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Front Cover: Mangrove leaf slugs newly recorded from the Northern Territory: *Elysia leucolegnote* (above) and *E. bangtawaensis* (below). (Adam Bourke)
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New insights into Holocene economies and environments of Central East Timor: Analysis of the molluscan assemblage at the rockshelter site of Hatu Sour

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Abstract

In the central region of East Timor (the proper name for this nation being Timor-Leste) little is known of prehistoric economies beyond 2000 years ago, most previous archaeological studies having been concentrated around the Baucau plateau and eastern end of the island. The village of Laleia on the Laleia River is located 20 km east of the main district town of Manatuto on the central northern coast. Recent excavations at the nearby rockshelter site of Hatu Sour have revealed a deep archaeological sequence that dates from approx. 11,000 years ago until the recent past. This paper examines the shellfish (i.e., molluscan) assemblage from the excavation at Hatu Sour for what it can reveal about prehistoric economies and the environment of this strategic region throughout the Holocene.

Introduction

This study was undertaken as part of an Australian Research Council Discovery Project investigating cultural and environmental shifts in Holocene East Timor. This investigation involved a programme of excavation at various archaeological sites to document cultural and environmental histories. One of the key cultural remains in these sites is shellfish (Mollusca), which provide evidence not only of what dietary preferences existed but also what shellfish were locally available and therefore what niches were being exploited. This is also an excellent indicator of past environments, as shellfish are extremely sensitive to changes in their environment. Up until 2010 most of our research had been undertaken at the far eastern end of the island of Timor in the Lautem District. One of our collaborators, the Secretariat of Culture, East Timor Government, was keen for us to investigate other areas of East Timor. In 2010, we located several prospective sites in the Manatuto District on the central northern coast. Here we discuss the results,
In the central region of East Timor little is known of prehistoric economies beyond the last 2000 years, most previous archaeological studies being concentrated around the Baucau plateau and the eastern end of the island (Almeida & Zbyszewski 1967; Glover 1986; O’Connor, Spriggs & Veth 2002; O’Connor 2003; O’Connor & Veth 2005; Selimiotis 2006; O’Connor 2007; Oliveira 2008, 2010; O’Connor 2010; O’Connor et al. 2010, Oliveira 2010; O’Connor et al. 2011, Reepmeyer et al. 2011; O’Connor et al. 2012). Recent excavations at the rockshelter site of Hatu Sour on the central northern coast have revealed a deep archaeological sequence that dates from approx. 11,000 years until the recent past. The site contains a large shellfish assemblage, vertebrate faunal remains and stone artefacts, and was occupied during a period of dramatic climatic and environmental change that profoundly affected the subsistence and settlement patterns of coastal dwellers. It encompasses the Neolithic transition dating from some 3500 years ago in Timor, which originally brought pottery and possibly the domestic dog, and later subsistence agriculture and other domestic animals (O’Connor 2006). It also covers the contact period from about 1000 years ago when outside influences from Chinese and later Makassar traders to Dutch and Portuguese colonisation deeply affected the indigenous culture and economy (O’Connor et al. 2012).
Study Area

East Timor (the proper name for this nation being Timor-Leste) is located 400 km northwest of Australia in the Timor Sea, and 8 degrees south of the Equator. It shares a land border with West Timor (the proper name for this part of the nation of Indonesia being Timor Barat). Geologically, Timor is an aggressively uplifted coral limestone island. The northern and southern coasts are divided by a steep mountain range rising to 3000 m. The climate is dry tropical with a long dry season from May to November and a shorter wet season from December to April.

The study was undertaken on the central northern coast, east of the capital, Dili (Fig. 1 inset). The Hatu Sour rockshelter is located on the Laleia River, 20 km east of the main district town of Manatuto and adjacent to the village of Laleia (Fig. 1). The rockshelter (Figs 2, 3) is about 7 m by 5 m and is located in a limestone outcrop about 1 km west of the village of Laleia. Today Hatu Sour is 4 km south of the northern coastline, which drops steeply away to the continental shelf (O'Connor 2007: 530). There appears to have been Holocene infill within the embayment as indicated in Fig. 1, where the dark green marks represent recent deposition within the Laleia River estuary.

Previous investigations

Previous investigations immediately to the east, west and south of the current study area (Spriggs, O'Connor & Veth 2003; Chao 2008; Lape & Chao 2008; Forestier & Guillard 2012) have revealed a range of archaeological sites in the region: open sites and rock shelters with cultural assemblages containing stone artefacts, invertebrate remains and, in the upper levels, earthenware and imported tradeware; rockshelters with painted art; shell middens; and fortified hilltop settlements containing ancestral graves and the remains of house sites. All these sites are dated within the last 8000 years, but most within the last 2000 years.

Our surveys in the Laleia region revealed a similar range of sites, including remains of old villages with house stones and concentrations of stone artefacts, marine and estuarine shell remains, pottery and Chinese tradeware. One open site next to the Laleia
River containing a scatter of shell and stone artefacts produced the unexpectedly early date of 9500 cal. BP for the bivalve *Anadara granosa* (Table 1). There are also indigenous sites on hilltops fortified with stone walls. One particular hilltop settlement known as Leki Wakik has been occupied within living memory. These sites contain surface scatters of shell, stone, pottery and Chinese ceramics. One open site dated to 400 cal. BP from a surface shell sample (of the gastropod *Telescopium telescopium*) (Table 1). This is consistent with the ages of other indigenous fortifications, both regionally and elsewhere in Timor, mostly dated to less than 1000 years old (Chao 2008; Lape & Chao 2008; O'Connor et al. 2012).

Table 1. Radiocarbon dates from Laleia region (Hatu Sour dates after Cooling 2012: 56)

<table>
<thead>
<tr>
<th>Site</th>
<th>Spit no.</th>
<th>Lab no.</th>
<th>Sample</th>
<th>C14 (years BP)</th>
<th>Cal. years BP (95.4% probability)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kampung Baru 1</td>
<td>Surface</td>
<td>Wk-28440</td>
<td><em>Anadara granosa</em></td>
<td>9007±31</td>
<td>9840–9460</td>
</tr>
<tr>
<td>Kampung Baru 2</td>
<td>Surface</td>
<td>Wk-28441</td>
<td><em>Telescopium telescopium</em></td>
<td>798±32</td>
<td>500–250</td>
</tr>
<tr>
<td>Hatu Sour</td>
<td>3</td>
<td>ANU#26606</td>
<td>charcoal</td>
<td>315±25</td>
<td>460–305</td>
</tr>
<tr>
<td>Hatu Sour</td>
<td>12</td>
<td>ANU#27105</td>
<td>charcoal</td>
<td>6165±40</td>
<td>7168–6849</td>
</tr>
<tr>
<td>Hatu Sour</td>
<td>35</td>
<td>ANU#26609</td>
<td><em>Anadara granosa</em></td>
<td>9650±45</td>
<td>11,198–10,787</td>
</tr>
</tbody>
</table>

Methods

We excavated a 1 m x 1 m square in arbitrary 5 cm spits using standard archaeological techniques. The deposit was sieved though a 1.5 mm wire mesh screen. Finds were initially washed and sorted on site by category (bone, stone, shell, etc). When we returned to our laboratories at the Australian National University (ANU) in Canberra, the finds were further analysed. In this case, molluscan shells were sorted by species, counted and weighed. Results were entered onto a spreadsheet according to MNI (Minimum Number of Individuals), NISP (Number of Individual Specimens) and weight (g). Where the shells were broken, MNIs were based on the part of the shell that was most commonly preserved. The same part of the shell was used consistently for each excavation unit to estimate MNI. While this method potentially underestimates the true number of specimens it ensures that no individuals are counted more than once where pieces of one shell may be distributed over more than one excavation unit. Examples of taxa that could not be identified at ANU were sent to RCW for final determination.

Results

The excavation reached the limestone bedrock at 2 m, which was unexpectedly deep given the small size of the shelter (Fig. 3). The site contained large quantities of stone, shell, some bone and a few small pottery sherds on the surface. Chinese tradeware made of high-fired porcelain was also restricted to the surface. Three dates were obtained – 400 cal. BP at spit 3, 7000 cal. BP at spit 12, and 11,000 cal. BP at spit 35 (Atkinson...
2012: 6; Cooling 2012: 56), as detailed in Table 1.

Shellfish Analysis

As can be seen from Fig. 4, there was a peak in shellfish remains at spit 35 (approx. 11,000 years BP), which subsequently declined, then increased again around spits 15–12 (approx. 7000 years BP), peaked at spit 8, and declined up until the recent past.

The site contained a mixture of marine, mangrove and freshwater associated molluscan species (Table 2, Fig. 5). The major marine species were *Chiton* sp., rock dwellers mainly found in shallow water, and *Anadara granosa*, *Nerita* spp. and *Turbo* spp., all found in the intertidal zone in shallow water. There was a large number of *Turbo* opercula, as opposed to *Turbo* shells themselves. The dominant mangrove species were *Telescopium telescopium*, *Terebra palearis* and *Geloina erosae*. *Telescopium telescopium* is typically found in intertidal mudflats and mangrove forests (Willan 2013). There was only one species of freshwater or brackish taxon, *Stenomelania* sp.

Fig. 6. Distribution of marine (blue), mangrove (red) and freshwater (green) molluscan species (MNI/kg).

Fig. 7. Distribution of molluscan species by habitat and weight.
Table 2. Distribution of major molluscan species at Hatu Sour (MNI).

<table>
<thead>
<tr>
<th></th>
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<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ostreidae</strong></td>
<td>5. avorota</td>
<td>3. dundukum</td>
<td>1. vosticardium</td>
<td>1. australis</td>
<td>1. granosa</td>
<td>1. cardiae</td>
<td>1. naticae</td>
<td>1. braehmani</td>
<td>1. placenta</td>
<td>1. maxima</td>
<td>1. rubra</td>
</tr>
<tr>
<td><strong>Cardiidae</strong></td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td><strong>Venericardia</strong></td>
<td>1</td>
<td>3</td>
<td>1</td>
<td>4</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>1</td>
<td>&lt;1</td>
</tr>
<tr>
<td><strong>Haliotidae</strong></td>
<td>1</td>
<td>3</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>&lt;1</td>
</tr>
<tr>
<td><strong>Calepinae</strong></td>
<td>1</td>
<td>3</td>
<td>1</td>
<td>4</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>&lt;1</td>
</tr>
<tr>
<td><strong>Gastropoda</strong></td>
<td>1</td>
<td>3</td>
<td>1</td>
<td>4</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>&lt;1</td>
</tr>
<tr>
<td><strong>Bivalvia</strong></td>
<td>1</td>
<td>3</td>
<td>1</td>
<td>4</td>
<td>1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>1</td>
<td>&lt;1</td>
</tr>
</tbody>
</table>

Notes: Values in the table represent the number of individuals. MNI stands for Minimum Number of Individuals.
From 11,000 to 7000 years BP the MNI shows that there is steady foraging of both mangrove and marine molluscs with marine species dominating from spit 30 until spit 15, when mangrove species begin to take over. *Stenomelania* sp. was being exploited, but only in low numbers until spit 15, when its presence increased significantly. From this time post-7000 years BP, species from all habitats increased with mangrove species dominating and peaking in spit 8 (Fig. 6). There is a significant decline in all species from spit 6 onwards, although mangrove species continue to dominate. Shellfish weights show this pattern even more clearly than MNI (Fig. 7).

**Other archaeological evidence**

Turning to the evidence from the stone artefact assemblages that were analysed by Cooling (2012), there was a large peak in artefact numbers between spits 3 and 9 post 7000 BP. On the basis of dates for peaks in stone artefact deposition from other rockshelter sites in East Timor, Cooling (2012: 57, 70) cautiously assigned the peak at Hatu Sour in spits 6–8 to between 5000–3000 years BP.

Other faunal remains at Hatu Sour are mostly fishes, rodents (murids), crabs and bats (all endemic fauna), most of which appear burnt. There is also a small amount of reptile vertebrae including snake, and a few unidentified bird bones. At least three species of murid exist, one of which is quite small (probably *Rattus exulani*), another larger, and the third a giant rat now extinct in Timor (two bones were found in spits 17 and 18). Most of the fish are parrotfish, a common reef fish. Exotic fauna are only found in upper levels and include remains of pig and dog (Stuart Hawkins, pers. comm.).

The few small sherds of pottery and Chinese tradeware that were found were restricted to the surface of the site (Cooling 2012: 34).

**Discussion**

Hatu Sour's current distance from the shoreline is about 4 km. Due to the depth of the Ombai Strait between East Timor and the island of Alor to the north, the northern coast of the island drops away sharply (O'Connor 2007: 230). Consequently, the coastline would have been more or less stable throughout the Holocene period despite sea level rise. The distance of the rockshelter from the Lalcia River is currently 1 km. Before sea level stabilisation approx. 6000 years BP, rising seas would have flooded former embayments and river valleys, subsequently infilling them with sediment derived from both the land and the sea (Chappell 1988; Woodroffe 1988; Woodroffe, Thom & Chappell 1993). This scenario is suggested for the Lalcia River estuary by the Google Earth image where the green shading indicates recent infill (Fig. 1). Infill would have encouraged the expansion of mangroves on the floodplains of the river (Chappell 1988; Woodroffe 1988; Woodroffe, Thom & Chappell 1993) and potentially rendered mangrove resources closer than previously. Increased sedimentation within river systems in East Timor could also be the result of increased rainfall in the mid-Holocene. A recent review (Reeves *et al.* 2013) from northern Australia suggests that the early to mid-Holocene was warmer and
wetter than at present across tropical northern Australia with drier and more variable conditions beginning sometime after the mid-Holocene.

The persistent presence of marine, mangrove and freshwater molluscan species from 11,000 years BP until recently at Hatu Sour implies that the rockshelter was occupied continuously throughout the Holocene and the occupants had access to all these habitats. The large number of *Turbo* spp. opercula (as opposed to actual *Turbo* spp. shell remains) may relate to the foraging strategy. Living *Turbo* spp. could have been processed near to their site of collection to separate the flesh from the heavy shells. However, the opercula would firmly adhere to the foot of the animal and would therefore be returned to the rockshelter, distorting the ratio (Szabó 2009: 197, 201).

The peaks in shellfish distribution at 11,000 and 7000 years BP suggest increased regional occupation at these times with a decline in occupation between these dates. Increases in occupation can be correlated with increased productivity of the environment. The exponential increase in numbers of mangrove shellfish species from 7000 years BP can be associated with an expansion of mangroves in the river estuary as a consequence of Holocene infill, as seen in Fig. 1, and closer proximity of mangrove resources to the rockshelter.

The significant increase in the presence of *Stenomelania* sp. at Hatu Sour from spits 15 to 7 could be suggestive of increased rainfall and freshwater in the environment in the mid-Holocene that would also have allowed for an expansion of regional occupation. There is some evidence from northern Australia that this may be the case (see below). The collapse of all shellfish numbers post spit 7 suggests decreased occupation.

The argument for increased occupation post 7000 BP is supported by the large peak in stone artefact numbers around spit 7 (Fig. 8). If Cooling (2012) is correct with her extrapolation regarding stone dates based on depth, this peak period is from 5000 to 3000 years BP. Can the decline in occupation at Hatu Sour in the late Holocene, as indicated by the decrease in numbers of shellfish and stone artefacts, be associated with drier conditions? Could it also mark a concurrent increasing reliance on subsistence farming introduced about 3500 years BP? After spit 3 dated to 400 years BP, there is a drop off of overall artefact numbers suggesting a decrease in occupation (Cooling 2012: 38). Is this reorganisation of occupation coincident with European colonisation of the region? (cf O'Connor et al. 2012).

Northern Australia has the same climatic regime as Timor, being located in the wet/dry tropics. To some extent, the events recorded at Hatu Sour mirror what was happening
at sites in northern Australia during the Holocene. Following the Last Glacial Maximum approx. 20,000 years ago, rising seas flooded down-cut river valleys in the early Holocene. Subsequent sedimentation led to infill and the expansion of highly productive mangrove swamps that dominated the floodplains of northern rivers. This is known as the ‘Big Swamp Phase’ and is dated from 7000–5000 BP in northern Australia (Woodroffe, Thom & Chappell 1985; Chappell 1988; Woodroffe 1988; Woodroffe, Mulrennan & Chappell 1993). The high productivity of these swamps is reflected archaeologically in the form of extensive estuarine shellfish middens in rockshelters along the floodplains of the East Alligator River indicating widespread exploitation of mangrove environments during this period (Schrire 1982; Allen 1987, 1996; Hiscock 1999). The decline of mangrove species in Hatu Sour from spit 6 could be similar to the period approx. 5000–3000 years BP in northern Australia where further sedimentation choked off the tidal influence and restricted mangroves to the coastal fringe and river margins (Woodroffe, Thom & Chappell 1985; Chappell 1988; Woodroffe 1988; Woodroffe, Mulrennan & Chappell 1993). This was reflected archaeologically by the concurrent decline in mangrove shellfish exploitation in the rockshelters of the East Alligator River (Schrire 1982; Allen 1987, 1996; Hiscock 1999).

At Hatu Sour, relationships between climate change, changing environments and economic strategies can be clarified with further dating and isotope analysis of the archaeological shells (Bourke et al. 2007).

Conclusion
There has been continuous Holocene occupation in the Laleia region of East Timor from approx. 11,000 years BP until the recent past. Early Holocene occupation was associated with exploitation of marine and estuarine resources, with some terrestrial fauna. Increases in artefacts and mangrove shellfish after 7000 BP suggest an increase in regional occupation linked to the spread of highly productive estuarine environments. Hatu Sour shows a subsequent decrease in occupation in the late Holocene. A similar decrease in site use has been noted at other sites in East Timor after 3000 cal BP, and has often been associated with changing settlement patterns as the population is assumed to have predominantly occupied open village sites once subsistence practices changed to a farming economy. Cave sites would have continued to be used, but as hunting bivouacs rather than base camps (Glover 1986: 206–207). Hatu Sour shows further decrease in use post 400 years BP which may be due to changing settlement patterns associated with European occupation (O’Connor et al. 2012).

Acknowledgements
Thank you to the people of Laleia for allowing us access to their sites, Ministry staff, Secretariat of Culture, Timor-Leste for support, Australian Research Council Discovery Project (DP0878543) and the Australian National University for funding. Thank you to Stuart Hawkins (Australian National University) for analysing the non-molluscan faunal
assemblage. This paper was much improved by the comments of the reviewer Patricia Bourke.

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The pathogen Myrtle Rust (*Puccinia psidii*) in the Northern Territory: First detection, new host and potential impacts

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Abstract

The plant pathogenic fungus Myrtle Rust (*Puccinia psidii*) was detected within the Northern Territory on Melville Island in May 2015, five years after its arrival in New South Wales. In July the rust was found on mainland Northern Territory on the outskirts of Darwin and in September in the Darwin suburbs. Four myrtaceous plant species were found infected by the rust including the indigenous shrub *Lithomyrtus retusa*, which represents a novel host for *P. psidii*. The mode of arrival and the ecological implications of the spread of Myrtle Rust infection across Top End vegetation and plant industries are discussed.

Introduction

Myrtle Rust affects plants of the Myrtaceae family which includes many well-known natives such as eucalypts (*Eucalyptus, Corymbia*), paperbarks (*Melaleuca*), bottle brush (*Melaleuca*, formerly *Callistemon*), tea tree (*Leptospermum*) and lilly pillies (*Syzygium*). Ten percent of Australia’s flora belongs to the Myrtaceae and a considerable proportion of these plants may be vulnerable to Myrtle Rust infection.

Infection by Myrtle Rust typically causes distortion or loss of new growth and partial defoliation/dieback; thus it reduces photosynthetic capacity in susceptible plants and reduces reproductive capacity in some species if fruits are also infested. This fungal pathogen could have serious impacts on commercial and native plants, affecting plant nurseries, garden centres and forestry, tea tree and Australian native food industries.

Myrtle Rust taxonomy and biology

Rusts are plant diseases caused by fungal pathogens, specifically basidiomycete fungi of the order Pucciniales. *Puccinia psidii* is an exotic fungal pathogen of a complex of closely related species referred to as Myrtle Rust, Eucalyptus Rust or Guava Rust. It was first described from a specimen collected in 1884 from Guava (*Psidium guajava*) in South America (Winter 1884 cited in Glen et al. 2007). Myrtle Rust is now regarded as native to Central and South America (Ferreira 1983 cited in Carnegie et al. 2015, Glenn et al. 2007, Ramsfield et al. 2010). It has subsequently spread to the USA (Marlatt & Kimbrough...
1979), across the Pacific to Hawaii (Uchida et al. 2006), Japan (Kawanishi et al. 2009), China (Zhuang and Wei 2011 cited in Carnegie et al. 2015) and Australia. Myrtle Rust has recently been recorded in South Africa (Roux et al. 2013) and also Indonesia (McTaggart 2015).

Native rusts do occur in Australia (on multiple plant families) but are rare on Myrtaceae with only two rusts indigenous to three host plants. Knowledge of the rust and its life cycle are important in understanding the impact of the organism in its environment. Rusts can exhibit complex life cycles with multiple spore types (vegetative as well as sexual) and alternative hosts. Myrtle Rust can, however, complete its life cycle on a single host, rapidly producing enormous numbers of readily dispersed infectious urediniospores via asexual means. Furthermore, Myrtle Rust sometimes produces teliospores which can recombine genetic material with compatible mating types, important yielding adaptive variation (Makinson 2012). A characteristic of rusts that makes them formidable plant pathogens is their ability to evolve rapidly under selective pressure.

When Myrtle Rust arrived in Australia it was thought to differ morphologically from the holotype of P. psidii by lacking teliospores. Australian material was placed in the genus Uredo which produces solely urediniospores, and was described as a new species, Uredo rangelii (Simpson et al. 2006). However, teliospores have since been found on Australian Myrtle Rust specimens and in concurrence with a lack of molecular differences U. rangelii is now synonymised as a biotype (a strain with differential physiological characteristics) of P. psidii and not recognised as a unique species (Carnegie & Cooper 2011).

Results from recent molecular analysis indicate that P. psidii specimens from Australia are closely related to those from Hawaii (Machado et al. 2015) and also those recently studied from Indonesia (McTaggart et al. 2015). More significantly, Australian P. psidii specimens appear to be genetically uniform and not undergoing sexual recombination, suggesting that only a single predominantly asexual biotype is currently present here. Introductions of novel strains of P. psidii would however increase the likelihood of mating compatibility leading to more genetic diversity in local Myrtle Rust populations.

**Dispersal**

Unlike many fungi that can survive on dead and decaying organic matter, rusts are obligate biotrophs dependent on living host tissue for reproduction and survival. Rusts produce huge numbers of spores for wind dispersal from one host to another (Brown & Hovmoller 2002). Rusts are renowned long-distance dispersers with, for example, one race of Wheat Stripe Rust spreading from Australia to New Zealand in two months and another race spreading from western Australia to eastern Australia within a year (Grgurinovic et al. 2006). Rust pathogens are in fact intercontinental travellers (Gregory 1963, Viljanen et al. 2002 cited in Brown & Hovmoller 2002).

Myrtle Rust produces vast numbers of tiny urediniospores which are highly suited to aerial dispersal over long distances. The spore’s thick walls resist desiccation and their pigmentation resists ultraviolet radiation allowing them to survive high in the air column.
for long periods without degradation. Spore longevity is thought to be approximately 90 days (Glen et al. 2007) but would depend on ambient conditions.

Thus, vast production of spores and their ability to travel long distances enable the disease to spread rapidly. For example, \textit{P. psidii} infecting Allspice in Jamaica covered an area of 5000 km$^2$ within one year (Smith 1935 in Glen et al. 2007) and in Hawaii the disease spread to all (but one) islands within nine months (Killgore and Hue 2005).

In addition to dispersal by wind, Myrtle Rust spores are spread by moving infected plant material including nursery stock or cut flowers. At times of movement, plants can appear asymptomatic as the infection may be dormant until conditions are conducive.

Rust spores are also dispersed by human-assisted or animal-assisted means. Spores are inadvertently transported attached to clothing, vehicles, machinery, tools and other equipment (Tommerup et al. 2003) that may come in close proximity to infected plants. Animals such as bees, bats and birds can transport rust spores if they contact infected plant parts during feeding and foraging. Native bees (\textit{Tetragonula} spp.) have been observed harvesting rust spores (possibly due to the resemblance of bright orange spores and pollen) and are thus potentially implicated in transfer of the disease.

**Detection in Australia**

Myrtle Rust was first detected in Australia in April 2010 on the central coast of New South Wales in a cut flower nursery (Carnegie et al. 2010). Since this initial detection, it had spread to Queensland by late 2010 and to Victoria in 2011 and is now present across much of eastern New South Wales and Queensland. It has also been found in 2015 in northern Tasmania.

**Detection in the Northern Territory**

In May 2015, during a routine plant health inspection by Northern Australia Quarantine Strategy (NAQS), officers detected Myrtle Rust on Melville Island of the Tiwi Islands, Northern Territory. During the NAQS plant health survey Myrtle Rust was observed at four locations over the western part of Melville Island (Fig. 4) on three host species:

- cultivated Beach Cherry (\textit{Eugenia reinwardtiana}) plants;
- native mature \textit{Lithomyrtus retusa} shrubs (Figs 1, 2); and
- minor (light) infection on cultivated Weeping Ti-tree (\textit{Leptospermum madidum}) (Fig. 3).

