenotypes. This constituted a test of the hypothesis that the distinctly different phenotypes were intersterile and represented separate taxa in the genus *Omphalotus* from Australia.

Fig. 2. shows the complete compatibility of all crosses and a rejection of the hypothesis. Note that VT 1946.02 and VT 1946.04 were selected because they were intersterile when crossed and represent tester strains from two of the four bifactorial pairs. The amended description of *O. nidiformis* includes the broad phenotypic variation encountered in this species and is given below.

Description

***Omphalotus nidiformis* (Berk) O. K. Miller, comb. nov.** Figs. 3–5.

Pleurotus nidiformis Berkeley 1844. Dec. n. 1. Lond. Journ. 3: 185.
Pleurotus lampas Berkeley 1845. Dec. n. 25, Lond. Journ. 4: 44.

Figure 1.

	.02	.06	.09	.13	.03	.04	.15	.16	.07	.12	.08
.02											
.06	-										
.09	-	-									
.13	-	-	-								
.03	+	+	+	+							
.04	+	+	+	+	-						
.15	+	+	+	+	-	-					
.16	+	+	+	+	-	-	-				
.07	-	-	-	-	-	-	-	-			
.12	-	-	-	-	-	-	-	-	-		
.08	-	-	-	-	-	-	-	-	+	+	
.14	-	-	-	-	-	-	-	-	+	+	-

Note: + denotes a successful pairing of the isolates and the presence of clamp connections within the populations.
- denotes an unsuccessful pairing of the isolates.

Figure 1. Mating tests using 12 single spore isolates O. nidiformis *(VT1948).*

Figure 2.

	VT1948					VT1949					VT1490	
	.02	.03	.04	.06	.07	.01	.02	.03	.04	.05	.08	.09
VT1946 .02	+	+	+	+	+	+	+	+	+	+	+	+
VT1946 .04	+	+	+	+	+	+	+	+	+	+	+	+

+ = presence of numerous clamp connections. VT1946 .02 and VT1946 .04 are mating incompatible.

Figure 2. Cross matrix using two tester strains with single spore isolates from 3 collections of O. nidiformis.

Figure 3. O. nidiformis *(OKM 23886, VT 1946) dark brown form with appressed radially arranged surface fibrils. – Bar = 1 cm.*

