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OCCURRENCE OF MYCOBIOTA IN EASTERN AUSTRALIAN SEA TURTLE NESTS

A.D. PHILLOTT, C.J. PARMENTER AND C.J. LIMPUS

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Major loggerhead (*Caretta caretta*), green (*Chelonia mydas*), hawksbill (*Eretmochelys imbricata*) and flatback (*Natator depressus*) sea turtle rookeries in eastern Australia were surveyed for the presence of fungi in turtle nests. While the fungi Fusarium oxysporum, F. solani and *Pseudallescheria boydii* were not universally present, they were isolated from the exterior of failed eggs of all turtle species and at all rookeries. Predominant fungi species varied spatially and temporally. *Sea turtle, egg, nest, fungi.*

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Globally, fungi have been described from the exterior and/or embryonic tissue of several species of sea turtle eggs (Table 1). *Fusarium oxysporum* Schlecht., *F. solani* (Mart.) Sacc. and *Pseudallescheria boydii* Negroni & Fischer have been identified from the exterior of failed eggs in sea turtle nests at Heron Island (Phillott et al., 2001). There have been no studies to determine the identity of fungi from failed eggs on turtle rookeries along a single coastline.

Heron Island (23°26'S, 151°55'E), Wreck Island (23°21'S, 151°57'E). Peak Island (23°20.5'S, 150°56'E), Milman Island (11°10'S, 143°00'E) and Mon Repos Conservation Park (24°48'S, 152°27'E) are major rookeries for the loggerhead (Caretta caretta), green (Chelonia mydas), hawksbill (Eretmochelys imbricata) and flatback (Natator depressus) turtles. These were surveyed in the 1996/97-1998/99 turtle nesting seasons (Table 2). Nests were located after hatchling emergence and excavated by hand to determine hatch success. A single unhatched egg that appeared by visual inspection to be colonised by fungus was taken from some nests (Table 2) and swabbed using MW170 TRANSTUBE®, Amies Clear Media and refrigerated at 3-5°C. Fungal swabs were incubated on half-strength Potato Dextrose Agar with 0.05gL⁻¹ chloramphenicol to inhibit bacterial growth and then subcultured onto a range of media (e.g. Potato Dextrose Agar, Nutrient Agar, Carnation Leaf Agar) as required for identification. Identification of species not included by Booth (1971) and Seifert (1996) was confirmed by Professor D.E. Ellis. Voucher specimens were lodged with the Queensland Department of Primary Industries (Accession

Numbers: *F. oxysporum* BRIP 28368, *F. solani* BRIP 28369, *P. boydii* BRIP 28370).

F. oxysporum, F. solani and *P. boydii* were isolated from sea turtle nests at all rookeries investigated (Table 2). The proportion of nests containing each fungal species varied between rookeries and, at Heron I. (the only rookery to be sampled in multiple years), between nesting seasons. Most samples were of single isolates, however 6.54% of all swabs revealed mixed cultures (1 *F. oxysporum* + *F. solani*; 2 *F. oxysporum* + *P. boydii*; 4 *F. solani* + *P. boydii*). While individual rookery/species census data often show absence of a particular fungus, all fungi were recorded from all turtle species.

As all 3 of the mycobiota isolated from Heron I. (Phillott et al., 2001) are common soil saprophytes, their presence in nests at other turtle rookeries in eastern Australia is not surprising. The temporal variation in predominant fungi observed at Heron I. (1996: *F. solani*; 1997: *P. boydii*) suggests that spatial differences in species composition cannot be solely attributed to geographic location, as it may equally have been influenced by environmental factors (e.g. hydric and thermal) and/or temporal variation.

A comparison of rainfall distribution and volume (1996: 237mm; 1997: 183mm), and air temperature (\pm SD 1996: max. 29.8 \pm 1.7°C, min. 23.1 \pm 1.6°C; 1997: max. 30.9 \pm 2.0°C, min. 24.0 \pm 1.9°C; beach temperature data not available), differed at Heron I. over the November to February sampling period between the 2 seasons. This may have contributed to the change in dominant nest mycobiota, as *F. solani* is more prevalent in areas with relatively high rainfall and low temperature (Burgess &

Turtle	Rookery	Isolate	Source	
Green Chelonia mydas	-	Aspergillus	Solomon & Baird, 1980	
Kemp's Ridley Lepidochelys kempii	-	Fusarium	Wyneken et al., 1988	
Leatherback Dermochelys coriacea	St Croix, U.S. Virgin Islands	Fusarium	Eckert & Eckert, 1990	
Loggerhead Caretta caretta	Göksu Delta, Turkey	Mucor	Peters et al., 1994	
Olive Ridley Lepidochelys olivacea Nancite, Costa R		Allescheria, Aspergillus, Cunninghamella, Gliocladiopsis, Fusarium, Homodendrum, Saksenaea	Mo et al., 1990	
	Naranjo, Nancite, Costa Rica	Fusarium, Monosporium	Acuña-Mesén, 1992	

TABLE 1. Records of fungi associated with failed sea turtle eggs.

Summerell, 1992). The germination rate and hyphal growth of all 3 fungi at varying hydric and thermal microclimatic conditions (experienced within natural sea turtle nests) is under further investigation.

Loggerhead turtle nesting populations in Queensland are in rapid decline (50-80% over 10-15yrs dependent on study site: Limpus & Reimer, 1994). Loggerhead nesting in the south Pacific Ocean occurs almost entirely in the southern Great Barrier Reef (the most significant being at Wreck I.) and Bundaberg coastline (including Mon Repos Conservation Park: Limpus, 1985). However loggerhead nests demonstrate high mortality (and associated fungal presence) on Wreck and Heron Islands when compared with green turtle nests at the same location, or loggerhead turtle nests on the adjacent mainland (Limpus et al., 1983; Phillott, 2002). Determining the role of fungi as an opportunistic contaminant of failed turtle eggs or infectant of living embryonic tissue is, therefore, of great conservation importance.

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Rookery, Season	Turtle	# Nests Excavated	# Nests Swabbed	% Nests		
				F. oxysporum	F. solani	P. boydii
Heron I. 1996/97	Green	86	22	22.7	63.6	13.6
Heron I. 1997/98	Loggerhead Green Loggerhead	2 108 5	2 19 3	0.0 5.0 0.0	100.0 5.0 0.0	0.0 100.0 100.0
Milman I. 1998/99	Green Hawksbill	7 32	5 8	40.0 12.5	60.0 75.0	0.0 12.5
Mon Repos 1998/99	Loggerhead	12	5	20.0	100.0	20.0
Peak I. 1998/99	Flatback	44	16	12.5	62.5	25.0
Wreck I. 1998/99	Green Loggerhead	39 7	16 4	18.8 50.0	62.5 50.0	37.5 25.0

TABLE 2. Spatial and temporal variation in fungal presence in sea turtle nests.

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