![Fig. 1. \textit{Lithomyrtus retusa} shrubs infected by Myrtle Rust (\textit{Puccinia psidii}) on Melville Island, May 2015. (John Westaway)](image)
Fig. 2. Foliage (left) and fruit (right) of *Lithomyrtus retusa* infected with Myrtle Rust on Melville Island, May 2015. (John Westaway)

Of 20 different myrtaceous species inspected on Melville Island in May only these three host species displayed symptoms, with the indigenous *Lithomyrtus retusa* most seriously affected, suggesting this species to be highly susceptible to Myrtle Rust infection.

The Northern Territory Department of Primary Industry and Fisheries (DPIF) had been conducting surveillance for Myrtle Rust in Darwin plant nurseries since its arrival in Australia. Following the detection on Melville Island, surveillance was undertaken in nurseries and mainland properties associated with the Tiwi Islands but the rust was not found. Highly susceptible plants in Darwin, including *Eugenia reinwardtiana* and a stand of mature *Syzygium jambos*, were checked periodically by the author and found to be symptom free. It was thought the most likely pathway for introduction of the disease was via human agency with nearly all visitors to the Tiwi Islands transiting through Darwin. However an alternative pathway of cyclone-assisted wind dispersal was possible as category 4 Tropical Cyclone Lam passed from Queensland through the Gulf of Carpentaria and onto coastal Northern Territory during the February 2015 wet season, which could potentially have transported Myrtle Rust fungal spores to Melville Island.

Such an interstate dispersal event would not be without precedent as Sugarcane Smut (*Sporisorium scitamineum*) dispersed from Western Australia to Queensland on a particular weather event (Croft *et al.* 2008) and it is also likely that the fungal Grapevine Leaf Rust that appeared in Darwin in 2001 was a result of wind-born inoculum from Timor-Leste or Indonesia, where the disease is widespread (Daly & Tran-Nguyen 2008).
During a plant health survey of Garug Gunak Barlu National Park, Cobourg Peninsula, in June 2015, NAQS had an opportunity to investigate native and cultivated myrtaceous plant species for symptoms of Myrtle Rust infection. Eighteen different myrtaceous plant species were examined in the field at a range of locations over the eastern parts of Cobourg Peninsula and no evidence of Myrtle Rust infection was observed. Plants inspected included the three species found infected at Melville island – Lithomyrtus retusa, Eugenia reinwardtiana and Leptospermum madidum – the first two being highly susceptible hosts. Evidence of Myrtle Rust on Cobourg Peninsula would certainly have lent weight to the cyclone pathway hypothesis.

In July 2015, DPIF plant biosecurity officers and the author inspected local Darwin populations of Lithomyrtus retusa, the plant severely infected on Melville Island. The nearest populations are located at Berry Springs (Fig. 4) and these were found to be infected by Myrtle Rust, albeit more lightly than on Melville Island.

Myrtle Rust was subsequently detected on Syzygium armstrongii in a plant nursery in outer Darwin in September 2015. The infected plants were later destroyed. Syzygium armstrongii

Fig. 4. Locations where Myrtle Rust was found in 2015 on Melville Island, near Berry Springs and in Darwin.
had previously been recorded infected by Myrtle Rust (Giblin and Carnegie 2014), but those host plants were presumably cultivated as this species is endemic to the Northern Territory (Northern Territory Herbarium 2015). Populations of *S. armstrongii* occurring in the wild may also be susceptible to infection by *P. psidii*.

Myrtle Rust was also found to have infected two cultivated Beach Cherry (*Eugenia reinwardtiana*) plants (Fig. 5) at Darwin's Jingili Water Gardens in late September 2015.

**Potential Impacts**

Myrtle Rust infects 'new growth', i.e. actively growing shoots and sometimes also buds and fruits (Fig. 2) of susceptible myrtaceous host plants resulting in foliage die-back, reduced photosynthetic and reproductive capacity and increased likelihood of secondary disease. Infection can even lead to tree mortality in some hosts (Carnegie et al. 2015). Potential impacts include economic loss for plant nursery industries growing rust-susceptible varieties of myrtaceous plants as Myrtaceae constitute an important component of native plant nursery stock. As mentioned above, Myrtle Rust has been detected on *Syzygium armstrongii* in a plant nursery situation.

There are some small-scale horticultural enterprises in the Northern Territory that may be vulnerable to Myrtle Rust, for example Guava (*Psidium guajava, P. cattleianum var. cattleianum*) crops and edible fruiting trees variously termed Rose/Water/Malay Apple or Jambu (*Syzygium aqueum, S. jambos, S. malaccense, S. samarangense*). Currently, Myrtle Rust has not been seen infecting these cultivated Guava or 'bush apple' hosts in the Northern Territory.

Monocultures of susceptible species are particularly vulnerable. There is potential for nursery Myrtaceae to be treated with appropriate fungicides but this is less feasible for commercial crops such as orchards and silvicultural plantations. For example, commercial bush-food plantations of Aniseed Myrtle (*Anetholea anisata*) and Lemon Myrtle (*Backhousia citriodora*) have been affected in New South Wales. Commercial
Guava and Eucalyptus plantations are affected in Brazil and the Pimento industry in the Caribbean was devastated by the Myrtle Rust.

Amenity plantings are likely to suffer a decline in aesthetic value as myrtaceous plantings experience dieback. This is potentially significant in Darwin and Palmerston where myrtaceous trees are commonly used in streetside amenity plantings.

Further to the direct commercial implications inferred above, there is potential for significant negative impact on economic and spiritual values held by Indigenous people of the Top End. Two examples are the importance of Syzygium fruit ‘bush apples’ as bush tucker and the spiritual importance of vegetation communities dominated by An-binik (Allosyncarpia ternata) in the western Arnhem region (Director of National Parks 2016).

Myrtaceae

The Myrtaceae is a large and diverse family of trees and shrubs distributed primarily in the southern hemisphere with considerable tropical representation. Although worldwide, the Myrtaceae is particularly significant ecologically in Australia as genera occur in 11 of the 13 major Australian plant formations (Specht 1981) and much of the Australian landscape is characterised by vegetation communities dominated structurally and/or floristically by myrtaceous plants (Pryor & Johnson 1981). On a continental scale, myrtaceous plants represent a high proportion of plant biomass in Australia, and are thus responsible for much of the plant gaseous exchange with the atmosphere and much of the nutrient recycling with soils – both critical environmental services.

The Myrtaceae is a key iconic plant family in Australia accounting for approximately 10% of the Australian flora, with more than 2250 species from amongst 95 genera (Australian National Botanic Gardens 2015a). The Myrtaceae contains the greatest number of species of any family of plants in Australia (Beadle 1981, Anonymous 1993) and more than half of the world’s approximately 3000 species of myrtaceous plants are Australian (Australian National Botanic Gardens 2015b).

A considerable proportion of this diverse Australian Myrtaceae flora occurs in the relatively moist climatic zone east of the Great Dividing Range that has rainfall and temperature conditions suitable for survival of Myrtle Rust. Puccinia psidii has now been reported from more than 300 Australian Myrtaceae species from 57 genera. Two hundred and thirty species have been infected in the wild and a further 100 by inoculation only (Giblin & Carnegie 2014). The risk Myrtle Rust poses to the conservation of Australian flora is accentuated by the fact that more than
140 species of Myrtaceae are nationally threatened (Environment Protection and Biodiversity Conservation Act 1999; Glen et al. 2007). It is unknown how many of these threatened Myrtaceae are susceptible to Myrtle Rust infection but examples of known susceptible species include *Uromyrtus australis* and *Gossia gonodada*, both listed as endangered under the EPBC Act.

Twenty-six genera of Myrtaceae are present in the Northern Territory. Many species of these genera occur in the semi-arid and arid zones that are not conducive to persistence of fungal rust infections in general. *Puccinia psidii* is unlikely to establish in arid regions due to its requirement for an extended period of leaf wetness (Ruiz et al. 1989 cited in Glen et al. 2007). The Top End of the Northern Territory can be defined as the area subject to a tropical monsoon climate (approximating the Territory north of 16 degrees latitude or receiving greater than 600 mm annual rainfall) and the climate here is more similar to the Australian east coast than to arid Northern Territory. Long-term impacts of Myrtle Rust on the natural environment of the Top End are not known but may be of grave concern as the disease inevitably spreads. A risk map for the spread of *P. psidii* (Fig. 6) developed by Booth and Jovanovic (2012) depicts the high-risk area of suitable climatic parameters to include the Top End of the Northern Territory.

Recent predictive climatic modelling (Kriticos et al. 2013) indicates a low ecoclimatic suitability for *P. psidii* in a limited area of eastern Arnhem Land and the Tiwi Islands (Fig. 7), though the authors caution uncertainty concerning the modelled risks in the tropics. These areas coincide with those predicted by the preliminary assessment of Booth et al. (2000) (Fig. 7).

Fortunately the long extended dry season across the Top End of the Northern Territory (with no effective rainfall for 5 to 6 months) may help restrict the spread and impact of Myrtle Rust as it thrives best in humid mesic conditions. The Rust's requirement for leaf wetness (Zauza et al. 2010a) may limit its effectiveness in strongly seasonal environments although how this disease behaves in the monsoonal tropics is presently unknown. A plausible scenario may see Myrtle Rust radiating out from moist sheltered environments during the wet season and then contracting annually by the harsh conditions of the dry season back to refuges such as irrigated gardens, spring jungles and riparian vegetation. *Melaleuca* or *Syzygium* species in Top End riparian habitats may however present suitable

![Fig. 7. Climate suitability map for *Puccinia psidii* in Australia as indicated by the CLIMEX Ecoclimatic Index (Kriticos et al. 2013) left, and risk areas identified by Booth et al. (2000) for the Northern Territory, right.](image-url)
host and microclimatic conditions for the pathogen to survive the dry season, permitting more or less permanent naturalisation of *P. psidii* in at least some parts of the Top End.

The number of Myrtaceae taxa present in the Top End of the Northern Territory can be calculated by subtracting the number of arid zone Myrtaceae (Albrecht et al. 2007) that do not extend their distributions into the Top End from the total Northern Territory Myrtaceae flora (Northern Territory Herbarium 2015; Department of Land Resource Management 2014). This yields some 151 Top End Myrtaceae taxa from 21 genera; with 66 species (of 14 genera) recorded for the Darwin region alone (Dunlop et al. 1995). In contrast to most rusts that infect only a few species, Myrtle Rust is remarkable for its wide host range. Under laboratory conditions about 90% of Australia Myrtaceae tested proved susceptible to Myrtle Rust to some degree (Morin et al. 2011). Given the diversity of species found to be susceptible to Myrtle Rust in Queensland (Giblin & Carnegie 2014, Queensland Government 2015), it seems reasonable to surmise that many/dozens of Top End species are also likely to be susceptible. Great variation has been observed in the level of susceptibility of myrtaceous plants to this rust ranging from relatively tolerant (e.g. many eucalypts) to extremely susceptible (e.g. *Eugenia reinwardtiana, Melaleuca quinquenervia*). Some species, e.g. *Rhodamnia rubescens* and *Rhodomyrtus psidioides*, are impacted to the extent that many individuals die (Carnegie et al. 2015). Most susceptible species however are not killed but their reduced fitness and health are likely to affect their recruitment capacity. Susceptibility is also highly variable even among individuals of the same species (Zuaza et al. 2010b; Carnegie et al. 2015).

Many widespread common Myrtaceae species of the Top End (e.g. *Eucalyptus tetrodonta, Eucalyptus miniata*) as yet show no signs of infection and hopefully this suggests a level of tolerance, supported by the observation that mature eucalypts in eastern Australia, where Myrtle Rust has been established for longer, appear resistant to the disease. However as most Australian Myrtaceae are naive to rust disease they may yet prove to be susceptible, as pathogens are often more virulent on naive hosts (Glen et al. 2007). There is possibly a delay time frame before such species become susceptible, perhaps related to Rust strain (biotype), local inoculum loads and mutations rates.

*Lithomyrtus retusa*, the new host record for Myrtle Rust, appears to be especially susceptible based on observations that nearly all individuals (*n*=100s) inspected on Melville Island were infected (with most being severely infected) (Figs 1–3) whilst all other Myrtaceae (e.g. *Eucalyptus, Corymbia, Melaleuca, Lophostemon, Calytrix*) in close proximity showed no symptoms. The genus *Lithomyrtus* has its evolutionary centre in the Northern Territory with all but two of the ten species

![Fig. 8. Lithomyrtus retusa collections at Australian herbaria. (Map courtesy of AVH)](image-url)
occurring here, *Lithomyrtus retusa* is a widespread species across northern Australia (Fig. 8).

By contrast, seven of the other eight *Lithomyrtus* species that occur in the Northern Territory are endemic to the Northern Territory including the fire sensitive *Lithomyrtus linariifolia* which occurs amongst sandstone outcrops on the western Arnhem Land Plateau. Applying IUCN conservation criteria *L. linariifolia* is listed under the *Territory Parks and Wildlife Conservation Act 2000* as ‘Vulnerable’ to inappropriate fire regimes on account of its obligate seeding regeneration method and also vulnerable to stochastic events due to its small population size (estimated at <1000 mature individuals). It is not known whether *L. linariifolia* is susceptible to Myrtle Rust. The related *L. obtusa* that occurs in coastal Queensland is reported as being susceptible (Giblin & Carnegie 2014) but conspecific status does not appear to confer susceptibility as the diverse list of susceptible versus tolerant Myrtaceae indicates (see Queensland host list, Queensland Government 2015).

Although *L. linariifolia* is the only threatened Myrtaceae species in the Top End there are several restricted range Myrtaceae of conservation value that may be susceptible to Myrtle Rust, including the iconic Arnhem Land monsoon forest dominant tree *Allosyncarpia ternata*. *Allosyncarpia* is taxonomically significant as a monospecific genus and *A. ternata* is a keystone monsoon forest plant endemic to the specialised geology of the sandstone plateau in western Arnhem Land. Although *A. ternata* is locally common in sheltered or less fire prone sites on the plateau, its distribution globally is a very limited area. Myrtle Rust has not been found on *A. ternata* in the wild but the species has been infected in a deliberate inoculation test by CSIRO (Giblin & Carnegie 2014). If *Allosyncarpia* trees were susceptible, potential impacts may include reduced recruitment and vigour, canopy loss and marginal attrition of the forest community which could expose this Arnhem Land monsoon ecosystem to further fire and weed incursion.

*Calytrix* is another Myrtaceae genus with many endemic or restricted range species in the Top End. There are six *Calytrix* species (*C. decussata*, *C. fanerola*, *C. inopinata*, *C. minima*, *C. rupestris* and *C. surdiviperana*) endemic to the Arnhem Plateau Sandstone Shrubland Complex ecological community (Department of the Environment 2015a), with all but the first two being listed with a conservation status of ‘Near Threatened’. It is unknown whether any of these endemic plants are susceptible to Myrtle Rust. A single *Calytrix* species, *C. tetragonoloba* from eastern Australia, has tested positive to *P. psidii* but only by inoculation test, not in the wild. The reduced leaf surface area and sclerophyllous nature of *Calytrix* may confer some anatomical resistance to infection.

A further nine Top End Myrtaceae – *Eucalyptus kooplinsensis*, *Kunzea* sp. Keep River, *Melaleuca stipitata*, *M. triumphalis*, *Ochrosperma sulphatum*, *Stenostegia congesta*, *Asteromyrtus hysiephala*, *Syzygium claviflorum* and *S. hemilamprum* (the first six listed being Northern Territory endemics) are all considered of conservation concern and listed as ‘Near
 Threatened” in the Northern Territory (Northern Territory Herbarium 2015). It is not known whether or not these species are susceptible to infection by Myrtle Rust.

Rock Myrtle (*Petraeomyrtus pumicea*) is another key endemic Myrtaceae species of the threatened Arnhem Plateau Sandstone Shrubland Complex ecological community (Department of the Environment 2015b) and its susceptibility to Myrtle Rust is also unknown.

Vast tracks of the Top End landscape support vegetation comprised of paperbark trees of the Myrtaceae genus *Melaleuca*, sometimes occurring as monospecific and/or dense stands. There are seven *Melaleuca* species in the Top End that form extensive vegetation communities, typically on poorly drained or seasonally inundated soils with *Melaleuca leucadendra* and *M. cajuputi* amongst the tallest and best formed tree species in the Northern Territory (Dunlop *et al.* 1995)

*Melaleuca viridiflora*, *M. cajuputi* and *M. leucadendra* are all of high ecological significance in the Northern Territory as significant character species of several swamp forest, wetland and riparian vegetation communities across the Top End. They could be regarded as keystone species for these communities due to their provision of nectar, pollen, foraging and sheltering substrates and other resources for wildlife such as birds and including migratory species. On account of their community dominance across broad geographic ranges, these *Melaleuca* species also contribute substantially to the ecological services of water regulation and carbon sequestration. As *M. leucadendra* is particularly dependent on perennial water sources its potential demise due to Myrtle Rust may have negative hydrological and biodiversity repercussions in sensitive riparian habitats. *Syzygium armstrongii* is another important Myrtaceae tree of Top End riparian habitats and this Northern Territory endemic species has recently been observed infected with Myrtle Rust in a nursery situation.

Myrtle Rust has been recorded in New South Wales and Queensland on *M. viridiflora* and *M. leucadendra*, both of which are rated as ‘highly susceptible’, and also on the closely related Broad-leaved Paperbark (*Melaleuca quinquenervia*) which is rated as ‘extremely susceptible’ (Queensland Government 2015). Myrtle Rust severely damaged naturalised (introduced) *M. quinquenervia* in Florida in 1977 (Carnegie & Lidbetter 2012) and has been reported to impact on growth rate and tree structure in eastern Australia (Makinson 2014). *Melaleuca viridiflora* is an integral component of diverse tropical lowland environments across northern Australia. If indigenous populations of *M. viridiflora* were to succumb to the effects of this pathogen then there would likely be significant detrimental ecological flow-on effects depending on the degree to which this species is impacted. Even if individual plants are not killed, reduced plant health fitness means less nectar production. Furthermore, their reproductive capacity is likely to be impaired, resulting in lower recruitment and perhaps a slow demise of this significant vegetation community with unknown but likely deleterious implications for its dependant wildlife.
Eucalypts (Eucalyptus, Corymbia and Angophora) constitute the structural and/or floristic dominant tree species of much of non-arid Australia. Nearly 80 eucalypts (approx. 10% of total) are known to be susceptible to Myrtle Rust though most of these records are from laboratory inoculation tests rather than field observations (Giblin and Carnegie 2014). Furthermore, most mature eucalypts show some resistance or have only a low level of susceptibility. It appears that the vital life stages of seedlings and saplings, as well as epicormic and coppice growth, are most susceptible to Myrtle Rust infection. This is significant ecologically in Australia for post-fire regeneration and cohort-recruiting species in native ecosystems. Some of the susceptible eucalypts include important forestry species with the major impact for native forestry likely to be on succession, as regenerating seedlings are most vulnerable (Makinson 2014).

Across the Top End and perhaps indeed northern Australia, Darwin Stringybark (Eucalyptus tetrodonta) and Darwin Woolybutt (Eucalyptus miniata) are probably the most prevalent and widespread tree species. It is not understood if either of these two species are susceptible to infection by Myrtle Rust and if so to what degree infected plants may be impacted. Testing in eucalypts indicates there is substantial variation in susceptibility within the same species and between plants from different areas (Zuaza et al. 2010b).

Ecological interactions

There is likely to be interaction at the plant community level between the pathogen, the plant host and abiotic factors such as climate and fire. The impacts of Myrtle Rust may be most significant in situations where host plants are already stressed due to climatic conditions such as drought, fire regimes, competition from weeds and other factors that have reduced the resilience of the native vegetation communities. Myrtle Rust's greatest impact may be on plant community succession. If Myrtle Rust hampers regeneration of key or dominant Myrtaceae species thus impeding their ability to compete, there is potential for major changes in plant community composition at the landscape scale. Such changes would spell habitat loss for native flora and wildlife amounting to fundamental alteration of Australia's ecology. Poor recruitment and succession resulting in canopy decline may also increase fire impacts and promote invasion of weeds into light or canopy gaps. Furthermore, abiotic consequences such as soil erosion and reduced water retention and quality may be exacerbated.

Depending on Rust strain, degree of virulence, environmental conditions and development of tolerance, this disease has the potential to alter the composition and function of forest, woodland, heath and wetland ecosystems. The extended severe dry season conditions typical across the Top End are however not conducive to the prospering of fungal rust pathogens. Top End temperatures may not always suit Myrtle Rust as spore longevity is apparently diminished at temperatures greater than 30°C (Glen et al. 2007) and spore germination rates reduce in overnight temperatures greater than 20°C (Kriticos et al. 2013).
Susceptibility and impact on host plants may vary into the future as climatic parameters such as rainfall seasonality change, possibly making some areas more favourable to Myrtle Rust and others less so. The potential for greater impacts may arise if the genetic diversity of the pathogen increases through recombination with novel strains. A genetic/evolutionary ‘arms race’ may ensue between plant hosts developing tolerance and the fungal pathogen evolving to more virulent strains. Due to the relatively rapid reproductive cycle of fungi compared to that of long-lived perennial vascular plants, the odds favour the pathogen.

Myrtle Rust cannot be eradicated and will continue to spread, as the fungus produces incalculable numbers of spores that disperse readily via wind, animals and human activity. As there is no practical way to manage the airborne spread of spores, land managers may have to adapt their management practices where possible, for example by addressing other/concomitant pressures such as fire and weeds to alleviate overall impacts. Land managers may be able to utilise management tools such as fire to assist with protection of vulnerable high conservation value vegetation.

Though we cannot eliminate Myrtle Rust from northern Australia, we can slow down its spread, manage its impacts and undertake research to discover its full host range whilst seeking longer term solutions. Maintenance and strengthening of quarantine and biosecurity practices to avoid new genetic strains of Myrtle Rust arriving in Australia will help in limiting the pathogens impacts. The Northern Territory Department of Primary Industry and Fisheries has a website with information about Myrtle Rust, and it makes the following recommendations regarding what the public can do to help reduce the spread of Myrtle Rust in the Northern Territory:

- avoid importing Myrtaceae plants from New South Wales and Queensland;
- if bringing plants in from New South Wales and Queensland, make sure they have been treated with an approved fungicide; and
- practise good hygiene when working with plants. Cleaning equipment such as secateurs after use will help reduce the spread of other plant diseases as well.

Territory residents and nursery growers are asked to report suspected infected plants by contacting the Exotic Plant Pest Hotline on 1800 084 881. It is also important to avoid plant movements into uninfected areas. If people suspect they have come into contact with Myrtle Rust then careful decontamination of clothing and equipment is required.

Makinson (2012) provides detailed advice on appropriate responses to the threat of Myrtle Rust spread including vulnerable asset identification, risk assessment, precautions, decontamination methods, hygiene protocols and options for risk reduction.

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**Asystasia gangetica** subsp. *micrantha*, a new record of an exotic plant in the Northern Territory

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**Abstract**

An herbaceous weed of the acanthus family, *Asystasia gangetica* subspecies *micrantha*, sometimes known as Chinese Violet, was found naturalised in Darwin in April 2015 and was immediately eradicated. Although cultivated as an ornamental, this plant is regarded as an invasive weed in eastern Australia where it has been established for 15 years, and is a recognised problem weed in neighbouring tropical countries. Identification and taxonomic aspects of this species are briefly discussed, as is its distribution in Australia and overseas, and its possible means of arrival in Darwin.

**Introduction**

*Asystasia gangetica* subspecies *micrantha* (Nees) Ensermu is a target weed species of the Northern Australia Quarantine Strategy which means that it has been identified as a plant that, if introduced, is likely to have substantial detrimental impacts on agricultural production and the environment. *Asystasia gangetica* subsp. *micrantha* is also on the Alert List for Environmental Weeds (Australian Government Department of Environment 2000), a list of non-native plants that threaten biodiversity and cause other environmental damage.

*Asystasia gangetica* subsp. *micrantha* is a form of Chinese Violet and belongs to the large, predominantly tropical plant family Acanthaceae. It is a perennial herb that can grow in a mat-forming habit and smother more desirable ground plants, thus potentially affecting agriculture or reducing biodiversity.

It is a major weed overseas, particularly in Malaysia, Indonesia and the Pacific Islands (Kiew & Vollesen 1997; Anonymous 2003; Hsu *et al.* 2005). In these places it infests plantations and competes effectively for soil nutrients, especially nitrogen and phosphorous (Barnes & Chan 1990: 148), reducing productivity and increasing crop management costs. It could also become an agricultural weed in Australia.

The taxonomy of *Asystasia* requires worldwide revision and *A. gangetica* is a variable species with two subspecies recognised, *viz.* subsp. *gangetica* and subsp. *micrantha*. 
Detailed botanical descriptions of *A. gangetica* subsp. *micrantha* are available in Ensermu (1994) and Kiew & Vollesen (1997), but in summary this plant is a sprawling perennial herb that grows rapidly to 0.5 m tall (higher on supporting vegetation) and can form mats due to its propensity to take root at stem nodes (Fig. 1). Leaves are oval shaped (to approx. 15 x 5 cm), opposite, and paler on the lower surface. The bell-shaped flowers (usually 15–25 mm long) are white with distinctive purple blotches in two parallel lines (Fig. 2). Club-shaped seed capsules (approx. 30 mm long) have four flattened seeds attached by hooks.