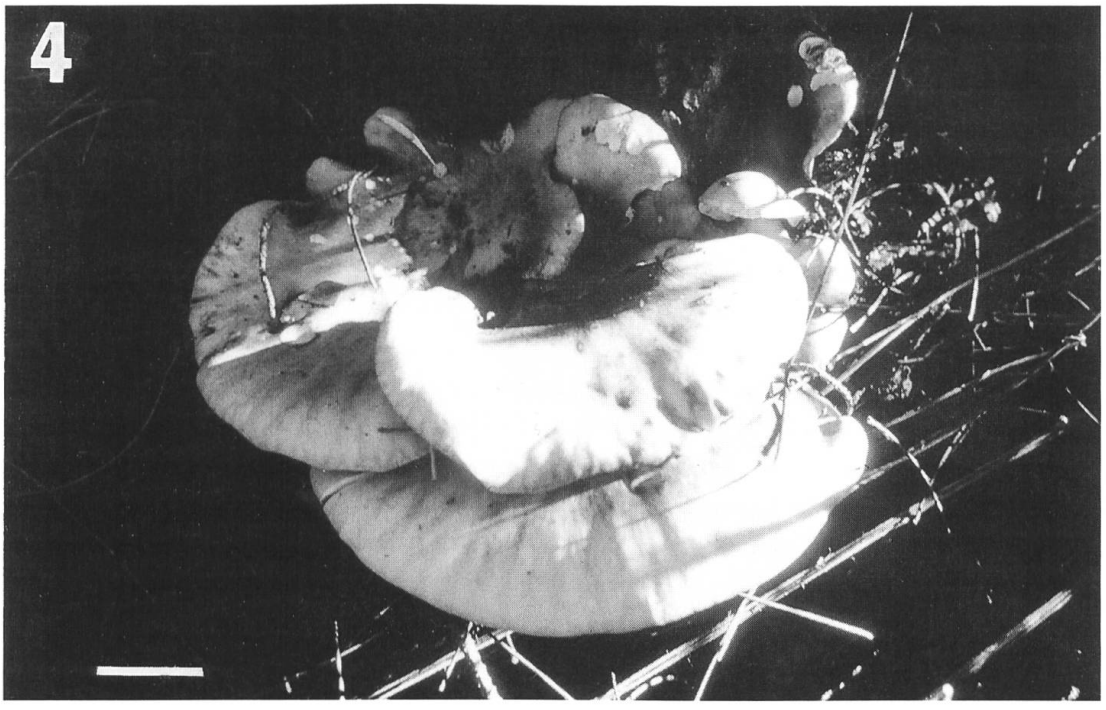
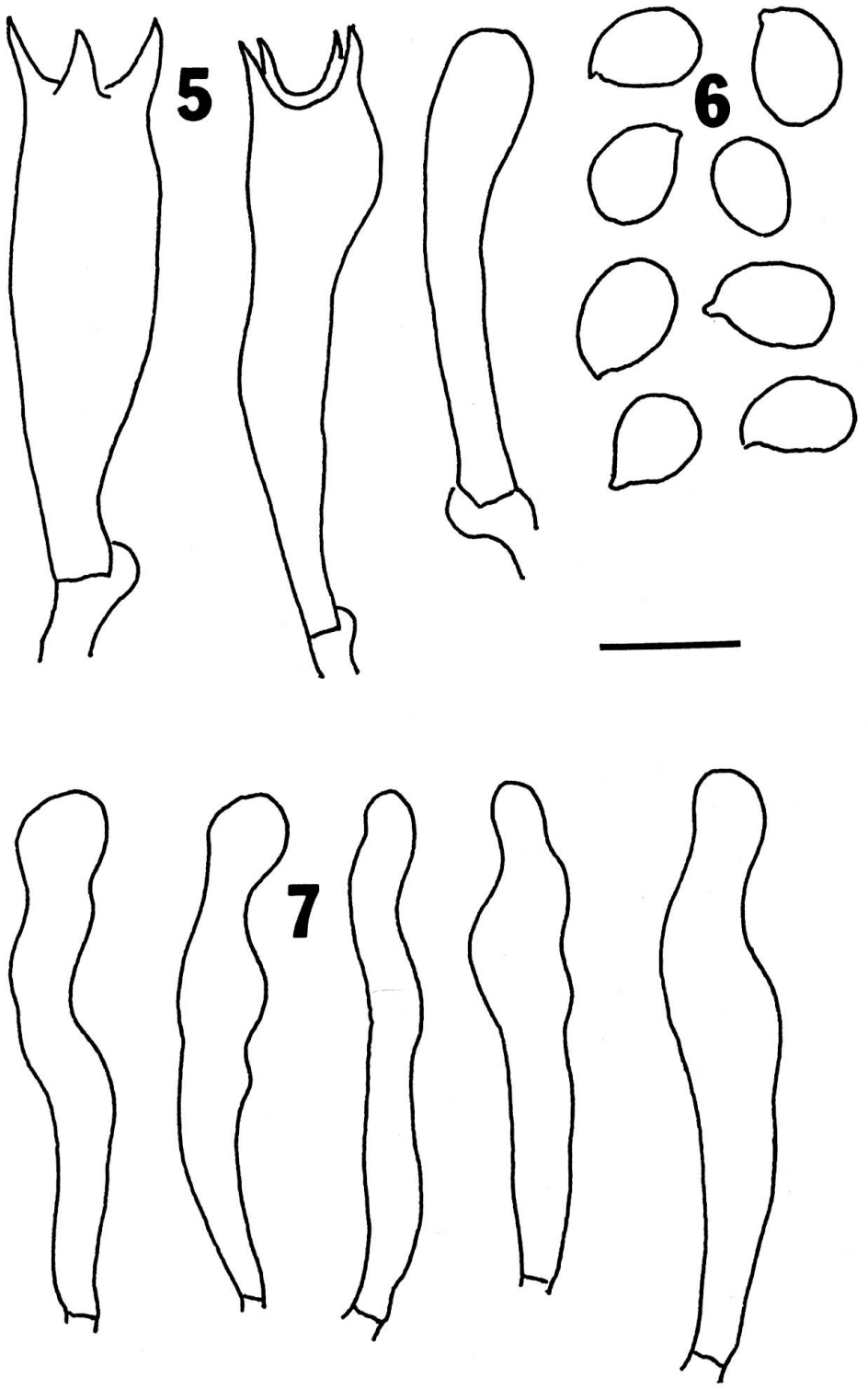


Figure 4. O. nidiformis *(OKM 2404) light form with nearly white margin and dark brown near point of attachment. – Bar = 3 cm.*

This contribution is dedicated to Prof. Dr. M. Moser (Innsbruck, Austria),
on occasion of his 70th birthday.