The two subspecies are closely related but differ in floral morphology with the typical subspecies having larger flowers (greater than 30 mm long) that may be blue, mauve, white or sometimes yellow but lack the purple blotches on the lower corolla lobe.
Asystasia gangetica subsp. gangetica is sometimes cultivated in tropical gardens for its large showy flowers and has also become naturalised in the Northern Territory and Queensland. The two subspecies also differ in their ecology with subsp. gangetica typically found as a relatively benign ‘garden escapee’ which does not seem as successful or aggressive as subsp. micrantha at invading bushland.

**Distribution**

Asystasia gangetica subsp. gangetica occurs from India to SE Asia but is cultivated more widely in tropical zones. This typical subspecies sometimes forms naturalised populations beyond gardens and has been recorded in settlements across the Northern Territory from Darwin to Croker Island, Maningrida, Numbulwar and Dhalinybuy in eastern Arnhem Land.

Asystasia gangetica subsp. micrantha is native only on the African continent, but is also cultivated and is now widely naturalised in Asia, the Pacific and central and southern America (for example, see Kiew & Vollesen 1997; Gorham & Hosking 2003; PIER 2006; Daniel & Figueiredo 2009; Luján et al. 2012). It can be found as naturalised populations in neighbouring countries of biosecurity interest to Australia such as Indonesia, Timor-Leste, Papua New Guinea and the Solomon Islands.

There are three main areas of establishment of A. gangetica subsp. micrantha in Australia (Fig. 3). It was first encountered in Australia in the Port Stephens area in 1999 (on the New South Wales mid-north coast) where it is established at a number of locations in and near Anna Bay and Boat Harbour (Anonymous 2003, Skinner 2015).

The species was later found at Shoal Water Bay Training area, north of Rockhampton in Queensland in 2011 where there are at least two naturalised populations established on this military land (HERBRECS). It is also established on the Gold Coast in south east Queensland by 2013 where it has ingressed into native vegetation in Currumbin Conservation Park (HERBRECS; Anonymous 2014).

Asystasia gangetica subsp. micrantha was found naturalised near the Darwin airport in April 2015 by Michael Schmid and Lesley Alford of Veg North whilst they were conducting weed management work in Darwin International Airport’s Rapid Creek Reserve (Fig. 4). This represents the first confirmed naturalised population of this taxon in the Northern Territory. It was present on Darwin International Airport land adjacent to the drain
running north to Rapid Creek from behind the Rydges Darwin Airport Resort barbecue area (12.4041°S, 130°.8807°E).

Identification was confirmed by JW and a specimen has been lodged at the NT Herbarium (JOW 4806). At this site many sprawling plants together occupied approx 5–10% ground cover (Fig. 4) over an area of about 60 m² in mulched brown clay loam at the edge of a minor drainage channel (Fig. 4). The vegetation there consisted of *Acacia auriculiformis* and *Corymbia bella*—dominated remnant grassy woodland with *Pandanus spiralis* common in the understory.

*Asystasia gangetica* subsp. *micrantha* had apparently been cultivated in the George Brown Darwin Botanic Gardens and subsequently eradicated (Anonymous 2003). There is an horticultural record of *Asystasia gangetica* from 1994 at these Gardens but the subspecies is not specified (Ben Wirf pers. comm.).

Should *A. gangetica* subsp. *micrantha* became more widely established in Australia, it could potentially impact commercial agricultural crops such as vegetables, legumes, cut flowers and horticultural and forestry (e.g. Fig. 5) enterprises (Anonymous 2003; Skinner 2015). Skinner (2015) provides a summary of this species impact and its management in Australia and internationally. Its competitive success over a wide geographical range is attributable to its fast establishment, stoloniferous growth form capable of rooting at stem nodes, rapid growth rate, early flowering and high seed production (Anonymous 2003). As an environmental weed, it is likely to have similarly detrimental effects, smothering native
flora and degrading wildlife habitat, particularly in already modified environments. The species’ ability to readily invade undisturbed native vegetation is unclear.

Asystasia plants at Rapid Creek were treated with the herbicide 2,4-D as part of proactive weed work being undertaken by Darwin International Airport who have a process in place for management of the environment of their leased lands.

Taxonomy

In a paper on morphological variation within Asystasia gangetica, Ensermu (1994) recognised two subspecies; one the large-flowered type of the species from India, throughout Asia to Indonesia and Pacific Islands, and the other a smaller-flowered African taxon, subsp. micrantha. Kiew & Vollesen (1997) tabulated morphological differences between the two subspecies. However the NSW Herbarium (PlantNet) and the Australian Plant Census (APC), an authoritative conspectus of Australian plant taxonomy, have recently relegated the two subspecies to synonymy under A. gangetica.

The Northern Australia Quarantine Strategy (NAQS) has observed and collected both subspecies in several locations in northern Australia and neighbouring countries during the course of plant health surveys and is of the view that the two subspecies are distinct on the basis of differing floral morphology and different invasion behaviour. NAQS has been conducting a molecular study to investigate genetic variation between the two subspecies (and closely related congeners) across the geographic range of A. gangetica subsp. micrantha. Preliminary results from this analysis, which will be published in the near future, strongly support the contention that A. gangetica subsp. micrantha is distinct from the typical subspecies. This is further supported by the existence of a difference in the number of chromosomes in the two taxa, with subsp. gangetica being tetraploid (four sets of chromosomes) and subsp. micrantha diploid (two sets of chromosomes). This creates reproductive isolation between the two taxa, with any hybridisation resulting in the production of sterile triploid plants (three sets of chromosomes).

Introduction to Australia

Asystasia gangetica (especially subsp. gangetica) is cultivated in tropical climates as an ornamental garden plant. It spreads by seeds released explosively from drying capsules, as well as by stems which are capable of taking root when in contact with moist soil. Dispersal over long distances is by human agency through accidental transportation of plant material in gardening, landscaping, roadworks, mining and defence activities.

The original incursion of A. gangetica subsp. micrantha in the Port Stephens area is thought to be derived from an horticultural introduction with subsequent populations having also spread from garden plantings or resulting from the dumping of garden waste. Care is required to ensure correct disposal of plant material as much of its spread has been attributed to poor disposal of plant parts which can contain seeds and broken stems that can readily establish and form new plants, causing the infestation to spread.
The means of introduction of the recent Darwin incursion at the airport may be via seed inadvertently transported by people or machinery from eastern Australia or overseas. A plausible scenario could entail a visitor from overseas staying at the cottages along the back of the resort and accidently depositing seed from his/her footwear or clothing that had travelled with them. The infestation occurred in a drain that runs off the airport so it is possible that plant material could have arrived on an aircraft, or machinery transported by air.

It is perhaps no coincidence that the Darwin incursion is near Defence land as is the Shoalwater Bay site in central Queensland, suggesting that transport via military hardware may have been implicated.

The Darwin population at Rapid Creek has been treated with herbicide and any regeneration will be monitored and treated as necessary. Soil removed from the site during drain maintenance is disposed of appropriately to ensure seed and plant material is not spread. Whether propagules were transported beyond the site prior to treatment (e.g. downstream in the Rapid Creek catchment) remains to be seen next wet season.

Acknowledgments

LA & MS thank Chris Collins of the Northern Territory Weed Management Branch for discussion of management of this species.

References


HERBRECS Queensland Herbarium database; accessed via AVH [Australian Virtual Herbarium].


Seed viability of native grasses is important when revegetating native wildlife habitat

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Abstract

Native grasses are a dynamic and essential component of the majority of terrestrial ecosystems in the Northern Territory. Restoring native grasses in disturbed environments is important for providing faunal habitat, reducing surface erosion and resisting weed invasion. However, establishing native grasses has been problematic in many regions of Australia due to seed viability issues. We investigated 48 seed lots of 29 Northern Territory native grass species to determine whether seed quality was an issue for establishment of tropical native grasses. Seed lots were largely collected by commercial seed suppliers, rather than by research staff, so the samples reflect seed lots that could be sourced for revegetation projects. The seed purity, proportions of filled seeds, visually viable seeds and metabolically active seeds were assessed. Viability responses to storage were investigated in 15 seed lots. The proportion of florets that contained a seed (caryopsis) ranged from 10—97% (average 62%) and between 0—79% of the florets contained metabolically active seeds (average 36%). Two seed lots had viability of 0—10% and 12 of the 48 seed lots had less than 30% seeds that were metabolically active and potentially viable. Thus, seed quality limits establishment of tropical native grasses from sown seeds in the Northern Territory. When using native grasses to establish native habitat it is important to assess the quality of the seeds and use a sufficient quantity of seeds for effective establishment of these grasses. Seeds of many species will maintain viability for several years if stored in cool dry conditions. Seed for revegetation projects can therefore be collected and stored over several years.

Introduction

Native grasses are a feature of the vegetation communities of the Northern Territory. Spear grasses (Heteropogon and Sorghum spp.) and spinifex (Triodia spp.) are particularly dominant grasses in tropical and arid vegetation communities. In tropical communities there is typically a considerable diversity of other native grass species present. Native grasses provide a range of valuable ecological functions including:

- providing food for granivorous mammals and birds;
- providing habitat for native fauna;
- resisting invasion by introduced weed and improved pasture species; and
- assisting with control of surface erosion.
When re-establishing native vegetation communities during revegetation activities, the establishment of native grasses is often problematic. Active establishment of native grasses as part of revegetation activities generally relies on sown seeds, and those seeds need to be of good quality. In many regions of Australia seed biology issues relating to seed viability and seed dormancy often limit establishment of sown native grasses. We suggest this is also the case in the tropics of the Northern Territory.

The seed of a grass is termed a caryopsis and this is typically enclosed within two sheathing covering layers (the palea and lemma) to create a floret and several florets are enclosed within two glumes (Fig. 1). The seed of a native grass may be dispersed either as a bare caryopsis or enclosed within the floret. Seed lots of native grasses generally contain florets, other inflorescence material such as the stigma and stamens (Fig. 2), and sometimes vegetative material, as well as the caryopses (Fig. 3). There may be a low proportion of viable seeds in a seed lot if the seed lot contains a high proportion of vegetative material, a high proportion of empty florets, or a high proportion of damaged or dead caryopses. A simple indicator of poor seed fill is to lightly press on the sides of the florets to feel the caryopsis. Alternatively, the floret

![Fig. 1. Mature 'seeds' of Cockatoo Grass (*Alloteropsis semialata*) showing the intact spikelets (middle) and the actual seeds (caryopses) after extraction from the covering structures (top). The bottom spikelet has been opened to remove the caryopsis and the remains of the outer glumes and the inner lemmas covering the two florets can be seen. Opening or cutting the spikelet to check for a filled caryopsis is an easy initial test for checking the proportion of spikelets that contain viable seeds.](image1)

![Fig. 2. Cockatoo Grass (*Alloteropsis semialata*) (above) and Giant Spear Grass (*Heteropogon triticeus*) (below) flowering. The Cockatoo Grass flowers have orange and yellow stamens and purple feather-like stigmas. The Giant Spear Grass has purple stamens and long brown awns protrude from the apex of the inflorescence.](image2)
can be cut in half to visually inspect the caryopsis. Sometimes caryopses may be of normal size and appearance but still be dead. To test viability in this case, a sample of the caryopses can be treated with tetrazolium chloride, a dye that will turn red if the tissues of the caryopsis are metabolically active (Merritt 2006).

Poor seed viability can be due to site factors affecting the grass plant, such as adverse seasonal growing conditions, habitat features that do not suit the species, or damage by fungi or insects. Some species are genetically disposed to poor seed production (Jacobs 1973). When collecting the seeds, seed maturity and collecting techniques can affect viability. After collection, storage conditions (including temperature, humidity, fungi and insects) affect the rate of deterioration of seed quality (Merritt 2006).

We investigated 48 seed lots of 29 native grass species to determine whether seed quality was an issue for the establishment of native grasses in the Top End of the Northern Territory. The proportions of filled seeds, seeds of normal appearance and metabolically active seeds were assessed. Viability responses to seed storage in an air-conditioned room were investigated by repeated testing of 15 of the seed lots.

Materials and Methods

Seeds were mainly supplied by Greening Australia NT and by Kakadu Native Plant Supplies, with a list of desired species sent to their collectors. Seeds were collected in the Jabiru, Darwin and Katherine regions. Four seed lots (Alloteropsis semialata Lot 3; Eriachne ciliata Lot 3, Eriachne schultzeana Lot 2 and Thaumastochloa major Lot 1) were collected with assistance from Charles Darwin University (CDU) staff and/or students. Seeds were collected between April 2005 and May 2011. The date or month of collection provided by the seed collector or seed supply company and the date of testing were recorded.

Fig. 3. Seeds and associated structures of Giant Speargrass (*Heteropogon triticus*) (top left), Kangaroo Grass (*Themeda triandra*) (top right), Love Grass (*Eragrostis spartinoide*) (bottom left) and Wanderrrie grass (*Eriachne schultzeana*) (bottom right). The small brown seeds or caryopses extracted from the covering structures are shown for Love Grass and Wanderrrie Grass.
Stored seeds were accepted but we requested that new seed lots were sent to CDU as soon as possible after collection and cleaning of florets from vegetative material.

Most seed lots were tested within two months of arrival at CDU. Four of the 48 seed lots were not tested until 9–10 months after arrival and five seed lots were tested 14–16 months after being received (Aristida inaequiglumis Lot 2, Eulalia aurea, Heteropogon contortus, Sorghum plumosum, S. timorense). If sufficient seeds were available, seed lots were resampled and retested after one or two years of storage.

Seed purity refers to the weight of undamaged florets and caryopses as a proportion of the total weight of the seed lot. Seed purity was assessed by removing all vegetative material, chaff and obviously damaged florets from undamaged florets and caryopses for small samples. Larger seed lots were sub-sampled using halving techniques prior to assessing purity of four sub-samples and the purity result is the average of those four sub-samples.

Seed fill and cut tests were conducted using four replicates of 25 florets. Seed fill data denoted the percentage of florets that contained a caryopsis within them when the florets were opened. For the cut test, the caryopsis was removed from the floret and inspected under a dissecting microscope, where the percentage of florets with visually viable caryopses were counted. Unfilled florets, shrivelled, discoloured or damaged caryopses and caryopses that had a missing embryo were assessed as not viable. Possibly viable seeds that were only slightly smaller or slightly discoloured were counted as viable and included in the tetrazolium assessment below. Eragrostis spardinoides did not have seed fill of florets assessed as the seeds disperse as caryopses and don’t retain the outer floret structures.

For those seeds that were visually assessed to be viable or possibly viable, 2,3,5 triphenyl tetrazolium chloride (TTZ) was used to determine any metabolic activity. This colourless solution becomes red in response to metabolic activity in the tissue. Four replicates of 25 seeds were preconditioned by placing them in water at room temperature for 24 hours. The covering structures were removed or pierced away from the embryo to ensure water uptake without causing damage to the embryo. After imbibition, the seeds were cut through the embryo (or close to the embryo if cutting caused damage) except for Eragrostis spartinoides seeds, which were too small to cut. Half of each seed was then placed into 1% TTZ solution in a glass vial covered with aluminium foil to keep the incubating seed in darkness. The vials were placed in an incubator at 30°C for 24 hours, after which the seeds were removed and inspected under a dissecting microscope. Seeds with deep red-stained embryos and storage tissues were considered viable. Seeds that were unstained were not viable. Seeds with the embryo stained pale pink, or mottled staining of the storage tissues, were considered possibly viable. This resulted in a minimum and maximum proportion of viable seeds as assessed by TTZ.
Results

Seed viability of the grass species was variable, with Aristida inaequiglumis Lot 2 and Themeda triandra Lot 1 having less than 2–6% of viable florets, whereas Heteropogon triticeus Lot 1 and Eragrostis spartinoides had 79% and 90–100% viable seeds respectively (Table 1). Average viability of all grasses was moderate (37–42%).

Table 1. Viability of native grass seed lots. Each row is a separate seed lot. Age is the approximate time in weeks from collection to testing. Purity is the proportion by weight of caryopses plus florets that contain caryopses relative to the seed lot weight. Seed fill is the proportion of florets containing a caryopsis. Cut test is the proportion of florets containing a visually viable caryopsis. The last columns are the minimum and maximum viability of florets after tetrazolium (TTZ) testing as a proportion of florets (or caryopses if caryopses are shed from the florets).

<table>
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<tr>
<th>Species</th>
<th>Age (weeks)</th>
<th>Purity (%)</th>
<th>Seed fill (%)</th>
<th>Cut test (%)</th>
<th>TTZ min (%)</th>
<th>TTZ max (%)</th>
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Seed viability of native grasses

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<td>72</td>
<td>51</td>
<td>46</td>
<td>49</td>
</tr>
<tr>
<td>Curly Spinifex <em>Triodia bitextura</em> Lazarides</td>
<td>40</td>
<td>65</td>
<td>21</td>
<td>20</td>
<td>17</td>
<td>18</td>
</tr>
<tr>
<td>Mean of all seed lots</td>
<td>47.7</td>
<td>60.7</td>
<td>62.6</td>
<td>49.6</td>
<td>36.7</td>
<td>41.8</td>
</tr>
<tr>
<td>Minimum</td>
<td>4</td>
<td>8</td>
<td>10</td>
<td>6</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Maximum</td>
<td>144</td>
<td>100</td>
<td>97</td>
<td>100</td>
<td>90</td>
<td>100</td>
</tr>
</tbody>
</table>

Seed purity was highly variable, ranging from 8–100%, but purity is dependent on the level of cleaning. The *Fragrostis* sp. seed batch had only 8% pure seeds but the seeds were tiny and numerous – the 17.8g seed lot still contained 24,200 seeds. Seed purity can also be highly variable between seed lots of a species; *Heteropogon triticeus* Lot 1 contained 100% pure seed whereas Lot 2 contained only 27% pure seeds.
Once chaff, vegetative material and damaged florets had been discarded, seed viability was largely dependent on the seed fill of the florets. On average, 37% of florets did not contain seeds. However, the proportion of filled seeds could be much lower – 90% of the florets of *Themeda triandra* Lot 1 did not contain caryopses and 79% of *Triodia bitextura* florets were empty. In contrast, other seed lots contained a high proportion of filled seeds – 72% of florets of *Themeda triandra* Lot 3 and 97% of the florets of *Aristida bolatbera* contained caryopses.

Closer inspection of the seeds was important for assessing viability as some filled seeds were not viable. Across all the seed lots, 62.6% of florets contained a caryopsis but only 49.6% of the caryopses were of normal appearance and contained an undamaged embryo. For the two seed lots of *Eriachne burkittii*, closer visual inspection determined that less than half of the filled florets contained viable caryopses. In contrast for the *Eriachne schultziana* florets, almost all of caryopses present appeared viable after microscopic inspection.

Some caryopses that appeared normal and viable when visually inspected were not viable as they were not metabolically active when tested with TTZ. For some species, such as *Eriachne schultziana*, visual inspection substantially overestimated the number of viable seeds, with 81% of seeds appearing viable in Lot 1 when inspected, but only 24% having any metabolic activity. For other species, such as *Cymbopogon bombycinus*, all the seeds that visually appeared to be viable were also metabolically active.

For the seed lots that were retested after at least one year of storage in the CDU laboratory, the decline in viability averaged 6% per year or a loss of 12% of viable seeds per year (Fig. 4). For some species, such as *Eriachne schultziana*, there was substantial reduction in viability after 1–2 years but for others, such as *Eragrostis spartinoides*, there was little decline in viability. The *Eragrostis spartinoides* seed lot investigated in this study maintained viability and germination levels after 4.5 years storage and hence some native grass seed lots are able to be stored for quite some time. *Eriachne obtusa* and *Chrysopogon fallax* seed lots were able to be stored for two years with similar levels of viability to the initial assessments, however, after 3–4 years storage viability levels were low. *Eriachne schultziana* and *Heteropogon triticeus* were able to be stored for 1–1.5 years, however, after further storage viability levels were low. *Alloteropsis semialata* seeds lost viability after 2 years storage with very low viability after 3 years.

**Discussion**

Generally, native grass seed lots newly received from seed suppliers in the Darwin region of the Northern Territory had reasonable levels of seed viability, similar to those for Australian native grass seed lots generally. Viability of four native grasses used for minesite rehabilitation in Western Australia was generally lower, ranging from 19–39% (Dixon 1997) and three seed lots of *Themeda australis* from New South Wales ranged from 52–68% (Nolan et al. 1997). Farley *et al.* (2013) assessed viability of 15 native grass
Fig. 4. Changes in grass seed viability over time (age since collection) of seed lots stored in sealed containers in an air conditioned room. Values are maximum viability of filled seeds that stained with tetrazolium chloride, a dye that turns tissue red if it is metabolically active. Where more than one seed lot was tested, the seed lot number is after the species name. Black: less than 1 year old; grey: 1–2 years old; white: 2 years old.

species that were hand collected and received at the seed laboratory within seven days of collection. Two species had higher than 80% viability and three species had less than 30% viability. The studies above all had viability tested using TTZ similarly to this study.

Commercial seed lots of exotic grasses used for sowing pastures and revegetation, which have been bred to have high seed production, often exceed 90% pure seeds (McCormick et al. 2009) and viability may exceed 85–90% (Cole and Johnston 2006; McKays Grass Seeds 2014; Grass Seed Online 2014) depending on the species. Tropical perennial grass cultivars from northern NSW purity ranged from 68–95% (Lodge and McCormick 2010). As purity and viability is lower for native grasses it is important to take this into account when determining seed application rates. To achieve the same number of
viable seeds as a seed lot with 90% purity and 90% viability, native grass seed lots with an average of 60% purity and 40% viability need a 3.375 times higher seed application rate (assuming seed sizes are the same). There is a tendency to apply less native grass seed because it is more expensive, but to achieve the same viable seed application rate, relatively more seeds need to be applied.

Purity of native grass seed can be increased, but this increases the cost of the seeds. Some seeds are also likely to be lost when carrying out further cleaning and sieving processes undertaken to increase purity. Another issue for cleaning the seeds of some species, is that structures such as long awns are damaged or lost and these may be important to help the seed orient itself correctly in the soil (Loch et al. 2004). Often the most economic option for native grasses is to apply seeds with a relatively high proportion of chaff rather than cleaning the seed lot to a high purity. It is important though, that the seed lot does not contain undesirable species, or insects or pathogens that will cause deterioration of the seed lot. In addition, for long-term storage, reducing the volume of non-seed material is important to reduce storage space.

Viability varies considerably between species and between seed lots of the same species. It is affected by the characteristics of the species, the seasonal growing conditions, the habitat where the plants were growing, maturity when harvested and by seed pathogens and granivores before or after storage (Merritt 2006). Wells et al. (2000) found that the optimum harvest window for Triodia spp. in the Kimberley region is just 5–7 days and if harvested before or after this, a substantial reduction in viability occurs. They also found a much higher quantity and quality of seed produced in more favourable locations and seasons. To control these factors, it is desirable to either test seed lots or use several seed lots collected from different sites.

Some native grass species have very broad ranges of viability but it is important that grass seed lots are regionally sourced. Obtaining seeds sourced from plants in the region is important to ensure the establishing plants are adapted to conditions that occur in the area where they are sown. For example, a trial in Brisbane investigated the performance of a seed lot of Heteropogon contortus sourced from Victoria compared to several seed lots sourced from Queensland. Plants of local and regional provenance seed lots grew and flowered whereas the seeds from Victoria produced robust plants that never flowered. The Victorian sourced plants likely required specific temperature and day length conditions to trigger flowering that did not occur in Brisbane (SMB unpubl.). Similar provenance effects on flowering are observed in Themeda triandra (Evans and Knox 1969).

Seed fill is an important first indication of seed quality. It can be tested when the seed lot is being collected by checking a selection of florets for filled seeds, either by pressing the side of the floret or by cutting them open to observe the caryopsis. If the grasses at a site have poor seed fill then collection can shift to a site with higher seed fill. The species characteristics also need to be considered. In the Triodia spp. studied by Jacobs (1973)
for example, seed production was restricted to just one or two seeds per spikelet with each spikelet containing six to eight florets. Even if grown in a glasshouse with sufficient water and nutrients, a maximum of three seeds per eight or more florets were produced, so even under optimum conditions many florets in these *Triodia* species remain empty.

The cut test could also be carried out in the field using a hand lens for species with larger seeds, but it requires familiarity with the grass seed to detect filled but abnormal seeds. In contrast, the tetrazolium test requires more time (two days), specific facilities and expertise, therefore is generally carried out by commercial seed testing laboratories. The cost can be a deterrent, but for larger revegetation projects the test is worthwhile when compared to the costs associated with repeating sowing activities due to inadequate amounts of viable grass seeds.

For larger projects, seed collection over several years and storage of seed lots is an option for obtaining sufficient seeds. The seeds used in this study were dried and stored in cool dry conditions in an air conditioned laboratory. Keeping the seed lots cool and dry is important as increasing the temperature by 5°C or increasing seed water content by 1% can double the rate at which seed viability is lost (Merritt 2006). Keeping tropical grass seeds at 30°C or higher temperatures and exposed to humid conditions greatly increases seed mortality (McIvor & Reid 2011) and could result in death of all seeds of some species within a year. Under low humidity conditions in a room that was air conditioned in summer, Silcock *et al.* (1990) in Charleville, Queensland, found 15 of the 20 native grass species tested could have seeds stored for at least three years with little deterioration. Twelve of the species could be stored for more than five years.

Native grasses are important for the structural integrity of native vegetation communities in the Northern Territory and it is important that they are included – along with trees, shrubs and forbs – when restoring habitat for wildlife. Many bird species are granivorous and dependent on native grass seeds. However, if native grasses are to be established in revegetation, it is necessary to use sufficient seeds of high quality and these seeds need to be stored to preserve viability. Seeding rates need to take purity, seed fill and viability into account. Some simple tests will give an indication of viability, but for large projects laboratory testing of viability using tetrazolium or germination testing is desirable.

**Acknowledgements**

This research was funded by Energy Resources of Australia Ltd, Environmental Research Institute of the Supervising Scientist and Northern Territory Department of Mines and Energy. Technical assistance was provided by Ms Julie Crawford and two seed lots were assessed by CDU student Ms Kathryn Sangster. The project support provided by Dr P. Bayliss, Mr P. Christophersen, Ms S. McGregor, Mr S. Crowder, Ms B. Saggars, Dr C. Humphrey, Dr M. Daws, Ms M. King, Mr A. Speechly, Mr J. Kepui and Mr P. Hickey is greatly appreciated.
References


Nest site fidelity of Flatback Turtles (Natator depressus) on Bare Sand Island, Northern Territory, Australia

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Abstract

The endangered Flatback Turtle (Natator depressus) is endemic to the continental shelf of northern Australia and is the only species of marine turtle with such a restricted geographical distribution. Most mature female Flatback Turtles show a high degree of fidelity to their chosen nesting beach, returning to the same beach within the same and successive nesting seasons. Natal homing has been well studied in other species of marine turtles and our findings support the supposition that all marine turdes display a similar degree of natal homing. Our study area is Bare Sand Island, Northern Territory, where we investigated nest site fidelity of female Flatback Turtles and the influence of wind speed, air and sand temperature, and relative humidity on nest site selection. The data were collected during a 46-day period from 12 June 2012. On Bare Sand Island, female Flatback Turtles demonstrate very strong nest site fidelity, with consecutive nests being located 247 m ± 198 s.d. apart. During the peak 2012 breeding season, sand temperatures, wind speed and relative humidity remained constant, however there was a significant difference in the air temperature between nesting days. Our study of the effects of environmental factors on the nesting environment of Flatback Turtles will contribute towards management practices to protect this endangered species.

Introduction

The range of the Flatback Turtle (Natator depressus) is limited to the continental shelf of northern Australia and its distribution is the most geographically restricted of all marine turtle species (Limpus 2009). Unlike other marine turtles, Flatsbacks lack an oceanic phase in their life cycle and remain in the surface waters of the continental shelf (Walker & Parmenter 1990). They are identified by four pairs of costal scales on a low-domed carapace and a pair of prefrontal scales on the head. The carapace of the hatchling measures approx. 6 cm and weighs approx. 43 g. At maturity, the adult female Flatback Turtle is olive grey, has an average carapace length of 92 cm and weighs around 90 kg (Limpus 2009). Flatback Turtles come ashore to lay their eggs on remote beaches along the tropical and subtropical northern Australian coastline (Whiting et al. 2008).

As a consequence of the destruction of nesting habitats and fishing bycatch, the Environmental Protection of Biodiversity and Conservation (EPBC) Act (1999) has classified
the Flatback Turtle as Endangered. The Flatback is the only marine turtle that the International Union for Conservation of Nature lists globally as Data Deficient (IUCN 2010). Marine turtles are becoming increasingly threatened as a result of commercial fishing bycatch, boat strike, illegal harvesting of eggs and adults, and an increase in nesting habitat destruction (Limpus 1995; Whiting & Guinea 2003). Of all the species of marine turtles, the Flatback is by far the most under-researched and under-reported (Pendolcy et al. 2014).

It has been suggested that the breeding site selection of marine turtles may be attributed to individual animals imprinting on magnetic fields in their natal area and then, years later, using this information to return to their natal site (Hueter 1998; Lohmann et al. 2008). However, uncertainty remains as to whether their homing is attributable to imprinting to the natal beach as a hatchling, or imprinting to the region of their birth and then the specific beach as an adult during their first breeding season (Limpus 2009).

Genetic analyses show that the precision of natal homing can vary considerably among different populations and species with homing to regions of coastline measuring several hundred kilometres being common (Lohmann et al. 2008). Female Flatback Turtles generally lay three to four clutches of eggs each nesting season with an inter-nesting interval of approx. 14 days (Hewavisenthi & Parmenter, 2002). Mature female Flatback Turtles show a high degree of fidelity to their nesting place, returning to the same beach to lay consecutive clutches within a nesting season and in successive nesting seasons (Limpus 2009; Oates 2010; Matos et al. 2012).

The environment of the nest site is important for marine turtles because it influences offspring sex, embryonic survival, hatchling development rates and hatchling size, mass and energy reserves (Hewavisenthi & Parmenter 2002; Koeh et al. 2007). To date, however, there is little understanding of the physical factors that contribute to the distribution of turtle nesting (Santana Garcon et al. 2010). What is known, is that sex determination is temperature-dependent; higher temperatures produce females, whilst lower temperatures result in male hatchlings (Santana Garcon et al. 2010; Hewavisenthi & Parmenter 2002).

The northern tropical Australian populations of Flatback Turtles have a protracted nesting period of around nine months of the year, reaching a peak in July (Whiting et al. 2008). Several studies have attempted to examine the influence of beach characteristics on nesting cycles (Parmenter & Limpus 1995; Hewavisenthi & Parmenter 2002; Koeh et al. 2007; Whiting et al. 2008). In this study we investigated nest site fidelity of female Flatback Turtles on Bare Sand Island within a single breeding season to assess the influence of environmental factors on nest site selection. The study site was free of introduced predatory species and human interference was minimal. The environmental factors considered were wind strength, relative humidity, and air and sand temperatures.
Nests of Flatback Turtles
Northern Territory Naturalist (2016) 27

Fig. 1. Location of nest sites where female Flatback Turtles laid more than once during the 2012 peak breeding season on Bare Sand Island, Northern Territory. (Courtesy Digital Globe 2012)

Materials and Methods

Our study site is Bare Sand Island (12°32.39'S, 130°25.02'E), which is 50 km west of Darwin, Northern Territory. It is located towards the end of a chain of eight islands (Whiting & Guinea 2003; Koch & Guinea 2006). The main nesting beach faces west and is composed of fine sand with a gentle rise, making the nesting beach easily accessible from the ocean (Koch & Guinea 2006). The data were collected during a six and a half week period from 12 June 2012.

Two-hour foot patrols either side of the evening high tide were undertaken to coincide with the turtles’ main nesting activity. Turtles were identified by their tags or were tagged on-site and the nest location was recorded using Garmin GPSmap 60CSx, or Garmin GPS72, or Garmin GPS72H. GPS points, and their corresponding tags, were uploaded into ESRI ArcMap 10.1 (© 2012, ESRI, Redlands, California) and a Microsoft® transposed onto a Google Earth Image Landsat© 2012 Google using the Landsat 7 Satellite (see Fig. 1). The image was geo-referenced against known latitude and longitude co-ordinates and projected into the WGS 84 Global Positioning System with our accuracy of approximately 5 m. A Hobo™ water temp Pro v2 data logger was buried in the sand on top of a dune at the same depth as that of a Flatback Turtle’s nest (50 cm) and another was positioned in the middle of the western beach. The sand temperature was recorded every half hour and air temperatures and wind speeds were recorded daily at 18.45 hr using a hand-held Air™ speed temperature meter (Dick Smith QF301). The meteorological data were compared with readings from the Australian Bureau of Meteorology weather station (AWS Station 014277) located on Dum In Mirrie Island (13 km south of Bare Sand Island) and we were satisfied that the Meteorological Station data were suitable for our study purposes.
For each female, the distance between consecutive nests during the 2012 season was calculated using ArcMap on-board measuring toolsets. These distances were compared with those from previous nest sites to an equal number of randomly generated potential nest sites and a two sample t-test was used to determine whether the two samples were significantly different. To determine whether there were differences between environmental conditions, paired t-tests were used to compare the environmental data from the different days when female Flatbacks nested. Both the paired and t-tests were analysed using alpha values of 0.05. All the data were analysed using Microsoft Excel 2010 (© 2010 Microsoft Corporation, Redmond, Washington, U.S.A) and Minitab 15 (© 2007 Minitab Inc., State College, Pennsylvania, U.S.A).

Results

Fifty-four female Flatback Turtles were observed coming onto Bare Sand Island to nest more than once. Of these, four returned three times, two nested four times and the rest nested twice. The mean inter-nesting period was 19 days ± 1.58 s.d. with the minimum and maximum inter-nesting periods being 15 days and 37 days, respectively. The locations of nest sites for females that nested more than once on Bare Sand Island are shown in Fig. 1.

The mean individual inter-nesting distance was 247 m ± 198 s.d. with a minimum of 7 and a maximum 687 m. We found that more than 50% of the females laid their second nest within 250 metres of their initial nesting site and that they were non-randomly selected (t = -4.79; P = 0.000; d.f. = 108). The mean distance between two nest sites of the same female was 247 m ± 198 s.d., whereas the mean distance between randomly selected sites is 456 m ± 277 s.d.. The mean air temperature on Bare Sand Island was 24.35°C ± 1.25 s.d. and was not significantly different from the meteorological recordings on Dum In Mirrie Island (t = -0.89; P = 0.376; d.f. = 52). The same was true for wind speeds, with a Bare Sand Island mean of 10.09 km/h ± 5.0 s.d. (t = 0.36; P = 0.718; d.f. = 42).

The mean wind speed was 9.75 km/h ± 2.60 s.d. and the mean relative humidity was 52.1% ± 19.0. The effects of four environmental elements on nesting behaviour were studied; namely, air and sand temperatures, wind speed and relative humidity. The mean values of the four sets of data were recorded on days when the same female turtle nested and were compared to determine whether they differed significantly (see Table 2). Using paired t-tests, we found no significant difference between the sand temperature, wind speed or relative humidity on different nesting days, however there was a difference in air temperatures.

Table 2. Comparison of environmental factors influencing nest site conditions of the same female turtle on consecutive nesting times.

<table>
<thead>
<tr>
<th></th>
<th>Mean Nest 1</th>
<th>Mean Nest 2</th>
<th>t- Value</th>
<th>P- Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sand temperature</td>
<td>28.16 ± 0.46 °C</td>
<td>28.17 ± 0.48 °C</td>
<td>-0.06</td>
<td>0.950</td>
</tr>
<tr>
<td>Wind speed</td>
<td>9.98 ± 2.55 km/h</td>
<td>10.23 ± 2.48 km/h</td>
<td>-0.52</td>
<td>0.608</td>
</tr>
<tr>
<td>Relative humidity</td>
<td>43.07 ± 15.50%</td>
<td>45.25 ± 18.07%</td>
<td>-0.69</td>
<td>0.494</td>
</tr>
<tr>
<td>Air temperature</td>
<td>24.72 ± 1.41 °C</td>
<td>25.47 ± 1.44 °C</td>
<td>-2.57</td>
<td>0.013</td>
</tr>
</tbody>
</table>
Discussion

The number of Flatback Turtles that returned to Bare Sand Island to nest during the 2012 breeding season was lower than in previous years – a fact that may be attributed to it being the coldest July in 35 years (Australian Government Bureau of Meteorology 2012). The nesting rates of the turtles observed in our study were generally less than the 3–4 clutches expected for Flatback Turtles during a nesting season (Hewavisenthi & Parmenter 2002), however, our data collection period was limited to the peak nesting period. The inter-nesting period we recorded was 19 days ± 1.58 s.d. This is longer than the mean 15-day inter-nesting interval that usually occurs with Flatback Turtles within the same season (Limpus et al. 1984; Hewavisenthi & Parmenter 2002). Again, this might be attributed to the cooler water temperatures that are known to reduce the rate of pre-ovipositional development of eggs during inter-nesting for Loggerhead, Olive Ridley and Green Turtles (Sato et al. 1998; Hays et al. 2002; Matos et al. 2012).

From Figure 1 it can be seen that the study animals preferentially nested on the western beach on Bare Sand Island. This is consistent with previous years and is attributed to the fine sand and the gently sloping beach that faces the open ocean (Koch & Guinea 2006). Although it is not entirely clear why some beaches are preferentially selected by sea turtles to deposit eggs, a number of factors have been identified. The beach must be easily accessible from the ocean, be high enough to avoid inundation at high tide and have temperatures conducive to egg development (Miller et al. 2003). The south-easterly section of the island is exposed to strong trade winds throughout the nesting season which explains its low nest density (Koch & Guinea 2006; Koch 2007).

The high degree of nest site fidelity observed in our study animals agrees with the similar findings of Limpus (2009), Oates (2010) and Matos et al. (2012). Our study suggests that Flatback eggs may be more tolerant of higher incubation temperatures than those of most other sea turtle species; a finding that is supported by the work of Hewavisenthi & Parmenter (2002). This change in air temperature may have contributed to the females’ nest site choice, as the nesting phase of the marine turtles’ reproductive cycle is thought to be largely determined by temperature (Santana Garcon et al. 2010). Higher temperatures will reduce the turtles’ progress across a beach to the ocean. However, as they typically emerge at night, their movements are not hindered by high temperatures (Koch et al. 2008). The mean inter-nesting distance of 247 ± 198 m for Flatback Turtles differs markedly from Olive Ridley Turtles (4.83 ± 4.37 km) and Green Turtles (0–5 km) (Matos et al. 2012; Lalith Ekanayake et al. 2003).

Sand temperature, however, plays a vital role in the development of turtles and influences hatchling size, sex, and energy reserves and successful incubation is only possible within certain thermal limits. Nest temperatures are variable not only on a single beach within a season and at different levels on the shore, but also vary with depth at a single nest site (deeper eggs are incubated at rather lower, more stable temperatures). The influence of short periods of extreme temperature is unclear. However, it has been reported that
the final third of the incubation period is particularly temperature-sensitive and eggs rarely hatch if exposed to temperatures below 23°C or above 33°C (Davenport 1997). Flatback Turtles appear to be more tolerant of high incubation temperatures and severe moisture stress than most marine turtle species (Hewavisenthi & Parmenter, 2002).

Sand temperature influences the timing of the emergence of Flatback hatchlings. Most left the nest during the same few hours each night because of thermal cues that are dependent upon a combination of threshold temperatures, thermal gradients in the nest, and rates of temperature change (Davenport 1997; Koch et al. 2008). As sand temperatures at the study site remained below 29.3°C (the pivotal temperature) and as sex determination is temperature-dependent, predominantly male hatchlings emerged (Koch et al. 2007).

Strong south-easterly seasonal trade winds have persisted throughout the nesting season on Bare Sand Island over the last 10 years and, while this has the potential to affect the depth of the nests, they were not reported to have been shallower than 20 cm and were not subsequently threatened by the high temperatures that occur late in the nesting season (Koch & Guinea 2006; Koch et al. 2007). We found no significant difference between the mean relative humidity (52.1% ± 19.0 s.d.) on successive nesting days. Favourable conditions for embryonic development and survival include high humidity influencing hatchling size, sex, and energy reserves (Hewavisenthi & Parmenter 2002; Miller et al. 2003).

References


Records of waterbirds and other water-associated birds from the 2014/15 migratory season in the Darwin region

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Abstract

Records of waterbirds, waterfowl, terns, gulls, egrets and herons, raptors, and resident shorebirds in the Darwin region, Northern Territory, were collected during fortnightly migratory shorebird monitoring. Eight study sites were monitored from August 2014 through to April 2015, which is considered the migratory season for most non-passerine birds in the Top End. Species abundance across the sites, breeding records, and new information on habitat use at an artificial habitat (East Arm Wharf) are presented. Across the eight study sites there were 39 species recorded, representing 15 taxonomic families.

Darwin Harbour in the Northern Territory has a rich coastal waterbird assemblage, owing to its diverse range of habitats. The coastal region supports resident and nomadic Australian waterbirds, waterfowl, resident and migratory terns and gulls and various raptors that inhabit coastlines (McCric & Watson 2003). A number of terns that breed in the northern hemisphere visit northern Australian coastlines during the austral summer where they feed over the ocean and along tide lines and then roost at beaches, rocky reefs, dykes and on floating buoys. The macro-tidal nature of tides in the Darwin region creates extensive mud and sand flats, available for foraging birds. Mangroves, saltpans and saltmarsh provide roosts during high tides.

Studies of waterbirds in the Top End have mainly focused on freshwater wetlands and floodplains in the Fogg Dam and Alligator River regions, east of Darwin (see Crawford 1979; Morton et al. 1993). The waterbirds in Darwin Harbour prefer coastal saline habitats, including fringing mangroves, brackish waste water ponds and dredge ponds nearby, and creeks and rivers. Extensive aerial and ground surveys along the Northern Territory coastline indicate that the region supports a variety of waterbirds (Chatto 2006).

During regular monitoring of migratory shorebirds I collected count data for all birds across eight study sites from August 2014 through to April 2015, which is when most migratory shorebirds and other water-associated migrant birds visit Australian shores. Birds were surveyed at each site most fortnights during spring tides, which were selected
to target when migratory shorebirds would be roosting. There were 184 surveys performed over the nine survey months. The sites were East Arm Wharf, Lee Point-Buffalo Creek, Ludmilla Bay, Spot On Marine boat yard, Nightcliff Rocks, East Point, Sandy Creek and East Arm Wharf Railway Mud, all within the Darwin region. This note summarises the results of all birds present at the study sites, excluding migratory shorebirds. Thirty-nine species of birds were recorded within the study period, including 5 species of heron and egret, 2 gull, 8 tern, 3 raptor, 8 resident shorebird, 8 waterbird and 5 waterfowl species. The maximum count for each species and the corresponding site and date are shown in Table 1.

### Table 1. Results from waterbird monitoring in the Darwin region from August 2014–April 2015. Bird species are grouped and presented in taxonomic order following Christidis & Boles (2008).

<table>
<thead>
<tr>
<th>Family and grouping</th>
<th>Common name</th>
<th>Scientific name</th>
<th>Max. count</th>
<th>Site of max. count</th>
<th>Site co-ordinates</th>
<th>Date of max. count</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Waterfowl</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anatidae</td>
<td>Wandering Whistling Duck</td>
<td>Dendrocygna arcuata</td>
<td>149</td>
<td>East Arm Wharf</td>
<td>12.5325°S, 131.0639°E</td>
<td>4 Jan 2015</td>
</tr>
<tr>
<td>Anatidae</td>
<td>Radjah Shelduck</td>
<td>Tadorna radjah</td>
<td>200</td>
<td>Lee Point Buffalo Creek</td>
<td>12.3453°S, 130.9825°E</td>
<td>21 Nov 2014</td>
</tr>
<tr>
<td>Anatidae</td>
<td>Pacific Black Duck</td>
<td>Anas superciliosa</td>
<td>17</td>
<td>East Arm Wharf</td>
<td>12.5325°S, 131.0639°E</td>
<td>23 Dec 2014</td>
</tr>
<tr>
<td>Anatidae</td>
<td>Hardhead</td>
<td>Aythya australis</td>
<td>12</td>
<td>East Arm Wharf</td>
<td>12.5325°S, 131.0639°E</td>
<td>6 Apr 2015</td>
</tr>
<tr>
<td>Podicipedidae</td>
<td>Australasian Grebe</td>
<td>Tachybaptus novaehollandiae</td>
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Two subspecies of Gull-billed Tern occur in northern Australia, *affinis* being a migrant that visits Australia during the summer season. These subspecies can be separated in the field using morphological features; see Lilleyman and Hensen (2014).

East Arm Wharf, an artificial site made up of dredge ponds, situated within Darwin Harbour, consistently supported the most species of all the sites. The site attracts a diverse range of species because the ponds represent a mixture of freshwater and
marine/saline habitats, with input from the harbour. The ponds are in open terrain with good visibility for birds to detect predators, and situated next to the coastline. The site is also protected from human disturbance as public access is restricted, and the site excludes feral terrestrial predators like dogs and cats through trapping and fencing. East Arm Wharf supported the most species (18) of water-associated bird compared to the other sites during the monitoring period. Sixteen species were recorded at the

Fig. 1. Franklin’s Gull (right) and Silver Gull (left) in a dredge pond at East Arm Wharf in Darwin, 20 April 2015. (Amanda Lilleyman)

East Arm Wharf Railway Mud (adjacent to the dredge ponds at East Arm Wharf), but the assemblages between these two close sites varied. Twelve species of waterbird were recorded at Lee Point-Buffalo Creek during the monitoring period.

Across the sites, the month of November had the highest total count of birds, mostly weighted by terns, followed by March and then December, both weighted by gulls and terns. A vagrant gull, Franklin’s Gull (L. pipixcan) (Fig. 1), distinguished from the more common Silver Gull (C. novaehollandiae) by its black head markings or prominent hood and dark grey back and upperwings contrasting with white underparts, was recorded in March, initially at Buffalo Creek, and subsequently at Stokes Hill Wharf (Mark de Kretser, pers. comm. 18 April 2015), and lastly at East Arm Wharf (by AL). This species breeds in North America and spends the non-breeding season in South America (Handbook of the Birds of the World Alive 2015). This is the 19th record for Australia and the second time the species has been recorded in the Northern Territory (BirdLife Australia 2015). The first arrival and last departure records for migratory terns and one vagrant gull are shown in Table 2.
Crawford (1980) reported mean counts for Whiskered Terns (*Chlidonias hybrida*) with peaks in September at Fogg Dam and in Darwin (100 and 50 individuals, respectively) and Lesser Crested Terns (*Thalasseus bengalensis*) (40 individuals) along the coastline of Darwin, but noted in January for the highest mean counts of Crested Terns (*Thalasseus bergii*) (100 individuals). The maximum count of Crested Terns from the current study was 192 individuals in November from Nightcliff, which was a site not surveyed in the Crawford (1980) paper. The maximum count of Whiskered Terns from the current study is certainly an increase from the mean counts recorded by (Crawford 1980).

Outside the monitoring period, in June, July and August there were up to 10 Red-necked Avocets (*Recurvirostra novaehollandiae*) using the freshwater dredge ponds at East Arm Wharf. Records from eBird and a local online forum (NT Birds Yahoo group) show this species is recorded in the Top End every year or so, but mostly further south and east of Darwin city (i.e. South Alligator River, Mamukala, Shark Billabong, Adelaide River). This record at East Arm Wharf is the first record close to Darwin since 20 August 2013, when the species was recorded at Leanyer Sewage Treatment Ponds. Resident shorebirds were recorded nesting and raising young at East Arm Wharf, including Black-winged Stilts (*Himantopus himantopus*), with a maximum count of 74 individuals in April, after the nesting period. Red-capped Plovers (*Charadrius ruficapillus*), Masked Lapwings (*Vanellus

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<tr>
<td>Franklin's Gull</td>
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<td>April</td>
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Fig 2. Red-necked Avocets in a dredge pond at East Arm Wharf in Darwin, 19 July 2015. (Amanda Lilleyman)
miles) and Pied Oystercatchers (Haematopus longirostris) were also recorded breeding along the muddy edge of one of the dredge ponds. Red-capped Plover also regularly breeds along the sandy beach at Lee Point.

Other localities around Darwin provide quality habitat for waterbirds, shorebirds and other water-associated birds, including Holmes Jungle, Knuckey Lagoon, McMinns Lagoon, Leanyer and Palmerston Sewage Treatment Ponds. These sites were not surveyed in the study period as they were not included in the migratory shorebird monitoring program; however, future monitoring of these sites would improve our knowledge of birds in the region.

The Darwin region coastline and associated freshwater ponds support a diverse range of water-associated bird species and high abundances throughout the austral summer season. Of the sites surveyed for this study, East Arm Wharf is the most important site (based on species diversity and number of individuals recorded) for a range of waterbirds, terns and gulls, waterfowl and breeding resident shorebirds.

Acknowledgements

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References


Fluctuations in use of urban roost and foraging sites in Darwin by Pied Herons (*Ardea picata*)

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Abstract

Pied Herons (*Ardea picata*) are a common bird of Australia's north, strongly associated with shallow freshwater wetland and estuarine habitats. However they also use urban sites for foraging, and in particular many feed at the Leanyer Sewage Treatment Works and the Shoal Bay Waste Depot site in Darwin's northern suburbs. Pied Herons roosted on Catalina Island in Darwin Harbour from at least 2010 to 2013. Seasonal fluctuations in numbers and use of that, and nearby roost sites, are documented here, as is the more recent abandonment of harbour roost sites in favour of constructed riverside habitat at Crocodylus Park, close to the foraging sites.