Figures 5–7: Omphalotus nidiformis: *5. Basidia and basidiole on right. – 6. Basidiospores. – 7. Cheilocystidia. – Bar = 10 µm.*

Pleurotus phosphorus Berkeley 1848. Dec. n. 192. Lond. Journ. VIII, p. 572.
Panus incandescens Berkeley & Broome.

Pileus (6.5–)8–14 (–18) cm broad, broadly convex to spathulate, often imbricate, dry, with appressed radially arranged fibrils or fine squamules, deep blackish to fuscous brown in center (p7F5–6, p7F4–5 to p5F4–7, p5F7–8), tinted brown (p7D7, p5C5–6) to orange brown (p5B5–6) over the margin which may also be more yellowish (p3A2) to nearly white, margin enrolled, and often somewhat crenulate in age. Lamellae decurrent to a fine line on the upper stipe, subdistant with two tiers of lamellulae, with some intervenose connections, broad, margins smooth young to eroded in age, white at first with a pinkish cast (p5A2) to yellowish orange (p4A2–3) in age. Stipe 1.3–5.4×0.7–2.5 cm wide, broad at apex tapering to a blunt base, eccentric, upper one half white, tinted buff (p3A2), lower half blackish brown (p7F6–7, p5F 6–7 to p5F4), dry with a low dense pubescence. Flesh pure white, firm and tough in cap and stipe, at stipe base often yellow under the cuticle. Odor strongly fungoid but not distinctive.

Spore deposit white. Basidiospores 6.5–8.5×(4.5–) 5–6 (–6.7) µm (E = 1.08; 1.63; Em = 1.28), short elliptic to subglobose, thin-walled, with a small apiculus, hyaline in Melzer's solution and 3% KOH. Basidia 28–47×6–7 µm clavate, thin-walled, 4-spored, with a basal clamp connection, hyaline in 3% KOH and Melzer's solution. Cheilocystidia 32–35×4–5 µm, numerous to scattered and infrequent, fusiform, thin-walled, hyaline in Melzer's solution. Pleurocystidia less frequent, but similar to cheilocystidia. Pileipellis of tightly interwoven, thin-walled, hyphae 3.5–5 µm diam, hyaline in Melzer's solution. Pileotrama of broad, interwoven, thin-walled to slightly thick-walled hyphae 4–18 µm diam, yellowish in Melzer's solution with numerous clamp connections. Lamellar trama nearly parallel to irregular, with filamentous, oval to swollen, thin-walled to slightly thick-walled hyphae 8–21 µm diam. Subhymenium a narrow layer of hyphae 2–4(–11) µm, nearly parallel, hyaline in Melzer's solution with clamp connections.

Habit, habitat, distribution: Occurs in caespitose clusters usually consisting of 3 to 5 or more fruiting bodies, laterally attached, usually imbricate, on stumps, logs, buried roots, and wounds of living trees. On a wide variety of hosts including the following recorded in this study: *Acacia* spp.; *Allocasuarina fraseriana* Miq.; *Banksia attenuata* R. Brown; *B. menziesii* R. Brown; Jarrah; *Eucalyptus marginata* (Donn ex Smith) Tuart, *E. gomphocephala* DC.; *Casuarina* sp., *Agonis flexuosa* Schau.; *Melaleuca* spp. and *Nuytsia floribunda* R. Brown. Fruiting from May to July in most years.