Introduction

Pied Herons (*Ardea picata*) (Fig. 1) are a common bird of Australia’s north, associated with shallow freshwater wetland and estuarine habitats (McKilligan 2005). However they

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Fig. 1. An adult Pied Heron (*Ardea picata*) at Knuckey Lagoon, Darwin, showing nuptial plumes on the head just prior to the wet season. (John Rawsthorne)
also use urban sites for foraging, and in particular a large number arrive each day to feed at Leanyer Sewage Treatment Works and the Shoal Bay Waste Depot site in Darwin’s northern suburbs (McCrie & Noske 2015; JR pers. obs.).

I was initially intrigued by large early morning flocks flying north over Darwin Railway Station at East Arm in 2010, and a suggestion by Richard Noske that the north-south daily movements were a well-established pattern by then. Further observations of evening flocks flying south over Kormilda College, Berrimah, during 2012 and a chance observation of Pied Herons arriving from the north at dusk at Catalina Island in East Arm, Darwin Harbour were the catalyst for a more formal investigation of these movement patterns.

Here I document roosting places of these urban birds, their flight path – including potential conflict with Darwin Airport flight paths – and seasonal fluctuations in numbers.

Methods

Bird activity sites

Catalina Island (12.4900°S, 130.9070°E) is a small (50 x 200 m, 1 ha) island in East Arm, Darwin Harbour. In addition to terrestrial trees including one tall Peanut Tree (*Sterculia quadrifolia*) (Richard Willan pers. comm.), it has fringing mangroves including Grey Mangrove (*Avicennia marina*) and Mangrove Apple (*Sonneratia alba*), which approximately double the vegetated area to around 2 ha. At low tide, a sand spit and rocky areas are exposed around the island.

Shoal Bay Waste Depot and the Leanyer Sewage Treatment Works are on the north-east fringe of suburban Darwin, about 12 km and 14 km, respectively, north of Catalina Island. Crocodylus Park is close to the waste depot and sewage works, about 3 km south of the waste depot.

Evening roost surveys

Pied Herons were counted flying from the north to evening roosts in East Arm (Fig. 2) monthly from November 2012 to October 2013. Counts were performed from either East Arm Boat Ramp (about 0.7 km north-east of Catalina Island) or Berrimah Road near the Vopak Fuel Depot (about 1.5 km north of Catalina Island), depending on wind conditions and roost site being used. In September 2015 a follow-up count was performed at Crocodylus Park.

Birds generally arrived to roost in flocks, mostly of less than 100 birds, but occasionally in much larger flocks of several hundred individuals. Counts were of individual birds where flocks were less than approx. 50 in number, but for larger flocks or for those in quick succession, estimates of the number of birds were made based on ‘blocking up’ from smaller counts.
Results

In November 2012 several preliminary counts and tests of counts were made (4357 individuals on 1 November and 5895 individuals on 8 November). These counts were treated as training exercises to refine the counting procedure. Some tests were made of my counts of larger flocks against photographs of those same flocks, confirming the general accuracy of my counts. On 22 November, two independent counts were made by Gavin O’Brien and me, producing counts of 5138 individuals (GO’B) and 4953 individuals (JR) arriving to roost on that evening. These tests confirmed the broad accuracy of the counts, and suggested that they might be regarded as being ± 5%, rather than precise counts of individuals. In the data that follow, I have used the average of the two 22 November counts (5045 birds) as the data point for November 2012.

The number of birds counted in individual surveys (Fig. 3) ranged from a high of 5895 individuals (preliminary count, November 2012) to a low of 1705 individuals (February 2013). The average number of birds by count across all months in 2012/13 was 2771 individuals.

Roost counts were conducted for 12 months, from November 2012 to October 2013. In the first few months of the survey, birds exclusively flew to Catalina Island to roost, and were easily counted flying directly overhead of the East Arm Boat Ramp (Fig. 2). In later months birds also sometimes appeared to roost on South Shell Island, a smaller treeless island about 2 km west-south-west of Catalina Island, while in some evening
surveys birds flew further west beyond the islands in East Arm, and at least as far as the mangroves fringing Wickham Point.

Follow-up observations in September 2015 indicated that Pied Herons had ceased to roost in East Arm, and were now roosting much closer to the sewage ponds and waste depot, at an artificial river at Crocodylus Park on the corner of Vanderlin Drive and McMillans Road. Construction of this habitat commenced in 2007 and was finalised in May 2014. A one-off count on 28 September 2015 counted approximately 3300 Pied Herons roosting at this site.

**Flight paths**

All birds arriving to evening roosts arrived from the north. Observations of mid-flight paths for flocks indicated a quite direct flight path between the Shoal Bay Waste Depot and East Arm, with no intermediate stops and no birds coming on to the flight path any further south than about McMillans Road (Fig. 2). Evening and morning observations in the northern suburbs did not detect birds arriving to or leaving from the northern sites in any direction apart from the south. I did not conduct any night-time surveys at the day time feeding sites.

The actual flight paths used by commuting Pied Herons were within about 1 km of Berrimah Road. Depending on the wind direction, birds deviated in their evening southward flight slightly east or west of Berrimah Road, but corrected their path as they came closer to roost. The flying height of birds was measured using binocular focus distances. I focused on birds flying directly overhead, and then measured the distance along the ground to the same focus distance. Birds typically flew at a height of about

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**Fig. 3.** Monthly count of Pied Herons (*Ardea picata*) arriving from the north to roost in Darwin Harbour, November 2012–October 2013. There is a notable peak in the early wet season, with lowest numbers in the late wet season.
50–70 m above me at observation points, but appeared to be flying higher, up to 100 m high, at the mid-point of their commute.

The flight path of birds travelling to Crocodylus Park was directly south of the waste depot. Some birds had a direct path over Holmes Jungle to Crocodylus Park. However, most birds appeared to follow Vanderlin Drive, with a distinct left-hand turn shortly before McMillans Road, for the short distance across to the roost site at Crocodylus Park.

**Discussion**

All the Pied Herons using the East Arm roost came from the Shoal Bay/Leanyer/Holmes Jungle area of Darwin’s northern suburbs. I cannot be sure that some birds did not also roost at the feeding sites or at other roosts such as the later-identified roost at Crocodylus Park, although the large numbers roosting at Crocodylus Park is a post-2014 phenomenon (Grahame Webb pers. comm.). Thus, my urban population estimates of 1700–5900 individuals should be regarded as minimum counts. Chatto (2000) estimated the Pied Heron breeding population in the Top End to be over 22,600 individuals based on aerial surveys of breeding colonies, with the majority of breeding occurring in the north-western part of the Top End, within about 300 km of Darwin. Morton et al. (1993) estimated that the maximum Pied Heron population in the Alligator Rivers region alone was around 50,000 individuals. The peak population of Pied Herons in Darwin of more than 5000 individuals in the late dry season represents a moderate fraction of the overall Top End population. The food available at the sewage ponds and waste depot appears to be an important resource for a small but significant proportion of the Top End population of this species during the late dry season, and may be maintaining the overall population in the north-west Top End at a slightly higher level than would otherwise be possible.

Pied Herons are present, but do not breed, in the Darwin urban environment throughout the year. The fewer birds remaining during the late wet season and early dry season correspond with the identified active period of breeding colonies from January to May (Chatto 2000), suggesting that the majority of Pied Herons leave the urban area for breeding sites in the wet season. The closest breeding colonies – within 100 km of Darwin – are in the mangrove-lined mouths of the Finniss and Adelaide rivers. The largest colony observed by Chatto was about 3000 individuals, while the average colony size was around 1000 individuals (Chatto 2000). The drop in urban population from the 2012 late dry season peak to the 2013 breeding season low of over 3000 birds is larger than the size of any one breeding colony, thus most likely comprising individuals that arrive from and depart to more than one breeding colony.

An alternative to local movements is that the significant increase in the number of Pied Herons in Darwin’s urban sites in the early wet season is made up of migratory birds, recently returned to Australia from the north where they had migrated to escape the
Habitat use by Pied Herons

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food and habitat shortages of the late dry season. Pied Herons are known to migrate regularly across the Torres Strait between North Queensland and Papua New Guinea for the dry season (Garnett & Bredl 1985; Marchant & Higgins 1990; McKilligan 2005). Although they are present in Papua New Guinea year-round, they are not known to breed there (Coates 1985; Beehler et al. 1986). I am not aware of any records of Pied Herons departing from or arriving to the Top End from the north, but migration of the Top End populations to Papua New Guinea or Indonesia should not be ruled out and is worthy of further investigation.

The seasonal cycle of movements of Pied Herons in the Darwin region is not as clear as for other Top End systems. For example, a study on the Magela Creek floodplain in April 1981 (i.e. towards the end of the Pied Heron breeding season) identified that over 90% of individual Pied Herons observed were in immature plumage (Recher et al. 1983; see also Garnett 1985). This is consistent with a broader seasonal pattern in the Magela Creek system of near-complete absence during the wet season and a gradual increase over the dry season, with corresponding later offsetting pulses of birds in different nearby systems linked to wetting and drying cycles of the different floodplains (Morton et al. 1993). Most likely, the April 1981 birds were newly fledged from one of the now-identified breeding colonies on the East or South Alligator rivers (Chatto 2000), and they are gradually joined on the floodplains by adults as they depart the heronries each May. The seasonal offsetting patterns identified by Morton et al. (1993) weakly suggest that birds from that area do not migrate north to Papua New Guinea or Indonesia, but are able to find suitable habitat in the late dry season within the mosaic of drying wetlands in the western Arnhem Land region.

There is a pool of non-breeding Pied Herons in Darwin through each wet season that do not congregate in heronries, and there may be others scattered across a wide area of wet floodplain or other habitat. This would explain the larger population estimate of 50,000 birds in the Alligator Rivers region by Morton et al. (1993) compared to Chatto’s (2000) estimate of 22,600 birds present at all breeding colonies in the western Top End. Pied Heron plumage varies by age, with juvenile and immature birds having different crown feather colour and other more subtle differences to adults. HANZAB indicates a juvenile plumage and two immature plumages (Marchant & Higgins 1990), suggesting that Pied Herons most likely do not breed until at least the end of their second year of life. Pied Herons are never completely absent from the urban area of Darwin, unlike other waterbirds such as Magpie Geese that visit the urban area for limited periods and then have seasonal absences, and it may be that the remaining (approx. 2000) herons are immature. As a starting point, closer observation of the plumage of individuals foraging in Darwin at different parts of the seasonal cycle may shed further light on population and breeding dynamics.

An alternative explanation for the presence of Pied Herons in Darwin during the breeding season may be that the food supply in Darwin in the late dry season is limiting and that the urban area acts as an ecological trap (sensu Robertson & Hutto, 2007) from...
which some birds struggle to escape. Pied Herons are regularly trapped accidentally at Crocodylus Park within food preparation areas, and are often noted to be either very skinny or otherwise injured or in poor condition (Simon Ferguson, pers. comm.). In addition to studying the plumage of birds through the seasons, observation of body condition of individuals roosting at Crocodylus Park at different parts of the seasonal cycle may provide insights into source/sink dynamics for the Top End population.

**Flight path conflict with Darwin airport**

There is some potential for conflict between Pied Herons when they are flying to or from the harbour roost sites and the short final approach of aircraft arriving at Darwin International Airport from the east. Given the large size of flocks identified in this study, the potential for multiple strikes exists, although the most common flight path of flocks of Pied Herons appeared to be safely lower than the approach paths of aircraft arriving from the east. Planes taking off to the east appear to climb steeply after take-off, and birds appeared much less likely to conflict with departing planes. Although Darwin airport has a high number of recorded bird strikes, Australian Transport Safety Bureau (ATSB) statistics for the period 2001–2013 indicate only five identified bird strikes at this airport were associated with Pied Herons, out of 1004 bird strikes involving identified species (ATSB 2012, 2014).

The flight path to the new roost site at Crocodylus Park does not cross the airport approach paths, so potential for bird strikes does not currently exist. However, the ability of Pied Herons to change roost sites is demonstrated here, and if in future the birds abandon Crocodylus Park in favour of new harbour roosts then careful monitoring may be required. Roost sites further west than Catalina Island in Darwin Harbour, e.g. the mangroves around Reichardt or Bleezers creeks, may present a more direct threat to aircraft.

**Roost site fidelity**

Observations through the 2012–2013 study suggest that the roost site fidelity towards Catalina Island may have broken down during that time. Increased activity in the East Arm of Darwin Harbour associated with Inpex, including substantial night-time activity including bright lights, may have caused the Pied Herons to change roost sites. Alternative nearby roost sites over 2013–2014 include the settlement ponds at East Arm (Amanda Lilleyman pers. comm.) while in some evening surveys Pied Herons flew further south-west to South Shell Island, or beyond to at least as far as the mangroves fringing Wickham Point. These longer commutes between feeding and roosting sites added around 1–5 km to the twice-daily flight of Pied Herons.

The follow-up observations in 2015 show that Pied Herons have abandoned the harbour roost sites, and now roost quite close to the feeding sites at Crocodylus Park. The roost site itself, the artificial tree-lined river habitat at Crocodylus Park, does not appear to be a limiting factor for Pied Herons, as they only roosted on a fraction of all available
trees. Simon Ferguson, Zoo Supervisor living on-site at Crocodylus Park, noted that the roosting of birds became apparent after completion of the artificial river and also that there had been many Pied Herons roosting at this spot continuously from around May 2014 to the present (September 2015). My single count at the end of September 2015 of about 3300 birds is broadly consistent with the September and October 2013 counts of about 2100 and 3900, indicating that this is a complete count of local roosting Pied Herons, and that the overall population of Pied Herons feeding and roosting in Darwin has not changed dramatically with the change in roost site.

It appears that there is a range of roosting options, so the abandonment of Catalina Island, possibly due to disturbance associated with Inpex, does not appear to be a critical disturbance to the urban population of Pied Herons.

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References


Evidence of rock kangaroo seed dispersal via faecal seed storage in a tropical monsoon community

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Abstract

While some of the plant species of the ‘Sandstone Country’ along the escarpment of western Arnhem Land produce fleshy fruits and appear to rely on biotic methods of seed dispersal, little is known about the methods by which this is achieved — and few potential dispersers co-occur in the sandstone outcrop communities. For the present study, scat collections were made on outcrops in the northeastern area of Kakadu National Park with the hope of uncovering relationships between local frugivores and fruit-producers, and providing evidence for seasonal storage of mammal-dispersed seeds in scat prior to germination. The goals of the present project were to collect and identify sandstone community macropod scat, determine the identity of seeds present in the scat, and provide support for the role of browser/grazer macropods as effective seed dispersers via faecal seed storage in an otherwise disperser-poor local fauna. Scat containing seeds was identified as belonging to the Black Wallaroo (*Macropus hernae reinhardt*), a rare and locally-endemic macropod considered an intermediate browser/grazer. These seeds were successfully germinated and the seedlings identified using molecular phylogenetic techniques as *Gardenia fucata* (Rubiaceae), an endemic rock-specialist species — thus establishing the first confirmation of effective seed dispersal by a ‘rock kangaroo’ in this region and the first identification of a seed disperser for this uncommon *Gardenia* species. The results provide support for the role of browser/grazer macropods as occasional effective seed dispersers of rock-specialist plant species in the northern monsoon tropics of Australia via faecal seed storage.

Introduction

Effective seed dispersal is defined not only by the movement of seeds, but also by the successful recruitment of new individuals following that movement (see Schupp *et al.* 2010). For fleshy-fruited plant species relying on endozoochory, identifying effective dispersal requires knowledge of whether a given animal ingests the seeds, whether those seeds survive passage through the digestive tract, and whether seed germination follows deposition of seeds in faecal material.
Production of fleshy fruits is relatively common among the woody rock-specialist plants occurring in the monsoonal and fire-prone ‘Sandstone Country’ along the escarpment of western Arnhem Land (Northern Territory, Australia). While numerous species appear to rely on endozoochorus seed dispersal, little is known about the methods by which this is achieved in this habitat – and few potential dispersers co-occur with the fruit-producing plants found growing there (Menkhorst & Knight 2011).

Rock-dwelling macropods are fairly common in the northern monsoon tropics of Australia and reflect an unusually high regional species diversity there (Telfer & Bowman 2006). The Nabarlek (*Petrogale concinna*), Short-eared Rock Wallaby (*Petrogale brachyotis*), Common Wallaroo (*Macropus robustus*) and Black Wallaroo (*Macropus bernardus*) (Fig. 1) all occur in sympatry (Menkhorst & Knight 2011). Because these northern ‘rock kangaroo’ taxa are largely nocturnal and generally shy, empirical knowledge of their behaviour, diet and distribution is limited (Richardson 2012).

In a 1998 paper, Telfer et al. compiled Aboriginal knowledge of rock kangaroo feeding habits that included numerous anecdotal accounts of opportunistic macropod frugivory – even though the dominant understanding of foraging behavior has defined these animals as intermediate browsers/grazers (Tuft et al. 2011). A later study (Telfer and Bowman 2006) using scat contents to explore niche separation among northern macropods found that at least one of the species, *Macropus bernardus*, may be able to inhabit the most rugged and severe habitat because of its ability to utilise leaves, fruits and seeds from a range of rock-specialist plants during dry seasons. In conjunction with this work, Telfer et al. (2006) designed a scat identification key to determine current

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**Fig. 1.** Black Wallaroo (*Macropus bernardus*). (Stephen Zozaya, by permission)
distribution patterns for macropod species in the Top End region of the Northern Territory; a study that set the stage for a follow-up analysis correlating these distributions with habitat characteristics (Telfer et al. 2008).

Coupling the work of Telfer and colleagues with field observations, Martine & Anderson (2007) postulated that rock-dwelling macropods play an important role in short-distance seed dispersal of sandstone endemic plants through a three-step process consisting of fruit ingestion, “faecal seed storage,” and “seasonal redispersal” via wet season rains. However, the authors did not test experimentally whether particular animal taxa actually functioned in this role for specific plant taxa.

The goals of the present project were to:
1. Collect and identify sandstone community macropod scat, examine it for seeds and, if present, germinate them; and
2. Identify the resultant plant(s) species using morphology and, if needed, molecular tools.

Materials and Methods

In May 2013, 80 scat pellets were collected from upper elevation scat piles on sandstone outcrops in the vicinity of Merl Campground and Cahills Crossing, East Alligator region, Kakadu National Park (Northern Territory, Australia). Two investigators gathered scats using a haphazard sampling scheme during which all intact scats encountered were collected during a single day over a period of roughly eight hours. At the time of collection, the scats were run through the identification key published by Telfer et al. (1996). Although the scats were relatively dry at collection, they were allowed to further air dry before being packed in paper coin envelopes and shipped to Bucknell University for future work.

In July 2013, the scats were dissected and searched for the presence of seeds. Seeds were removed and directly sown into a soil tray without pre-treatment in order to ensure that any germination success might be based on natural processes alone (notably the passage through an animal’s gut and, perhaps, time passed in the scat). Because seedling recruitment on rock outcrops appears to be highest in cracks and fissures where scats/seeds are covered with accumulated detritus (CTM, pers. obs.), experimental seeds were shallowly planted below the soil surface. The tray was then placed in a growth chamber with temperature, light, and humidity settings previously found to be successful for two species of Solanum endemic to the area where the scats were collected (as per Lionheart 2014). The soil was kept moist at all times.

Once seedlings were apparent, they were transplanted into pots and moved to the Bucknell University research greenhouse, where a temperature and light regime matching present conditions in Kakadu was already in place. Following establishment, leaf material was removed and dried on silica for future DNA work.
In April 2014, CTM returned to Kakadu, compared leaves of the germinated greenhouse plants to plants on the site where the scat was collected and, finding a potential match, collected voucher material. This material was examined and identified by staff at the Northern Territory Herbarium in Palmerston, then accessioned at the Manning Herbarium at Bucknell University for later use in DNA extractions.

Dried leaf material was extracted using a modified CTAB protocol (Doyle & Doyle 1987). Leaf tissue was pulverised using a GenoGrinder (SPEX Sample Prep) and steel beads in 2 ml microfuge tubes. The CTAB plant solution was incubated for 30 min at 37°C, centrifuged, and moved to a clean tube. The aqueous solution was extracted with chloroform: isoamyl alcohol (24:1) twice. The DNA was precipitated with ice cold 100% isopropanol for 20 min at -20°C, then pelleted for 20 min at 4°C in the centrifuge at 7000 rpm. The DNA pellets were cleaned with two consecutive washes of ethanol, 75% and 95%, respectively, and re-suspended in 100 ml ddH₂O. The re-suspended pellet was incubated at 37°C with RNase for one hour, and let to sit overnight at 4°C. The DNA was then frozen at -20°C for storage.

The ITS (internal transcribed spacer) gene region was amplified using PCR with the following protocol for a 30 µl reaction volume: 2 min denaturing at 95°C; 29 cycles of denaturing for 30 sec at 95°C, annealing for 30 sec at 56.8°C, and elongation for 30 sec at 72°C; with a final extension for 5 min at 72°C. The reaction mixture for the PCR includes 6 µl GoTaq Flexi buffer (Promega, Madison, WI), 2 µl of MgCl₂ (1 mM), 1 µl each of forward and reverse primers (0.2 mM, Invitrogen, Carlsbad, CA), 0.6 µl dNTP mix (0.2 mM, Promega), and 1.25 units of GoTaq Flexi (Promega). The primers designed for these studies were as used in Jordon-Thaden et al. (2010) for the ITS of the ribosomal DNA sequence. The primers amplified the ITS2, ITS1, and the 5.8 S rDNA region. This was done with the ITS-18 forward 5'-GCA TGT TIT CCC AGT CAC GAC GGA AGG AGA AGT CGT AAC AAG G-3' which includes an M13 extension (the last 19 bases). The reverse ITS-25 primer was 5'-ACT TCA GGA AAG AGC TAT GAC GGG TAA TCC CGC CTG ACC TGG-3' which also includes an M13 extension (the first 21 bases). For Sanger sequencing of the ITS region, the M13 extension primer alone was used for the PCR products that had been generated with the M13 extension attached to the ITS forward and reverse primers. The M13 extension for forward primer is 5'-GCA TGT TTT CCC AGT CAC GAC-3' and reverse is 5'-ACT TCA GGA AAC AGC TAT GAC-3'. The PCR products were cleaned with the Promega Wizard cleaning system (Promega), and Sanger sequenced with an ABI sequencer at the Heck Genomics Institute at Pennsylvania State.

Sequenced gene regions were processed with Geneious R7 (Biomatters Ltd.) and then compared via BLAST search to confirm the match and the identification of the taxon grown from seeds recovered from the scat.

Finding a generic match, multiple ITS accessions of congeners were downloaded, along with an outgroup taxon, and used for phylogenetic comparisons with the two
“unknown” accessions. Sequences were aligned in Geneious R7 (Biomatters Ltd.) and a maximum likelihood ITS tree was generated with GARLI (Zwickl et al. 2006), with bootstrap values based on 10,000 reps. Novel sequences generated from the individual collected from scat and the specimen taken from the potential match in the field were deposited in GenBank (see ID numbers in tree figure).

Results

Use of the Telfer et al. (2006) scat key confirmed that the scat collected on our site was that of the Black Wallaroo, an escarpment-restricted species listed as Near Threatened by the IUCN (Woinarski 2008) largely because of a small total global population limited to a geographic range of about 30,000 km² (Telfer & Calaby 2008). Of the 80 scats examined, only one contained seeds, with seven seeds removed from the same sample. These seeds germinated at a rate of 100% without pre-treatment.

Plant voucher material collected at the scat collection site was identified by Ian Cowie at the Northern Territory Herbarium as Gardenia fucata R.Br. ex Benth. (Rubiaceae), a small tree endemic to the sandstone escarpment country (Puttock 1997) in the Top End region of the Northern Territory (Fig. 2). While no previous ITS accession existed in GenBank for G. fucata (or any other Australian Gardenia), the BLAST search confirmed that the ITS sequences of the greenhouse-grown seedlings (GenBank KP657895) and the wild-collected specimen of G. fucata (GenBank KP657896) were closely allied to other members of the genus Gardenia. Using Gardenia ITS sequences from GenBank (and Ixora pavetta as the outgroup taxon) for comparison to our greenhouse-grown and wild-collected accessions, phylogenetic analysis provided support for their close alliance (Fig. 4). With this evidence, we can conclude that the seed found in the scat was most likely Gardenia fucata, a species occurring in fairly low abundance in the site where the scats were collected.

Discussion

While previous scat analyses (Telfer & Bowman 2006) and compilation of local Indigenous knowledge (Telfer et al. 1998) suggested that Black Wallaroos occasionally eat fruits and seeds, no empirical evidence for their role as effective seed dispersers has been previously established. Our results show that Black Wallaroos in the East Alligator region of Kakadu National Park occasionally ingest the fruits of Gardenia fucata and that those seeds are not only passed intact in scat but are also able to germinate at a high rate (100%; n=7). In
the sandstone habitats where this rock-specialist plant species occurs, few other potential seed dispersers are present – meaning that even if Black Wallaroos (and potentially other rock macropods) are not frequent frugivores, bouts of frugivory and seed dispersal may still play an important role in recruitment of new individuals of *Gardenia fucata*. The fruits of *G. fucata* begin ripening in July (peak dry season) and are not especially fleshy, the mesocarp instead having a fibrous nature (Puttock 1997). Local Indigenous knowledge compiled by Telfer & Garde (2006) includes accounts of *M. bernardus* (and two other rock kangaroo species) consuming the leaves of *G. fucata*, but not the fruits.

**Outcrop interactions** The sandstone outcrops of the escarpment country offer a unique suite of underlying opportunities and challenges to their resident flora and fauna. Because outcrops typically retain water, they offer a refuge for plant communities requiring more available resources than the surrounding eucalypt savannah communities (Brock 2001) and thus may support a richer suite of invertebrate and vertebrate animals. Outcrops support communities that are distinct from the surrounding landscape – including numerous endemic species (Brennan 1986; Brock 2001). At the same time, these outcrop communities, while similar to the nearby plateau country, also remain distinct from it by virtue of their smaller size and island-like isolation. In the most pronounced cases they serve as refugia for drought-intolerant species that, formerly more widespread, were forced to retreat to these islands of tolerable habitat during the Pleistocene – the best example perhaps being *Allosyncarpia ternata*, a tree common in previously widespread rainforests that is now found only in refugial monsoonal gorges along the Arnhem Plateau escarpment (Russell-Smith 2009).

These restricted rock outcrops may have also served (and continue to serve) as refugia for *Macropus bernardus*. Telfer & Bowman (2006) suggest that *M. bernardus*, the species with smallest range (Fig. 3) of six *Macropus* species in Australia (Woinarski 2008; Coulson and Eldridge 2010), was forced to retreat to these habitats during glacial periods of the Pleistocene and subsequently adapted to exploit rock-specialist plants (many of them chemically-defended). All rock kangaroos are considered refuge-dependent, with disjunct occurrences across broad geographic regions reflecting the scattered distribution of compatible habitat (Tuft et al. 2011); and this is strongly pronounced in *M. bernardus*. An analysis of environmental correlates found that *M. bernardus* distributions, more so than the patterns for three sympatric rock-dwelling macropods, are especially linked to the presence of rugged, rocky terrain (Telfer et al. 2008).

For plant communities on outcrops, these habitats operate like terrestrial islands, with isolated populations for which gene exchange is challenged through the limited movement of pollinators and seed dispersers (Martine & Anderson 2007; Roche et al. 2014). While little has been done on the effects of outcrop-to-outcrop distance on gene exchange, the relationship between two sandstone specialist sister species, *Solanum asymmetricphyllum* and *S. sejunctum*, shows that the taxa remain distinct (Brennan et al. 2006; Martine et al. 2006) even though they can hybridise and are separated by just 10 km at the nearest point in their combined distribution (Gilman et al. 2014). Previous inferences (Anderson
adapted for movement across flat ground (a consequence of being exceptionally adapted for climbing around rocks) (Telfer & Bowman 2006), they are most likely to facilitate short distance dispersal within outcrops. This might be accomplished through nightly dry season foraging bouts where seeds are moved into ‘faecal seed storage’ in/around daytime roosting spots (Telfer & Griffiths 2006), followed by ‘seasonal redispersal’ via wet season downpours – an inference supported by the frequent occurrence of young plants in steep washes, ravines and deep cracks (CTM, pers. obs.).

The present study shows that rock-dwelling macropods may occasionally function as effective seed dispersers for at least one sandstone outcrop endemic and, by nature of the locations and manner in which seeds were collected, appear to provide faecal seed storage as per Martine & Anderson (2007). Occasional seed and fruit opportunism on the part of the macropods (see Jarman 1994; Telfer & Bowman 2006) may thus be as important to the plants as to the animals, particularly during seasons when the availability of grasses and foliage is reduced.

Whether this interaction then leads to seasonal redispersal during the monsoon season is left to be determined, but knowing that seeds stored in faeces on upper elevation rocks can germinate without any additional treatment aside from the presence of a moist substrate allows us to assume that seedling recruitment can follow redistribution of seeds by water and gravity. Telfer et al. (2006) found that although scats of the Short-tailed Rock Wallaby may persist in nature for over two years, most scats are lost within six months, ostensibly being degraded and washed away by wet season rains. Faecal seeds redispersed during these rains might then reap the benefits of increased access to nutrients and carbon supplied by the faeces (Kobayashi et al. 2011).
Given the low numbers of seeds recovered in this study, we cannot discount the potential for post-dispersal seed predation to render our findings moot. Ants and rodents are known to predate seeds released in scat (e.g. Hulme 1998; Manzano et al. 2010; Pecco et al. 2014); and both are in abundance in the study area. However, seeds moved but not consumed during bouts of post-dispersal predation (see Azacarate & Manzano 2011) may benefit from redispersal similar to that hypothesised above.

There is growing evidence that small- and medium-sized mammals throughout the northern monsoon tropics of Australia have recently experienced and continue to face major declines, even in Kakadu National Park and other protected areas considered to be ecologically intact (Woinarsi et al. 2001; Ziembicki et al. 2012; Woinarsi & Fisher 2013). Further work is needed to help understand and protect the links between these mammals and the plants that may depend on them, at least in part, for their long-term survival. Descriptive studies like this one provide individually small steps that should prove cumulatively integral to conservation efforts, particularly for organisms whose natural histories are as poorly understood as those highlighted here.

**Conclusions**

This study establishes the potential role of rock macropods as dispersers of rock-specialist plants in the monsoon tropics of Australia. Scat containing seeds was identified as belonging to the Black Wallaroo, a rare and locally-endemic macropod considered an intermediate browser/grazer. Seeds were successfully germinated and seedlings were identified using molecular techniques as *Gardenia fucata*, an endemic rock-specialist species — thus establishing the first confirmation of effective seed dispersal by a rock kangaroo in this region and the first identification of a seed disperser for this uncommon *Gardenia* species. The results provide support for the role of browser/grazer macropods as occasional effective seed dispersers of rock-specialist plant species via faecal seed storage.

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References


Report of the presence of *Hapalotrema synorchis* and *H. postorchis* (Digenea: Spirorchiidae) in marine turtles (Reptilia: Cheloniidae) in Northern Territory waters

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**Abstract**

The spirorchiid digenean *Hapalotrema synorchis* was recovered from the heart of a juvenile Hawksbill Turtle (*Eretmochelys imbricata*) found deceased on a local Darwin beach. The turtle was in poor condition, showing many characteristics associated with spirorchiid infection. A second necropsied turtle showed signs of infection with spirorchiids but adult specimens were not recovered. Examination of specimens held at the Berrimah Veterinary Laboratory found another *E. imbricata* infected with *H. synorchis* and a Green Turtle (*Chelonia mydas*) infected with the related *H. postorchis*. Despite previous reports of infected turtles, this is the first confirmed identification of *H. synorchis* and *H. postorchis* from Northern Territory waters.

Marine turtles are hosts to spirorchiid digeneans that inhabit the cardiovascular system, including the tissues and blood vessels of all the major organs (Glazebrook *et al.* 1989; Chapman *et al.* 2015; Flint *et al.* 2015). Spirorchiids have been implicated as causes of strandings and mortality in turtles around the world, with infected turtles often showing muscle wastage, plastron shrinkage and general listlessness (Glazebrook *et al.* 1989; Stacy *et al.* 2010).

Clinical signs of infection with spirorchiids include sunken eyes, plastron shrinkage and generalised muscle wastage (Glazebrook *et al.* 1981; Gordon *et al.* 1998) and in some cases, neurological symptoms (Jacobsen *et al.* 2006). However, the direct correlation of spirorchiid infection with disease in turtles is poorly understood (Stacy *et al.* 2010). Apparently healthy turtles were found to harbour significant spirorchiid infections by Stacy *et al.* (2010), while Glazebrook *et al.* (1989) reported only 26% of infested turtles displayed any clinical signs, which were non-specific. Gordon *et al.* (1998) found that 98% of stranded turtles showed evidence of infection with spirorchiids, although only 45% contained adult digeneans. Flint *et al.* (2010) found that 75% of turtles presented
for necropsy were infected with spirorchids, which were deemed the primary cause of death in around 42% of cases. Egg granulomas can be found in high numbers in turtles where adult digeneans are apparently absent (Gordon et al. 1998; Flint et al. 2010).

Although reports of cardiovascular digeneans in stranded marine turtles from Northern Territory waters are known (Limpus 2009; Mackouras & Griffiths 2014), no species-level identification has been undertaken. Mackouras & Griffiths (2014) reported on the pathological examination of eleven marine turtles stranded from June 2012 to June 2014 in Northern Territory waters. One of six Green Turtles (Chelonia mydas) was found to have died as a result of severe cardiovascular digenean infection; two of five Hawksbill Turtles (Eretmochelys imbricata) were infected with cardiovascular digeneans, but these were not considered the cause of death.

A juvenile E. imbricata, was submitted to the Berrimah Veterinary Laboratories (BVL) for post-mortem assessment after it was found deceased on a local beach (in the suburb of Nightcliff (12.3783°S, 130.8453°E) on 10 July 2014. The turtle weighed 2.5 kg and externally appeared thin and in poor condition. During necropsy it was noted that the abdominal fat was absent except for remnants attached to the plastron (lower shell) and around organs and there was extensive clear fluid present within the abdomen and pericardial sac. Granulomas containing digenean eggs were present in the blood vessels associated with the lungs, liver, heart, intestine and fat. Digeneans were recovered from the heart and placed into a vial of 70% ethanol.

A second (female) E. imbricata was submitted to the BVL after it was found deceased entangled in a net at Lee Point (12.3292°S, 130.8844°E) on 8 January 2015. The turtle weighed 3.5 kg and had a curved carapace length of 35 cm. The turtle was in good body condition. No digeneans were recovered from the heart or lungs. A small number of egg granulomas were observed in the blood vessels of the intestinal system.

A subsequent search of the BVL parasite collection found two vials of digeneans identified as Hapalotrema sp. The first came from an Eretmochelys imbricata collected from Bare Sand Island, Fog Bay (12.5369°S, 130.4189°E) in 1997 and the second from a Chelonia mydas (collection data listed as Darwin) in 2003.

From each sample, the anterior end (anterior to the ventral sucker) of one specimen was removed, placed into 100% ethanol, and used for genetic analysis. The remaining portion of that specimen was stained with aceto-orcein, dehydrated through a graded ethanol series and mounted in Canada Balsam. The 1997 E. imbricata vial contained a number of specimens, so an intact specimen was also mounted. Further specimens were left intact as unmounted specimens.

The digeneans recovered from the 2014 E. imbricata were identified as Hapalotrema synorchis through a combination of morphological and genetic analyses. The specimens recovered from the 1997 E. imbricata were also identified as H. synorchis and the specimens from the Chelonia mydas were identified as H. postorchis through morphology; unfortunately the
Fig. 1. *Hapalotrema synorchis*. Lateral view of whole specimen orientated upside-down as is Fig. 2, the convention for digeneans. Abbreviations: OS, oral sucker; VG, vitelline (= yolk) glands; VS, ventral sucker. Scale bar = 1 mm. (Adam Bourke)

Fig. 2. *Hapalotrema synorchis*. Ventral view of stained whole mount specimen. Abbreviations: AT, anterior testes; IC, intestinal caecum; OS, oral sucker; PT, posterior testes; VG, vitelline (= yolk) glands, VS, ventral sucker. Scale bar = 1 mm. (Adam Bourke)

A genetic analysis was unsuccessful on these specimens, possibly because they may have been initially preserved in formalin.

The specimen collected in this study were easily identified to the genus *Hapalotrema* based on the morphological criteria of the genus (see Platt 2002). Morphologically, the specimens from *E. imbricata* exhibited the testes arrangement as indicated in Platt and Blair (1998) for *H. synorchis*, with testes forming compact anterior and posterior masses. Species identification was confirmed via genetic analysis using the PCR and sequencing methodology described in Chapman et al. (2015) for one specimen. A BLAST search was conducted on the resulting sequence, which indicated that the closest match (99% similarity) was *H. synorchis*. The specimens from *Chelonia mydas* were identified...
as *H. postorcbis* due to a similar number and arrangement of the testes (16 counted; 9 posteriorly, 7 anteriorly) and the vitelline glands as described by Dailey *et al.* (1993).

*Hapalotrema synorchis* was described by Luhmann in 1935, from a Loggerhead Turtle (*Caretta caretta*) from Tortugas, Florida (Platt & Blair 1998). Subsequent records of *H. synorchis* occurred from *Chelonia mydas* in the Gulf of Mexico (Caballero y Caballero 1962) and from *E. imbricata* in Puerto Rico (Fischthal & Acholonu 1976). Takeuti (1942) described *H. orientalis* from *E. japonica* (= *E. imbricata*) in Japanese waters, which was subsequently synonymised with *H. synorchis* by Platt & Blair (1998).

Platt & Blair (1998) reported *C. caretta* as a host for *H. synorchis* at Shark Bay, Western Australia (4 turtles), Heron Island (2) and Mon Repos (1) in central Queensland waters. Chapman *et al.* (2015) subsequently reported *H. synorchis* from *E. imbricata* from Redland Bay and Buddina Beach in south-east Queensland. In the study by Chapman *et al.* (2015), *H. synorchis* was found in two of three *E. imbricata*, but no *Chelonia mydas* from Queensland (of 22) or Hawaiian (of 10) waters. Stacy (2008) also reported *H. synorchis* from *C. caretta* from Florida (originally described as *I. pambanensis*, however later amended to *H. synorchis* when further sequences became available for comparative molecular analysis (see Chapman *et al.* 2015).

*Hapalotrema postorchis* was described by Rao from *Chelonia mydas* in the Gulf of Manar, India (Dailey *et al.* 1993). It has subsequently only been reported from the same host species in Hawaii (Dailey *et al.* 1993; Chapman *et al.* 2015: one of ten hosts examined) and various locations in south-east (Moreton Bay; Coolum) and central Queensland (Quoin Island, Gladstone Harbour) (Cribb & Gordon 1998: three infections recorded from an unstated total number of hosts examined; Gordon *et al.* 1998: three infections from 96 hosts examined; Chapman *et al.* 2015: total of three from 22 hosts examined).

Reports of infections with *Hapalotrema* from Australian waters are confusing as many reports of digeneans infecting turtles were undertaken prior to the review of the genus (see Cribb & Gordon 1998). Glazebrook *et al.* (1981) reported a *Hapalotrema* sp. from a heavily infected *Chelonia mydas* off Townsville, North Queensland. Although they stated that it was a different species to *H. synorchis*, Glazebrook *et al.* (1989) subsequently listed it as *H. synorchis* in their literature records of cardiovascular digeneans recovered from sea turtles. However, *H. synorchis* has never been reported from another *Chelonia mydas* in Australia, despite a large number of this turtle species being examined (see Chapman *et al.* 2015). Morphologically, *H. synorchis* is very similar to *H. pambanensis* (a synonym of *H. mebrai*), so it is more likely, without genetic confirmation, that the original record from Glazebrook *et al.* (1981) belonged to this species.

Four species of marine turtles are found in the waters of the Northern Territory (Mackouras & Griffiths 2014). During the period 2012–2014, a total of 60 marine turtles were reported as stranded in the Northern Territory; the most common species was the Green Turtle (*Chelonia mydas*; 26) followed by the Hawksbill (*E. imbricata*; 16) (Mackouras & Griffiths 2014). The vast coastline of the Northern Territory and the array of large
marine predators (saltwater crocodiles and sharks) unfortunately limits the access to fresh carcasses required for necropsy for a systematic survey of diseases of marine turtles.

During the 2012–2014 reporting period, 11 turtles were necropsied at the BVL. Of these, one (of six) *Chelonia mydas* was determined to have died from a severe cardiovascular digenean infection and one (of five) *E. imbricata* contained a heavy cardiovascular digenean infection, but this was not the suspected cause of death (Mackouras & Griffiths 2014). A further *E. imbricata* contained possible cardiovascular digenean egg granulomas, but no adult digeneans were observed (Mackouras & Griffiths 2014). Unfortunately, no digeneans specimens were available from these necropsies for examination in this study. A study into stranded Green Turtles from Gladstone, central Queensland, found that nine (of twelve) turtles were infected with spirorchiid digeneans at a level that could have contributed to their death (Flint et al. 2015). All turtles, however, were infected with a variety of spirorchiids that belonged to five genera, including *Hapalotrema*.

*Hapalotrema synorchis* is a widespread parasite that has been positively identified from three species of marine turtles in waters of Queensland and Western Australia, Japan, the Gulf of Mexico, Florida and Puerto Rico. *Hapalotrema postorchis* has only been reported in *Chelonia mydas*, with a distribution in waters of India, Queensland and Hawaii. This is the first confirmed report of the presence of *H. synorchis* and *H. postorchis* in waters of the Northern Territory.

The following voucher specimens have been deposited in the collections of the Museum and Art Gallery of the Northern Territory: *H. synorchis* from *E. imbricata* (2014) (D1532); *H. synorchis* from *E. imbricata* (1997) (D1545); *H. postorchis* from *Chelonia mydas* (D1546). The Genbank accession number for the *H. synorchis* is KT361641.

**Acknowledgments**

The authors are indebted to the staff at the BVL for access to specimens from turtle necropsies. The authors are also grateful to the Northern Territory Government Berrimah Farm Library staff for their speedy acquisition of reference material.

**References**


Hapalotrema in marine turtles


Coral communities in extreme environmental conditions in the Northern Territory, Australia

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Abstract

An extensive intertidal reef flat in the macro-tidal marine waters of the Northern Territory was chosen to investigate species composition and zonation persisting under extreme environmental conditions. Thirty-six visual belt transects were used to quantify scleractinian corals, benthic algae and other sessile invertebrates which varied in vertical and horizontal space. Thirty-four coral species were identified. Most species were represented by the family Merulinidae, with lifeform characteristics typical of species specialised in environmental tolerance to high sedimentation, turbidity and temperature (i.e. massive, sub-massive and encrusting growth forms with convex and steep sided morphologies, thick skeletal tissue and large polyps). Whilst the combination of environmental and ecological characteristics of this reef flat community can be viewed as distinctive to the Darwin region, a number of similarities can be compared to reef communities reported in extreme environments of the Arabian Gulf, Red Sea and other regions of tropical northern Australia.

Introduction

Coral communities which presently persist in extreme environmental conditions are of contemporary interest towards understanding species resilience and potential for adaptation to climate change (Hughes et al. 2003; Bauman et al. 2011, 2013a, 2013b; Dandan et al. 2015). Predictions for climate change stressors for coral communities include warmer sea temperature and changes in extreme episodic events such as heavy rainfall, storms and possible sediment and nutrient debouching from rivers and run-off (Gilmour et al. 2006). Such environmental perturbations are known to be associated with mass coral bleaching and sudden die off (Glynn 1993; Depezionski et al. 2013). Coral species of the coastal waters in the vicinity of Darwin Harbour, Northern Territory (Wolstenholme et al. 1997), the Kimberley coast, northwestern Australia (Dandan et al. 2015; Schoepf et al. 2015) and the Arabian Gulf (Sheppard & Sheppard 1991; Coles 1997; Coles 2003; Riegl 1999; Bauman et al. 2011; Rieg & Purkis 2012) survive in harsh conditions and offer insight to species with high climatic tolerances and adaptation.

Climatic trends in this region of Australia are changing. Since 1950, the Northern Territory average rainfall has risen 35.7 mm per decade during November–April and
fallen 0.4 mm per decade during May–October. From 1910 to 2003, the intensity of heavy daily rainfall gradually rose by 10% (Hennessy et al. 2004). Water temperatures at the shoreline may reach over 36°C at high tide, with tide pools and standing water bodies reaching over 43°C at low tide which is similar to the high temperature fluctuations in the Arabian Gulf, and well above temperature ranges corals traditionally considered limiting to coral survival (Coles 1997). Sea surface temperatures north of Australia have been at record-breaking highs. In 2010, temperatures north of Australia broke previous records by large margins and were also above average during the 2011–2012 La Niña event (Australian Bureau of Meteorology 2013).

An extensive intertidal reef flat community off the north-eastern shoreline of Darwin Harbour was chosen to investigate coral zonation on a macro-tidal shoreline. The marine waters of Darwin Harbour are subjected to daily and seasonal fluctuations in sea surface temperature, light availability and extreme levels of sedimentation and turbidity which provides insight to northern Australian coral species that have already adapted to extreme climatic conditions.

Methods

Study site

Nightcliff Reef (Fig. 1) was surveyed at low spring tide between September and October 1994. Darwin Harbour is a ria coast formed by the post-glacial marine flooding of a dissected plateau. Subsequent sedimentary infill has resulted in the formation of numerous embayments, islands and extensive mangrove-vegetated tidal flats (Semeniuk 1985). The dominant lithological type is ferricrete laterite with phyllite/siltstone and this is reflected by the presence of conspicuous medium to coarse grained lateritic pebbles of the upper beach sediments. The coral community at Nightcliff exists as a veneer reef by colonising hard substrates of rock and consolidated materials without accreting substantial calcium carbonate substrata (Hooper 1987; Mitchie 1987). The community extends seaward to a distance of approximately 500 m from the shoreline at low spring tide.

Tides in Darwin Harbour exhibit semidiurnal inequality with a spring tidal range is in the vicinity of 0.1–7.8 m. Tidal currents are very strong ranging from 0.25–1.4 m/s and recording as high as 2 m/s (Semeniuk 1985). The concentration of rivers, streams and creeks with accompanying discharge, and the strong tidal movement of the system, account for excessive sediment mixing and high turbidity (Michie 1987). Nightcliff Reef is fully emersed for 2–3 hours during low spring tides that are less than 1 m. Low spring tides occur between 1100–1600 hrs in the period September to March (wet season) and between 2300–0400 hrs in the period April to August (dry season).

Stratification

A topographic contour map of the reef was generated from random spot heights measured using a digital theodolite. All heights were referenced from a survey datum
point adjacent to Nightcliff Pier, and were converted to metres above the lowest astronomical tide (i.e. 0 m). The contour map was used to stratify the reef into three main vertical zones: (1) the upper reef flat which occupied a vertical height between 1.4–1.8 m; (2) the middle reef flat between 0.9–1.4 m; and (3) the lower reef flat between 0.3–0.9 m.

**Sampling**

Sampling was conducted using four replicate 1 m x 20 m contiguous belt transects that were haphazardly placed in each of the upper, middle, and lower reef flat vertical zones. The sampling was repeated in three localities (north, central and south locations) to account for any variations in substrata micro-topography, sediment deposition and taxa composition that may occur horizontally across the reef relative to the shoreline. In total, 36 belt transects were surveyed. Contiguous belt transect was chosen as the sampling unit to improve the recording of small and less abundant species, and representation of microhabitats (e.g. large or small coral colonies, sand patches, tide pools) in any given zone and locality (Chiappone & Sullivan 1991; Sullivan & Chiappone 1992). Species abundances of all sessile taxa were recorded by visual estimates of percentage cover and numbers of individuals for each 1 m² quadrat. Lifeform attributes of coral colonies were recorded using categories of English et al. (1994). When a particular species was encountered that could not be identified in the field, a sample of the species was collected for identification in the laboratory and also compared to collection specimens held at the Northern Territory Museum. Original taxonomic identifications for scleractinian corals followed that of Veron and Pichon (1976, 1980, 1982); Veron et al. (1977); Veron and Wallace (1985) and Veron (1986). Algal identifications followed that of Jaasund (1976); Cribb (1983); Cribb and Cribb (1985); Lawson and John (1987) and Price and Scott (1992). Updates to recent taxonomic revisions follow Wynne (2011); Budd et al. (2012); Guiry and Guiry (2013); Huang et al. (2014) and WoRMS Editorial Board (2015). All data presented in the text and figures are the arithmetic mean.

**Results and Discussion**

A total of 75 sessile species comprising scleractinian corals, algae, and sessile invertebrates were identified in the 36 transects sampled across the upper, middle and lower reef flat zones. 34 species of scleractinian corals from eight families were recorded (Table 1). The majority of species were members of the family Merulinidae, represented by 18 species (or 53% of all corals), followed by the Lobophylliidae and Poritidae, each represented by 4 species (23% of all corals). All remaining 5 families were represented by 1 or 2
species (24% of all corals). The more abundant corals, all with greater than 2% in mean percentage cover pooled for all 36 transects, were *Platygyra sinensis* (Fig. 2), *Porites cf. nigrescens*, *Astrea curta*, *Coelastrea aspera* and *Goniastrea retiformis*. Secondary dominant corals (1–2%) were *Porites lutea*, *Lobophyllia hemprichii*, *Platygyra daedalea*, *Galaxea astreata*, *Favites*

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**Fig. 2.** Massive colony of *Platygyra sinensis* at Nightcliff Reef, Darwin Harbour. This particular species and *Coelastrea aspera* have been sighted attaining colony sizes of up to 1.8 m in height. (Lawrance Ferns)
Table 1. Mean percentage cover of scleractinian corals, algae, other sessile invertebrates and physical substrates at Nightcliff Reef. Lifeform categories M = massive, S = submassive, E = encrusting, D/C = digitate/corymbose, R = ramose, T = turf, BF = bladed foliosc, F = foliosc, EC = erect coralline.

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<th>Species</th>
<th>Lifeform</th>
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<th>Middle Reef Flat (≥0.9–1.4 m)</th>
<th>Lower Reef Flat (0.3–0.9 m)</th>
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abida, Dipastrea speciosa, and Dipastrea rotumana. Corals with ‘massive’ and ‘sub-massive’ lifeforms were the most common growth morphologies with 20 species (or 59% of all corals). The other main lifeform was ‘encrusting’ comprising 12 species (35% total of all corals). Only one species with digitate and tabulate lifeform categories were recorded (6% of all corals).