Material examined: AUSTRALIA: Western Australia: Perth: M. Blackwell, 6 May, 1959 UWA 905 (WA); Kings Park, E. Tenraa, 10 May, 1968, UWA 1186 (WA); H. C. Broughton, 16 May, 1965, UWA 1261 (WA); M. Blackwell, June 1972,

UWA 1532 (WA); O. K. Miller, 31 May 1994, OKM 24688 (WA); Mt. Pleasant, Inez Tommerup, 11, June 1989, OKM 23908 (WA); Kings Park, O. K. Miller, 23 May 1991, OKM 24604; Kings Park, O. K. & H. Miller, 16 June, 1989, OKM 23934 (WA), OKM 23935 & OKM 24936; Kings Park, O. K. Miller, 25 June, 1989, OKM 24041 (WA); Wembley, O.K. & H. Miller, 18 June, 1989, OKM 23970; Wembley, O. K. & H. Miller 19 June, 1989, OKM 23969; Univ. of W. A. campus, O. K. Miller, 18 June 1989, OKM 23965 (WA). – Ludlow, Woonerup House, O. K. & H. Miller, 3 June 1989, OKM 23786. – Mandura, M. Pearce, 8, June 1989, OKM 23906 (WA). – Margaret River, N. Malajczuk, 22 June 1989, OKM 24038. – Denmark, Katie Syme & O. K. Miller, 10, June 1989, OKM 23886. – Mundaring, E. Paling, 13 July 1980, UWA 2406 (WA & VPI). – Parkersville, O. K. & H. Miller 17, June 1989, OKM 23959 (WA). – Two Peoples Bay Nature Reserve, O. K. & H. Miller, 22 June, 1989.

Panus incandescens Berk. & Br., Queensland, Brisbane, No. 26, F. M. Bailey (holotype; K).

Pleurotus lampas Berk., Western Australia, Swan River No. 9, Herb. Myc. Berk. (holotype; K).

Observations: Populations of *Omphalotus nidiformis* have at least two distinct color forms and are phenotypically variable in size and shape. There is a dark form with a deep brownish black center and a brown to orange-brown margin. The luminescence is confined to the lamellae, much reduced, and is visible in about 5 minutes in a dark room. There is both a dark and a light form which luminesce very brightly. The light form also has a blackish brown center but the margin is nearly white to light yellowish. The luminescence is visible in seconds to slightly over one minute and is intense throughout the lamellae, cap, and stipe. It is the strongest luminescence which I have witnessed in the genus. There is a brown form which also luminesces quickly but the luminescence is confined to the lamellae. As a result of these observations notes on fresh material were made on all the forms encountered. They were written up separately and both cultures and single spore isolates were made from each in order to ascertain if they represent more than one taxon. The results of these matings have been detailed above and it is very likely that several species described in the past are in fact phenotypic variants of a single taxon.

The general coloration is different from other members of the genus. The coloration of *O. olivascens* Bigelow, Miller & Thiers (1976) is the closest with brownish orange cap coloration but it is tinted olivaceous (Bigelow & al. 1976). Several species have an orange pileus and yellow-orange lamellae including *O. olearius* (DC.: Fr.) Sing., and *O. subilludens* (Murr.) O. K. Miller. The subglobose to broadly elliptical spores of *O. nidiformis* are about the size of those of *O. subilludens* but are a bit wider. The small flexuous cystidia, the cuticle and tramal tissues are similar to those found in other species of the genus as illus-

trated in Fig. 3 (Bigelow & al. 1976). All species in the genus, including the above taxon, have luminescent lamellae, are usually in caespitose clusters, occur on Angiosperm wood, have subglobose to short elliptical, inamyloid spores, and are toxic to man. A portion of UWA 1186 collection was eaten by a collector, who was an anthropologist, to experience the known vomicant effect. He did become ill but recovered with no lasting ill-effect.

There seems to be little doubt that *Pleurotus phosphoreus* Berk. is also a synonym of *P. nidiformis* but no type material of it or of *P. nidiformis* can be located at Kew. In addition, there is no original illustrations at Kew (pers comm., D. Pegler). Material in poor condition of *Pleurotus lampas* Berk. and *Panus incandescens* Berk. & Br. is at Kew and was sent to the author. However, both holotypes are badly decomposed and so filled with mold that no hymenium now exists. *Panus incandescens* holotype consists of three specimens pasted onto a paper label and Brisbane is listed on the label. They are typical in life form but yield no useful microscopic characters. *Pleurotus lampas* holotype consists of two small specimens with decurrent, narrow lamellae and short central stipes. The label says "Swan River No. 9" which flows through Perth to Freemantle in Western Australia. At this point all evidence points to the existence of one member of the genus *Omphalotus* which is widely distributed in Australia.

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