Coral abundance and species richness varied between vertical zones and horizontal localities across the reef. Physical factors such sediment type, micro-topography, standing water, with the relative influence of each factor shifting between and within vertical zones and localities, is the likely cause of the observed variation (Table 1). Species cover and richness increased markedly seaward from the upper reef flat zone (1.4–1.8 m) to the lower reef flat zone (0.3–0.9 m) which has the highest coral abundance and species richness. This is consistent with similar studies that have found intertidal corals to be more successful below 1 m tidal height elevations and reaffirms the period of emersion time is an important factor determining the their upper vertical limit (Morrissey 1980;
Bull 1982). Coral abundance and species richness was significantly higher on the lower reef flat at the northern extent of the reef (41% mean cover, 26 species), and decreased towards the southern extent (9% mean cover, 13 species). This is attributed to a notable decrease in fine sediments at the northern extent, combined with a higher proportion of coral rock and rubble, and the presence of small tide pools with standing water (Table 1). High levels of fine sedimentation on substrates are not conducive to coral recruitment and growth (Rogers 1990; Rogers et al. 1994). It is postulated that predominant north-westerly tidal movement drives local cross-shore transport of finer sediment from north to south.

The coral species and lifeforms in these extreme conditions are characteristic of those that occupy sheltered reefs with high sedimentation and turbidity (Rosen 1971; Chappel 1980; Done 1982). Species with massive, submassive and encrusting life forms are more tolerant to thermal stress (Marshall & Baird 2000), while convex and steep sided growth morphologies facilitate sediment flow off surfaces (Lasker 1980; Rogers 1990). The most diverse species were from the Merulinidae, which are represented by species that are tolerant of thermal stress (Depeczynski et al. 2013; Schoepf et al. 2015) and well defined association with turbid water and lengthy periods of emersion. Corals with large polyps, like most species from the Merulinidae are efficient at removing sediment (Marshall & Orr 1931), and species such as Dipastrea spp., Goniastrea spp, Lobophyllia hemsberchii, Astrea curta, Platygia spp., and Symphyllia recta can easily manipulate silt and fine sands (Stafford-Smith & Ormond 1992). Species of the genera Coelastrea and Goniastrea, for example, are often found in conditions where corals may not be expected to survive (Veron 1986) and are dominant in high latitude reefs in waters of high nutrients and turbidity (Thomson & Frisch 2010). In the Arabian Gulf region, the Merulinidae has a greater representation of species compared to other coral families such as Acroporidae which has greater species diversity in the Indo-Pacific region (Coles 1993; Foster & Foster 2013).

Species of the Acroporidae were scarce, with only two species represented at very low coral cover. The Acroporidae is reported to occur in other regions with high thermal stress (e.g. Craig et al. 2001; Bauman et al. 2013b; Dandan et al. 2015; Schoepf et al. 2015). At Nightcliff Reef it is probably further limited by the high turbidity as it is a poor sediment rejector with small polyps less than 2 mm diameter (Stafford-Smith & Ormond, 1992). However, a small-polyped species, Porites cf. nigrescens (Poritidae) was abundant on the middle and outer reef zones. This species has also been recorded as abundant amongst a coral community surveyed at Cobourg Peninsula, Northern Territory (Bilyard 1995) and to the west on macro-tidal reef flats and lagoons off Sunday Island at the mouth of King Sound, Western Australia (Dobson 1999). In further regions, Porites nigrescens is reported as common on turbid fringing reefs to the south of Saudi Arabia (Jeddah to Jizan), but disappears to the north in clearer waters (Sheppard 1985). The species is regarded as a shallow water reef builder in environments with low light intensities and low wave energy (Cabioch et al. 1999). In northern Australia it appears to have adopted a similar niche to that occupied by Montipora digitata that is common on more sedimented
inshore reefs of eastern Australia, such as the reef flats of Magnetic Island, Queensland (Bull 1982; Mapstone et al. 1992).

Sediments for the upper and middle reef flat zones across the majority of localities were predominantly muds and silts on rock with intervening patches of muddy sand. Benthic algae were both more abundant and more diversely represented in these zones and collectively contributed to 38 species (Table 1). The brown foliose species *Padina australis* dominated the upper reef flat zone. Other conspicuous bladed foliose algae included *Sargassacea* holdfasts (probably *Sargassopsis decurrens*), *Splatoglossum asperum* and *Dictyopteris* sp. Considerable turf forming algae were also present on the upper reef flat zone, and dominated in the middle reef flat zone with *Amphirola fragilissima*, *Portieria bornemanni*, *Trickocarpa fragilis*, *Acanthophora specifera*, *Gracilaria salicornia* and *Anadyomene plicata* visually conspicuous and varying in relative abundance across localities.

The lower reef flat zone, which was dominated by corals, exhibited minimal algal cover (0.03–0.05% mean cover, Table 1). There was no evidence of competition from algae with corals as described in eastern Australia (Morrisey 1980), the Arabian Gulf (Sheppard et al. 1992) and the Red Sea (Loya 1977b). It appears corals have a competitive advantage in this lower reef flat zone due to the greater water depth and ability to cope with the high turbidity which results in lower light availability to benthic algae. In the shallower upper tidal zones, longer and more frequent emersion times exclude most coral species, and this allows rapid colonisation of benthic algae which also gain improved light attenuation for photosynthesis.

The algal species recorded at Nightcliff Reef have been widely reported across the Indo-Pacific, from Tanzania (Jassund, 1976) to eastern Australia (Morrisey 1980; Ngan & Price 1980). The abundance of benthic algal species at Nightcliff Reef is likely to be seasonal with species exhibiting variable growth and dominance between wet and dry seasons (e.g. Benayahu & Loya 1977b; Lawson & John 1987; Vuki & Price 1994). The dominance of fleshy brown algae in the upper reef flat zone, with the co-occurrence of turfing algae is similar to the reef flat zonation described in the Gulf of Elat, Red Sea (Benayahu & Loya 1977a, 1977b).

A notable observation was the limited representation of erect coralline algae from the genus *Halimeda* which is an important contributor to calcium carbonate accretion and grows abundantly on reef flats in eastern Australia (Morrisey 1980), Guam (Merten 1971) and the Gulf of Mannar, India (Rao 1972). However, in similar extreme environmental conditions such as the Arabian Gulf region, *Halimeda* is not recognised as a major component of inshore reefs (Sheppard 1985; Sheppard et al. 1992).

The coral species of Darwin Harbour offer valuable insights to physiological and evolutionary adaptations to persist in the most extreme environmental conditions. It is evident from this study and comparative investigations elsewhere that species with large polyps, thick skeletal tissue and massive or submassive lifeform strategies are amongst the most successful to surviving high sedimentation, nutrients and water temperatures.
The close proximity of Nightcliff Reef to coastal development, land-based sources of pollution and marine development combined with the changing climate pose future conservation challenges for this reef which is currently deemed at high risk to integrated local threatening processes (Burke et al. 2011).

Acknowledgments

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First record of two mangrove leaf slugs, *Elysia leucolegnote* and *E. bangtawaensis* (Sacoglossa: Plakobranchidae), in mangrove forests in the Northern Territory

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Abstract

Here we report for the first time on the occurrence of the distinctive and highly ephemeral sap-sucking sea slugs *Elysia leucolegnote* and *E. bangtawaensis* from mangrove forests from Darwin Harbour, Northern Territory, Australia. Individuals of both species apparently attain smaller body size than their counterparts elsewhere in Australia and the Indo-Pacific region, with maximum extended crawling lengths recorded between 17–22 mm. It appears the northern Australian (i.e. Northern Territory and northern Queensland) populations of *E. bangtawaensis* differ consistently from their counterparts elsewhere in the world in aspects of (parapodial and rhinophoral) colouration.

The ability to retain functioning chloroplasts sequestered from algal host(s) is widespread in the group of sea slugs known as sap-sucking slugs (order Sacoglossa). Numerous species in the genus *Elysia* (family Plakobranchidae, the largest family numerically in the Sacoglossa) are known for their ability to sequester live chloroplasts within their extensively branched digestive diverticula, imparting a bright green colour (e.g. Trench et al. 1973; Rumpho et al. 2008; Jesus et al. 2010). Some of them match their host food precisely (Burn 1998). The genus *Elysia* is species-rich, with some 95 named species (Bouchet & Gofas 2015) and at least that number again undescribed (RCW pers. obs). Its members are mostly small in adult size (< 20 mm) and live on algae in tidal pools, seagrass meadows or in sublittoral algal beds (Swennen 1997). However, four comparatively large (25–50 mm) species are specific to mangrove habitats, aggregating on mud in shaded pools without any apparent algae in the immediate vicinity (Swennen 2011). These mangrove-dwelling species, which have all been described in the last 25 years, are *Elysia leucolegnote*, *E. bangtawaensis*, *E. singaporensis* and *E. bengalenensis* (Swennen 2011). Collectively, these four species are commonly called mangrove leaf slugs, as animals resemble a fallen mangrove leaf (in both shape and coloration) when they have their wing-like parapodia relaxed and fully extended (by contrast, all other species of *Elysia* have the parapodia folded up on the dorsal side, with the margins meeting more or less in a wavy line mid-dorsally). All these mangrove leaf slugs are restricted to the
tropical Indo-west Pacific region, but only *E. leucolegnote* and *E. bangtawaensis* occur in both hemispheres, ranging from the western coast of India to the north-eastern coast of Australia (Swennen 1997; Rudman 2007; Rudman 2009; Swennen 2011). Despite the wide distribution and presence of both species in eastern Australia, neither has been previously recorded from anywhere in the Northern Territory.

Here we document the first occurrence of *E. leucolegnote* and *E. bangtawaensis* in Northern Territory mangroves, specifically Darwin Harbour. Like most others throughout the world, the discoveries were of hundreds of individuals, not just one or two. During the early dry season (April) of 2014 approx. 115 adults and juveniles of *E. leucolegnote* were encountered, and during the dry season of the following year (April–May 2015) around 90 adults of *E. bangtawaensis* were encountered.

### Elysia leucolegnote (Figs 1–3)

On 14 April 2014 approx. 75 adults and juvenile *E. leucolegnote* were found in a puddle in the landward mangrove fringe at Bayview, Sadgroves Creek (12.4419°S, 130.8611°E). These individuals were identified by AJB and confirmed by RCW. *Elysia leucolegnote* is characterised by having a white or yellowish border to the parapodia, a distinctive white triangular mark on the head, and a white line over the dorsal side of each rhinophore that connects with the one from the other rhinophore on top of the head (Swennen 2011) (Fig. 1). All Darwin animals displayed these characteristic features. Living individuals ranged in size from less than 4 mm up to 22 mm in extended crawling length (ECL) and each had a yellowish-green coloured digestive gland (Fig. 2), indicating they had not fed for some months (Swennen 2011). Further specimens were found on 20 April when approx. 40 small (10–15 mm) dark green coloured slugs were recorded in shallow puddles in a low, closed Stilt-root Mangrove (*Rhizophora stylosa*) forest at Virginia, Elizabeth River (12.5690°S, 131.0156°E) (Fig. 3). All the *E. leucolegnote* observed in Darwin mangroves were notably smaller than the maximum size of 41 mm known for this species elsewhere (Swennen 2011).

These records of *Elysia leucolegnote* from Darwin Harbour can be added to those already known for this species from Australia – from northern New South Wales and southern Queensland (Allan 1950; Thompson 1973; Burn 1998). However, it is possible some (if not all) of these records might relate to *E. bangtawaensis* as there are no recent records of *E. leucolegnote* from this region (RCW pers. obs.). Its occurrence in the Kimberley region is to be expected.

### Elysia bangtawaensis (Figs 4–7)

*Elysia bangtawaensis* is characterised by the prominent reddish or orange glandular spots along its parapodial margin (Swennen 2011). The description of its colour in life stated "no epidermal pigmentation other than the red and white glands" (Swennen 1998) and noted that the tips of rhinophores were pale-coloured or orange (Swennen 2011). This species was first recorded in the Northern Territory on 21 April 2015, when approx. 20 small (13 mm) individuals were found in shallow, partly shaded puddles amongst
Figs 1–3. *Elysia leucolegnote*. Specimens photographed in the laboratory and *in situ*. 1. An individual with partially relaxed parapodia (scale bar = 5 mm); 2. Yellowish-coloured, starved individuals with fully relaxed parapodia congregated in a shallow puddle near the landward mangrove edge; 3. Dark-green individuals congregated in a shallow puddle (4 mm depth) in a *Rhizophora stylosa* forest. (Adam Bourke)

Porupan Mangrove (*Sonneratia alba*) pneumatophores in the seaward mangrove community (Fig. 4) about 120 m east of the East Arm Boat Ramp carpark (12.4842°S, 130.9132°E). These individuals were identified by RCW. One month later (i.e. on 20 May) approx. 70 small slugs (9–17 mm) were observed in a similar habitat (Fig. 5) about 240 m west of the East Arm Port Precinct (12.4831°S, 130.9239°E).

These records of *Elysia bangtavaensis* from Darwin Harbour can be added to those already known for this species from Australia – from northern New South Wales (Cobb 2007, which were also first identified by RCW; Riek 2015), and northern Queensland (Mitchell 2009). As with *Elysia leucolegnote*, its occurrence in the Kimberley region is to be expected.

All the *Elysia bangtavaensis* individuals recorded from Darwin Harbour differed from previously described specimens in having black tips to their rhinophores (Fig. 6) and microscopic, iridescent blue spots covering much of the dorsal and ventral margin of the parapodia (Fig. 7). Interestingly, *E. bangtavaensis* with tiny, metallic blue spots have also been recorded from mangroves in Cairns, northern Queensland (Mitchell 2009).
However, to our knowledge animals with black-tipped rhinophores have not been reported previously. As is the case with *E. leucolegnote*, all individuals of *E. bangtawaensis* from Darwin Harbour were notably smaller than the maximum live length of 52 mm known for this species (Swennen 2011).

That both of these large species of *Efysia* are highly ephemeral in time and space is shown by the fact that we systematically sampled in mangrove forests in Darwin
Harbour for 25 years and never encountered them previously. Therefore, their detection is unlike that of the caddis slug Aiteng sp., which remained undetected in the harbour because it is microscopic (Neusser et al. 2015).

Acknowledgements

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References


Field identification of the *Platevindex* mangrove slugs (Mollusca: Gastropoda: Onchidiidae) of Darwin Harbour

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Abstract

Darwin Harbour supports nine species of mangrove slugs (family Onchidiidae) and currently the names of all them are unknown. It appears that the characters distinguishing the two *Platevindex* species allow animals to be accurately identified in the field on the basis of external characters and on differences in habitat. This note provides descriptions and information on the external characteristics of the two *Platevindex* species.

Mangrove slugs as they are commonly known, comprise a family of shell-less, pulmonate (air-breathing) gastropod molluscs. Members of the Onchidiidae are particularly special ecologically as they constitute one of only four families of gastropods (out of more than 400 families) with species living in marine, brackish, freshwater and terrestrial habitats (Dayrat 2009a). Onchidiid slugs have a wide geographic distribution, but most genera are exclusively found within the tropical and subtropical Indo-West Pacific region (Dayrat 2009b). The Onchidiidae is a poorly-known family with its systematics in a state of confusion (Dayrat 2009b, Dayrat, Zimmermann & Raposa 2011). Animals in this family have been understudied since the last experts specialising in the taxonomy of the family were active more than 70 years ago and there is currently no expert able to reliably identify members of the Onchidiidae (Dayrat 2009a; Benoit Dayrat pers. comm. 2013).

At present, five genera of onchidiids are thought to inhabit the mangroves of Darwin Harbour, only two of which have been assigned names — *Platevindex* and *Peronia* (Benoit Dayrat & Trish Goulding, pers. comm. 2016). Among these genera, nine species have been identified, however none has been formally documented and the nomenclatural status of all of them remains unknown. Hence, scientists dealing with them are required to allocate operational taxonomic units (OTUs, or ‘working names’) to these unnamed species until they are documented by a taxonomic specialist.

This note provides descriptions and information on the external morphological characteristics (i.e. shape, structure, colour and pattern) of the two unnamed species of *Platevindex* — herein referred to as *Platevindex* sp. 1 (blue) and *Platevindex* sp. 2 (orange).

*Platevindex* is a common tree-climbing genus of onchidiid inhabiting mangrove forests of the Darwin region of the Northern Territory and the two species are frequently encountered on the trunks and branches of mangrove trees during low tides. The genus
is characterised externally by having a noticeably narrower foot than underside of dorsal surface (= hyponotum) (i.e., a ratio of 0.25–0.3 foot width to hyponotal width depending on the degree of body contraction) (see Figs 1, 3), in contrast to other onchidiids.

Field identification of Darwin’s *Platevindex* slugs

Most onchidiids cannot be easily identified by non-specialists as few species display distinctive external traits (Dayrat 2010). It is the author’s opinion, however, that the unique external features and colouration of Darwin Harbour’s two *Platevindex* slugs do allow them to be distinguished and identified accurately to species in the field. The following descriptions of the external characteristics plus photographs of living animals of *Platevindex* sp. 1 (blue) and *Platevindex* sp. 2 (orange) are presented as an aid to distinguishing between them in the field.

The characteristics provided here are based on living adult individuals. Caution must be exercised when identifying juveniles though, as animals may differ in shape, colour and pattern as they mature. Where notable differences in external morphology or colour are evident between adults and juveniles, descriptions and photographs are provided.

*Platevindex* sp. 1 (blue) (Figs 1, 2)

**Description:** Size to 43 mm in extended body length (pers. obs.). Body ovate-elongate in shape, juveniles and subadults more circular; distinctively flattened. Notum (= dorsal surface) leathery, either smooth or warty in appearance, always moist to the touch. Photoreceptors (dorsal eyes) single, present almost to edge of notum. Foot sole distinctively narrow. Colour of notum variable, but usually dark grey with lighter grey-brown mottled patches or broken bands, commonly with dark black-grey reticulated mottling around edge; mottling weaker in adults, but conspicuous in juveniles and subadults (Fig. 2). Head, oral lobes and tentacles usually darker than underside of body. Foot sole commonly yellowish to light or dark cream in colour. Hyponotum (= underside

![Fig. 1. Dorsal and ventral views of living adult *Platevindex* sp. 1 (blue). The specimen is 40 mm in extended crawling length. (Adam Bourke)](image1)

![Fig. 2. Subadult *Platevindex* sp. 1 (blue) displaying the distinctive reticulated dark mottling around the edge of the notum. The specimen is 24 mm in extended crawling length. (Adam Bourke)](image2)
of dorsal surface) always pale bluish grey colour in adults, *never* with a mottled dark
colour radiating outwards from foot sole; juveniles commonly lighter cream in colour.

**Remarks on preserved specimens:** Disregarding contraction of the body and
some loss of colouration, most of the diagnostic features described above are clearly
distinguishable for this species in preserved specimens (i.e. those preserved in 70% 
ethanol). In particular, the lack of dark mottling surrounding the foot sole remains
distinct in preserved specimens.

**Ecology:** This species may occur throughout the entire mangrove forest (pers. obs.),
but adults are most commonly encountered on the trunks and branches of trees within
forests dominated by Stilt-root Mangrove (*Rhizophora stylosa*) and Pornupan Mangrove
(*Sonneratia alba*). Individuals are regularly found feeding on large woody debris in the
more seaward mangrove forest zones, but rarely observed on the forest floor (pers. obs.).

**Global distribution:** The true extent of this species is uncertain. Currently it is only
known definitely from Halmahera, the Maluku Islands, Queensland and the Northern
Territory (Benoit Dayrat pers. comm. 2016).

**Platevindex** sp. 2 (orange) (Fig. 3)

**Description:** Size to 55 mm in extended
body length (pers. obs.). Body ovate-
elongate in shape, juveniles and subadults
more circular; distinctively flattened.
Notum (= dorsal surface) leathery,
commonly having a very warty apperance
resulting from laterally arranged raised
bumpy ridges; usually dry to the
touch. Foot sole distinctively narrow.
Photoreceptors (dorsal eyes) single,
ever present at edge of notum. Colour
of notum dark brown to brown. Head,
oral lobes and tentacles rarely darker
than underside of body. Foot sole always
distinctively orange colour in adults;
juveniles commonly pale to dark brown.
Hyponotum (= underside of dorsal surface) yellowish-orange in colour, *always* with a
mottled dark colour radiating outwards from foot sole.

**Remarks on preserved specimens:** Disregarding contraction of the body and
some loss of colouration, most of the diagnostic features described above are clearly
distinguishable for this species in preserved specimens (i.e. those preserved in 70% 
ethanol). In particular, the presence of dark mottling radiating outwards from the foot
sole is distinctive and remains distinct in preserved specimens.
Ecology: *Platevindex* sp. 2 is restricted to landward mangrove communities dominated by Smooth-fruitd Spur Mangrove (*Ceriops australis*) in the upper intertidal (pers. obs.). Individuals are commonly observed feeding on the trunks and buttress roots of *Ceriops australis* trees and after spring high tides and rain, individuals are active on the forest floor (pers. obs.). During the dry season, slugs aestivate in the sediment, taking refuge inside crab burrows and crevices at the bases of trees (pers. obs.).

Global distribution: Philippines, throughout the whole of Malaysia, Northern Territory (Benoit Dayrat pers. comm. 2016).

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References


Captain King’s lost weevil – alive and well in the Northern Territory?

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Abstract

The discovery of a ‘hairy’ yellow weevil in Kakadu National Park in 1995, akin to a widely distributed pest species of agricultural crops in South-East Asia (but not Australia), the so-called ‘Gold-dust Weevil’ (*Hypomeces ‘squamosus’*), prompted us to investigate the taxonomy and distribution of this weevil in order to determine the identity and origin of the Kakadu specimen. The ‘Gold-dust Weevil’, whose correct scientific name is *H. pulvinis* (Herbst, 1795), is a sexually dimorphic and variable species and has been described under various names in the literature, but its taxonomy and nomenclature have never been investigated. The results of our research to date indicate that it comprises a complex of closely similar species and that the Australian specimen is not conspecific with those occurring further west and north in South-East Asia. We also found that a female conspecific with the Kakadu specimen was likely collected by Captain Phillip Parker King during his surveys of the northern Australian coast in about 1820 and described in 1826 by W. S. Macleay as *Cenchroma obscura*. King’s weevil has been forgotten for over 200 years, but the discovery of the Kakadu specimen suggests that this species, correctly named *Hypomeces obscurus*, may be present in northern Australia, albeit scarce and seemingly of no current agricultural concern.

Introduction

An unexpected discovery

In September 2009 one of us (RGO) came across a ‘hairy’ yellow weevil (Figs 1, 2) in the insect collection of the CSIRO Tropical Ecosystems Research Centre (TERC) in Darwin. He recognised it as a species of *Hypomeces* Schoenherr, a genus distributed throughout South-East Asia, from eastern India and southern China southwards through Indochina and Indonesia to Timor and New Guinea, but not known to occur in Australia. *Hypomeces* currently comprises about ten species and belongs in the tribe
Tanymecini of the subfamily Entiminae, a large group of typically short-snouted weevils with wide host ranges as adults and soil-dwelling, root-feeding larvae. One species of Hypomeces, named *H. squamosus* (Fabricius) in the literature and ‘Gold-dust Weevil’ in vernacular language, is a major agricultural and horticultural pest in South-East Asia.

Significantly, the specimen in the TEiRC collection (Figs 1, 2) is labelled as having been collected at the Naramu Camp of the former Kapalga Research Station in Kakadu National Park, Northern Territory, in April 1995 by Lyn Lowe, who then participated in a fauna survey forming part of the Kapalga Fire Experiment (Orgeas & Andersen 2001; Andersen et al. 2003). Moreover, the specimen, a male, is in a teneral condition (freshly eclosed), both its mandibles still carrying the deciduous cusp that occurs in Entiminae upon eclosion from the pupal case but breaks off when the weevil starts feeding, and its coating of yellow wax, which grows as the specimen ages and is more prominent in males, is only slightly developed. Its teneral condition and pristine state of preservation indicate that the specimen was collected on the day it hatched from its cocoon and was pinned shortly afterwards, not stored in ethanol as this fluid would have dissolved its covering of wax and matted down its erect silvery setae.

Comparison of the Kapalga weevil with specimens of *Hypomeces* in the Australian National Insect Collection (ANIC) in Canberra revealed that, although similar to the well-known *Hypomeces ‘squamosus’* (an invalid name, see below), it differs in a number of characters from this species and agrees more closely with specimens from Timor. The status of the Timorese taxon is unclear from the literature; it is sometimes treated as a ‘variety’ of *H. ‘squamosus’* but has also been named as a different species. In their recent catalogue of Australian weevils, Pullen et al. (2014) settled on calling it *Hypomeces rusticus* (Weber, 1801), following the distinction made between this and *H. ‘squamosus’* by Marshall (1916) in his scholarly treatment of the weevil fauna of British India. However, Pullen et al. (2014) changed the name Marshall had used for it, *Hypomeces unicolor* (Weber, 1801), to *H. rusticus*, in accordance with a recent correction published by Ren et al. (2013) and necessary due to the fact that Weber’s original name *Curculio unicolor* Herbst, 1795 and hence nomenclaturally
unavailable. For the same reason, *H. squamosus* had to be renamed as *Hypomeces pulviger* (Herbst, 1795) (Ren et al. 2013), an unfortunate but unavoidable change of the name of a well-known pest species. The identification of the Timorese taxon as *H. rusticus* remained somewhat insecure, however, as Marshall (1916) had expressed some doubt about the distinction of this species from *H. pulviger* (as *H. squamosus*), considering the few differences he could find between them to be ambiguous in some cases. Also, there is no recent and proper taxonomic study of the genus *Hypomeces* to verify them. Due to the fresh nature of the Kapalga specimen, Pullen et al. (2014) treated *H. rusticus* as occurring in Australia.

Aims and objectives

In this paper we report the results of further research into the taxonomy and nomenclature of the Kapalga weevil and outline the apparent history of the species in Australia. Although additional study is required (and in preparation) to fully resolve its taxonomic affinities, we here aim to draw attention to the indicated occurrence of this weevil in the Northern Territory and to list and illustrate the morphological differences between it and the more northerly pest species *Hypomeces pulviger*. We hope that this report will assist in the determination of whether this weevil species is established in northern Australia.

Material and Methods

We undertook a morphological study of 113 relevant specimens (including 13 types) of *Hypomeces* from the following collections:

- ANIC — Australian National Insect Collection, Canberra, Australia;
- MAGNT — Museum and Art Gallery of the Northern Territory, Darwin, Australia;
- MMUS — Macleay Museum, University of Sydney, Sydney, Australia;
- NAQS — Northern Australia Quarantine Strategy Entomology Collection, Darwin, Australia;
- NHMD — Natural History Museum of Denmark, Copenhagen, Denmark.

Selected specimens were photographed using a Leica DFC500 digital camera mounted on a Leica M205C microscope, combining (“montaging”) image stacks in Leica Application Suite 4.4 and cleaning and enhancing the final images as necessary in Adobe Photoshop CS3. The genitalia of 15 specimens (mostly males) from different localities were dissected in the standard manner, temporarily stored in glycerine or KY Jelly® and photographed using the same equipment.

Results

Captain King’s lost weevil

No other Australian specimen of *Hypomeces* has been located in any collection so far, but Zimmerman (1993: 667), in his bibliographic notes on William Sharp Maclay,
asserted that a specimen collected during Captain Phillip Parker King’s survey of the northern Australian coast in the 19th century and described by Macleay (1826) as Cencbroma obscura is an “abraded, mislabeled Hypomeces squamosus (Herbst), an Indonesian species and not Australian, which remains in Macleay’s collection”. Zimmerman had examined this specimen, housed in the Macleay Museum in Sydney, and in ANIC’s copy of Macleay’s (1826) paper he scribbled next to description of Cencbroma obscura: “A specimen thought to be the type of this is in the Macleay Mus. It is labeled ‘Cencbroma / obscura / Capt’ King / Australia’. It is an abraded Hypomeces squamosus (Herbst) var. & is therefore a mislabeled specimen.”, and he dated this note as “Z. x. 83”. The author of the species name given by Zimmerman is incorrect, however; it is not Herbst but Fabricius. The addition of the epithet “var.” (variety) in Zimmerman’s note is important as Zimmerman had also labelled the Timorese specimens in the ANIC as “Hypomeces squamosus (Herbst) var.”, indicating that he was aware of their differences from the typical H. ‘squamosus’ (now H. pulviger) occurring further west and north, though regarding them as being conspecific.

The discovery of the Kapalga weevil thus raised the possibility that Cencbroma obscura might be the same species and also that Captain King’s weevil might indeed have been collected in Australia. We were able to borrow from the Macleay Museum the single specimen of Cencbroma obscura as well as two specimens labelled “Hypomeces lanuginosus” and “Timor”, in the handwriting of a former curator of the Macleay Museum, George Masters, who is known to have replaced many original labels with his own (Zimmerman 1993). Macleay (1826) indeed recorded another Cencbroma species from King’s voyages, as C. lanuginosa Dejean. This name had been published by the French Count P. F. M. A. Dejean in a catalogue of the Coleoptera in his collection (Dejean 1821), for a species occurring in Timor. Our study of these specimens revealed the following:

1. the specimen (holotype) of Cencbroma obscura is an abraded female, missing virtually all its scales and setae (Figs 4, 5);
2. its origin is given on its label as “Australasia”, not Australia as recorded by Zimmerman (Fig. 6);
3. the two specimens from Timor labelled *Hypomeces lanuginosus* are males, both with a complete vestiture of pale setae and a thick layer of yellow floccular wax between them (Figs 7, 8); and

4. the three specimens are conspecific with each other and also conspecific with a series of specimens from Timor in the ANIC and with the Kapalga specimen.

**Taxonomic and nomenclatural tangles**

The taxonomic boundaries of *H. pulviger* (formerly *H. squamosus*) have been unclear for a long time, as pointed out a century ago by Marshall (1916), and have not been satisfactorily resolved. The most noticeable difference between *H. pulviger* and the Timorese taxon is the presence of one or more tooth-like projections at the anterolateral corners of the prothorax, behind the eyes, in the former but not the latter (Figs 17, 18). However, as noted by Marshall (1916), these projections are sometimes only feeble in *H. pulviger* and evidently not a reliable distinguishing character. There are, however, a number of other, more consistent differences between the two taxa (Table 1). Although these differences are somewhat relative and difficult to assess when looking at only one species, they become clearer in a side-by-side comparison of both and appear consistent enough to enable a reliable distinction. Whether they are suitable indicators of species delimitations is difficult to assess in the absence of a comprehensive study of specimens from throughout the range of these two taxa, but they are similar to species differences in other entimine weevils and likely to have the same significance in this case too. This conclusion is supported by a preliminary analysis of the ‘barcoding’ fragment of the COI gene of a number of *Hypomeces* specimens, from Malaysia south to Timor, which revealed a clear divergence between the Timorese specimens and those from further west and north (Greg Chandler, pers. comm.), so congruent with the morphological differentiation. Furthermore, comparable differences in these as well as in other morphological characters indicate the existence of additional *Hypomeces* species on the Sunda Islands. A more comprehensive morphological and molecular analysis of additional specimens is in preparation to assess the number of species in this complex and the precise affinity of the Kapalga specimen.

What then is the correct name for the Timorese and Australian specimens? The oldest species name in contention is *rusticus*, which was given by Weber (1801), and also by Fabricius (1801), to specimens collected by the Danish naturalist O. K. Daldorff in Sumatra, probably at Bengkulu (Reid & Beatson 2015). Photos of the two type specimens of *rusticus* in Fabricius’ collection, kindly provided to us by the Natural History Museum of Denmark in Copenhagen, show these to possess a strong prothoracic tooth and thus not to be conspecific with the Australian and Timorese specimens (but apparently representing *H. pulviger*). The next oldest name is *lanuginosa*, which was proposed by Dejean (1821) for a species in Timor but not accompanied by a description and which is therefore unavailable for nomenclatural purposes (it was also never validated afterwards). Next in line of nomenclatural priority is *obscura*, which was established by Macleay (1826) with a proper description and is therefore nomenclaturally available, although it has not been used for almost two centuries. Given the existence of the holotype of *obscura* in the Macleay Museum and its agreement in characters with the Kapalga and Timorese specimens (rather than with *Hypomeces pulviger*), this species is to be named *Hypomeces obscurus* (Macleay, 1826) — the ending of the adjectival species name changing to accord with the different gender of the genus name (*Hypomeces* is masculine, *Cenchroma* feminine).

Table 1. Differences between *Hypomeces obscurus* (Macleay, 1826) and *Hypomeces pulviger* (Herbst, 1795) (see Figs 9–22).

<table>
<thead>
<tr>
<th>Structure</th>
<th><em>Hypomeces obscurus</em> (previously <em>H. rusticus</em>)</th>
<th><em>Hypomeces pulviger</em> (previously <em>H. squamosus</em>)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body scales</td>
<td>colour always creamy; separate from each other</td>
<td>colour usually iridescent green, at least in male; partly overlapping</td>
</tr>
<tr>
<td>Rostrum</td>
<td>longer</td>
<td>shorter</td>
</tr>
<tr>
<td>Admedian linear depressions on rostrum</td>
<td>indistinct, very shallow, straight</td>
<td>distinct, deep, curved</td>
</tr>
<tr>
<td>Antennal scapes</td>
<td>longer</td>
<td>shorter</td>
</tr>
<tr>
<td>Eyes</td>
<td>flatter, less prominent</td>
<td>more acute, very prominent</td>
</tr>
<tr>
<td>Anterolateral corners of prothorax</td>
<td>never tooth-like extended</td>
<td>usually tooth-like extended</td>
</tr>
<tr>
<td>Median pronotal groove</td>
<td>shallow, indistinct</td>
<td>deep, distinct (sharply edged)</td>
</tr>
<tr>
<td>Pronotal impressions</td>
<td>broad, shallow, transverse impression across base of median groove</td>
<td>pair of short, narrower, deeper, irregular longitudinal impressions parallel to median groove</td>
</tr>
<tr>
<td>Elytral bases</td>
<td>more strongly rounded</td>
<td>less rounded, partly straight</td>
</tr>
<tr>
<td>Elytral setae of female</td>
<td>very fine, slightly longer</td>
<td>shorter and thicker</td>
</tr>
<tr>
<td>Penis</td>
<td>shorter, more strongly curved; dorsally more open</td>
<td>longer, less curved; membranous dorsal strip narrower</td>
</tr>
</tbody>
</table>
Discussion

Captain King's voyages and collecting localities

Having clarified the identity and taxonomic status of Captain King's weevil, its origin remains to be determined. Captain Phillip Parker King (1791–1856) was one of the famous Australian explorers of the 19th century. He undertook four voyages around Australia between 1817 and 1822, charged by the British Admiralty and the Colonial Office to survey the north-west coast of New Holland, which his predecessor, Matthew Flinders, had not been able to chart during his circumnavigation of Australia in 1802–1803. The Admiralty thus instructed King to "examine the hitherto unexplored Coasts of [the Continent of] New South Wales, from Arnhem Bay, near the western entrance of the Gulf of Carpentaria, westward and southward as far as the North-West Cape, ...", and specifically to discover "any river or that part of the coast likely to lead to an interior navigation into this great continent." The Colonial Office wanted him "to obtain information" of, i.a., the "general climate ...", the "directions of the mountains ...", the "animals, whether birds, beasts, or fishes; insects, reptiles, &c., ...", the "vegetables ... applicable to any useful purposes, ..." and the "descriptions and characteristic differences of the several tribes or people on the coast" (King 1827).

On his first voyage, from December 1817 to July 1818, King sailed his sole ship, the cutter Mermaid, around the south and west coast of Australia and got as far east, on 26 March 1818, as Braithwaite Point on the coast of western Arnhem Land. He then turned westwards again, exploring the nearby Goulburn Islands and surveying the coasts of the Cobourg Peninsula, Van Diemens Gulf and Melville Island before heading to Timor to reprovision his ship and then returning to Sydney. On his second voyage, from May 1819 to January 1820, he sailed the Mermaid northwards along the Australian east coast, around Cape York and across the Gulf of Carpentaria and explored the Arnhem Land coast from the Wessel Islands to Bathurst Island as well as the Cambridge and Admiralty Gulfs on the Kimberley coast, then ran for Timor again to take on provisions and home to Sydney along the west coast. On his third voyage, from June 1820 to December 1820, he followed the same route, but the Mermaid was "nail-sick" (leaking badly) by then and allowed him little opportunity for exploration, and he limped back from the Prince Regent River mouth to Sydney, this time without replenishing in Timor. On his fourth voyage, from May 1821 to April 1822, he had a new and larger ship, the brig Bathurst, which he again sailed around Cape York and the Gulf of Carpentaria to the Goulburn Islands, but he surveyed and explored mainly the coast of the western Kimberley region south to the Dampier Peninsula, returning to Sydney via Mauritius. Although King failed to find the fabled waterway into the interior of Australia, he explored practically every inlet along the north-western coast of Australia for about 1200 km west of Cape Wessel. King published a two-volume Narrative of his surveys soon afterwards (King, 1827), and a comprehensive and splendid account of his voyages, as well as of the many trials and tribulations he and his crew experienced during them, was published by Hordern (1997).
Captain King was given two marine surveyors, Frederick Bedwell (1796–1853) and John Septimus Roe (1797–1878), to assist him in this task, and the botanist Allan Cunningham (1791–1839) joined him in Sydney. The animals collected by King, Cunningham and Roe on these voyages were studied and described in Appendix B of Volume II of King’s *Narrative*. William Sharp Macleay (1792–1865) studied the ‘Annulosa’, the ringed or segmented animals, the majority (188) being insects, among them 108 beetles (Coleoptera) and among these 20 weevils (Curculionidae) (Macleay 1826). He described nine of the weevils as new, although some turned out to have already been described by earlier authors and others belonged to different genera than those to which Macleay assigned them (Zimmerman 1993). Macleay did not provide the names of the collectors of these beetles or the localities where they were taken, and not all occur along the ‘intertropical and western coasts’ of Australia. The weevil specimens he named *Cenchroma lanuginosa* evidently originated from Timor, not only because this name had been published by Dejean (1821) for a species from Timor (and Dejean was an acquaintance of Macleay) but also because the two specimens with this name in the Macleay Museum carry a label reading “Timor”. King briefly visited the harbour of Kupang in western Timor on his first two voyages, and Cunningham collected specimens (mainly plants) in the vicinity of the town on both occasions (Hordern 1997; Orchard & Orchard 2013). In contrast, the single specimen of Macleay’s *Cenchroma obscura* is labelled as “Australasia”, in Macleay’s hand, suggesting that it was not collected together with the two males from Timor but separately and from somewhere else. But where?

**Looking for a weevil in a haystack**

The name “Australasia” was coined in the 18th century for the lands south of Asia, so encompassing Australia, New Zealand, New Guinea and their neighbouring islands in the Pacific Ocean (but not Timor). In King’s and Macleay’s times, the name “Australia” was not yet established and commonly used for the Australian continent, which was generally referred to as “New Holland” or, as on King’s instructions from the British Admiralty, “New South Wales”. Macleay used both names “Australasia” and “New Holland” on the labels of his insect specimens, the former probably when he was unsure of their exact origin. The holotype of *Cenchroma obscura* is not the only Macleay type labelled as having come from “Australasia”. Among the Macleay types in the ANIC there are another 11 with the same locality name on the label (*Acanthocinus piliger, Callidium erasum, Chrysomela klongjii, Chrysomela nigravaria, Cistela securesfera, Clerus cruciatus, Coccinella kingi, Lycus septemceonas, Lycus rhipidium, Notoclea splendidus, Teleborus pulchellus*), whereas nine others (*Chrysolopus echidna, Chrysolopus tuberculatus, Elater nigrifemoratus, Elater xanthomma, Epholosium reutilminum, Hybanchenia rodulosa, Oedemera punctum, Talaurinus kirbyi, Trox alternans*) are labelled as from “New Holland” instead. Most of the species whose Macleay types are labelled “Australasia” do not occur in Timor but only in Australia, i.e. the coccinellid *Coccinella kingi* (now *Archegleis kingi*; Pope 1989; Ślipiński 2007; Adam Ślipiński, pers. comm.), the cantharid *Teleborus pulchellus* (now *Chauliognathus lagadris* (Fabricius)), the lycids *Lycus rhipidium* and *L. septemceonas* (now both *Porrostoma rhipidium*; Lodislav Bocák,
Hypomeces weevils in Australia

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pers. comm.) and the cerambycids *Acanthocinus pihilger* (now *Rhytidophora piliger*; Adam Ślipiński, pers. comm.) and *Callidium erasum* (now *Pytheus erasus*; Adam Ślipiński, pers. comm.). The exact distribution ranges of the species described by Macleay from King’s material are often not known; some of them are widespread in Australia and others are restricted to the south-eastern or south-western parts, but at least two occur in the Northern Territory and northern Western Australia, i.e. the clerid *Cnthus cruciatus* (now *Orthrius cruciatus*) and the tenebrionid *Cistela securigera* (now *Nocar securigera*). It is thus manifest that most of the beetles described by Macleay (1826) and labelled “Australasia” must have been collected in Australia, and some indeed likely in the Northern Territory or north-western Western Australia, and there is no prima facie evidence that the type of *Cenchroma obscura* was not collected there either.

King and his crew explored almost the entire north-west coast of Australia and went ashore on many islands and points and bays on the mainland, and especially Cunningham collected specimens wherever and whenever he could (Curry et al. 2002). Among the likely places he (or King or Roe) could have taken the type of *Cenchroma obscura* are South Goulburn Island and Sims Island, where Cunningham collected specimens on all four of King’s voyages, and especially the banks of the South Alligator River, which King and Cunningham explored upstream for about 64 km from its mouth on the first voyage and where, on 8 May 1818, they collected near the present site of Kapalga (Curry et al. 2002: Map 8). In his journal Cunningham recorded some plants he encountered there on that day but nothing about any insects, but as his journal entries generally only deal with botanical specimens (Tony Orchard, pers. comm.), this does not mean that he could not have taken such a weevil there. An exact locality for the type of *Cenchroma obscura* can probably never be established, but it is very likely that it was indeed collected along the Northern Territory coast.

No further specimens of *H. obscurus* have been found in Australia to date, despite 25 years of quarantine inspection of numerous locations in the Northern Territory by the NAQS team in Darwin (Glenn Bellis, pers. comm.). A recent search at the Kapalga site also failed to find another specimen, but it was undertaken in July 2015, in the dry season when the parched condition of the vegetation greatly reduces insect activity. The absence of further specimens so far suggests that, if the species is present in the Northern Territory, it may have a restricted distribution and/or occur in very low numbers, and the time of collection of the Kapalga specimen (April) and also of King’s 1818 visit to the site (March) indicate that it may only be active during the wet season.

**Potential impact**

The indicated occurrence of a *Hypomeces* species in northern Australia is important as *H. pulviger* remains a target (under the name *H. squamosus*) of quarantine surveillance efforts in the area (Glenn Bellis & Luke Halling, pers. comm. 2015). This notorious pest (the ‘Gold-dust Weevil’) has a wide range of hosts in South-East Asia. Hill & Abang (2006) recorded it from 42 hosts in Malaysia alone. The highly polyphagous nature of both
adults and larvae can cause significant damage on a number of agricultural crops, the major hosts being rice, maize, sugarcane, cotton and tobacco (Kalshoven, 1981), along with *Citrus* spp. and sweet potato (Hill, 2008). Other hosts include cocoa, coffee, durian, guava, jackfruit, long-bean, mango, rambutan and sapote (Muniappan et al. 2012), and additional ones are listed, together with a summary of the weevil’s impact on crops and additional references, on CABI’s Plantwise Knowledge Bank (http://www.plantwise.org/KnowledgeBank/Datasheet.aspx?dsid=27783). In contrast, little information exists about the hosts of *H. obscurus* in Timor. Specimens in the ANIC have been collected on Pigeon Pea (*Cajanus cajan*, Fabaceae) and Jujube or Chinese Apple (*Ziziphus mauritiana*, Rhamnaceae) in West Timor, and it has been found defoliating mango and was also taken on guava, maize, long-bean, peanut, sweet potato, sorghum, cucumber and rice (Glenn Beilis, pers. comm.). This host range suggests that *H. obscurus* may also be able to feed on a variety of plants (both native and cultivated) in Australia if it is established here now or in the future.

**Conclusions**

Our intricate sleuthing work revealed that Lyn Lowe, quite unbeknown to her, succeeded in rediscovering Captain King’s lost weevil in the Kakadu National Park and that the name William Sharp Macleay gave it, forgotten in the scientific literature for almost 200 years, is in fact valid. While it seems impossible to determine the exact locality where King and his party may have collected this specimen nearly 200 years ago, King and his botanist, Allan Cunningham, did collect specimens in the vicinity of Kapalga, the site where Lyn Lowe took a freshly hatched male in 1995. As far as currently known, *Hypomeces obscurus* occurs mainly on Timor, and it is not the same species as *Hypomeces pulviger* (formerly *H. squamosus*), the notorious “Gold-dust Weevil” (a misnomer as its colour is neither golden nor due to dust). Further collecting efforts at the Kapalga site as well as in similar habitats elsewhere in Kakadu National Park and other parts of the Northern Territory are needed to confirm the presence of *H. obscurus* in Australia and verify whether King’s lost weevil is indeed alive and well in the Northern Territory. Such confirmation would indicate that the species is either native to Australia or was transported there by humans (e.g. by Indonesian fishermen) at least two centuries ago and has been established for a considerable time.

**Acknowledgements**

We sincerely acknowledge a range of associates who helped us in this complex investigation to discover the status and name of King’s lost weevil: Alan Andersen (CSIRO, TERC, Darwin) and Lyn Lowe (Charles Darwin University, Darwin) for validating the collecting details of the Kapalga specimen; Robert Blackburn and Jude Philp (MMUS) for the loan of King’s original specimens; Gavin Dally, Graham Brown and Richard Willan (MAGNT) and Glenn Bellis and Stacey Anderson (NAQS) for the loan of *Hypomeces* specimens from their collections; Miguel Alonso-Zarazaga (Museo Nacional de Ciencias Naturales, Madrid, Spain) for the names and locations of several other specimens; Sree...
Selvantharan (NHMD) for the excellent photos of critical Weber and Fabrician types; Glenn Bellis, Stacey Anderson and Greg Chandler (NAQS) for a preliminary molecular analysis of a number of critical specimens and for various pieces of information on Hypomeces; Luke Halling (Australian Quarantine and Inspection Service, Cairns) for information on Hypomeces pest species; Adam Ślipiński (CSIRO, ANIC) and Ladislav Bocak (Olomouc, Czech Republic) for information on the distribution of beetle species described by Macleay; Tony Orchard (Canberra) for information on Allan Cunningham’s records and journals; Ted Edwards and Russell Barrett (CSIRO NCRA) for important literature; John Westaway (NAQS) and Rachel Martin (Parks Australia, Kakadu) for help in searching for further specimens at the Kapalga site.

References


Rediscovery of the Spinifex Sand-skipper (*Proeidosa polysema*) in the Darwin area, Northern Territory

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Abstract

The Spinifex Sand-skipper (*Proeidosa polysema*) (Lepidoptera: Hesperiidae) is recorded from two sites near Noonamah-Berry Springs, approximately 28 km south-east of Darwin. The species is recorded breeding on the grass *Triodia bitextura* (Poaceae) growing in eucalypt open-woodland in sandy soil derived from laterite. The butterfly had not been recorded from the Darwin area for more than a century (since 1909) and its presence in the rural area confirms earlier historical collections made by renowned entomologist F.P. Dodd.

Introduction

The Spinifex Sand-skipper (*Proeidosa polysema*) (Fig. 1), has a wide distribution across the northern half of Australia where it occurs in hummock open-grassland on sand dunes and eucalypt open-woodland with a hummock/tussock grass understorey on sand and dry rocky sandstone, preferring shallow gullies and slopes of hills (Braby 2016). The larvae (Figs 3–5) specialise on a limited range of perennial ‘soft’ resinous spinifex tussock-forming grasses (*Triodia* spp.) (Poaceae), which in the Top End of the Northern Territory include *T. microstachya* (Common & Waterhouse 1981) and *T. bitextura* (Braby 2015). The larva also pupates in the final instar larval shelter.
Figs 1–5. Proeidosa polysema showing: 1. adult male hilltopping on sand dune in central Australia at Curtin Springs, Northern Territory; 2. larval shelter on Triodia bitextura at Wongalara Wildlife Station, Northern Territory; 3. early instar larva on T. microstachya at Fish River Station, Northern Territory; 4. final instar larva on T. microstachya at Fish River Station, Northern Territory; 5. final instar larva in diapause on T. pungens at Curtin Springs, Northern Territory. (Michael Braby)

(Meyer et al. 2006). Historical records further north in the Darwin area ('Port Darwin') have been dismissed or considered to require confirmation because of the lack of subsequent records for over a century. Indeed, Meyer et al. (2006: 15) stated that: “We believe it is unlikely that it will be encountered there in the future, due to a lack of suitable habitat.”

Waterhouse (1933) and Meyer et al. (2006) reviewed the historical literature records of P. polysema from the Darwin area and noted that at least five specimens were collected from Port Darwin by F.P. Dodd at the turn of the twentieth century: two males (both paratypes) in February 1909, other males in January and March, and a female in April. It has generally been assumed that the material was collected further south, possibly along the railway line such as between Adelaide River and Pine Creek, and that Port Darwin was the location at which the specimens were processed. In this note, we record an extant population of P. polysema from the outer Darwin rural area. Prior to our rediscovery of
the species near Darwin, the butterfly had not been recorded from the area for more than a century.

**Observations**

At the intersection of Finn Road and Middle Arm Road, approximately 6.5 km north of Berry Springs (12.6451°S, 131.0099°E), an adult of *Proeidosa polysema* in ‘fresh’ condition

**Figs 6–11.** Ecology of *Proeidosa polysema* near Berry Springs-Noonamah, Darwin: 6. breeding habitat comprising eucalypt open-woodland with a grassy understorey dominated by *Triodia bitextura*. 7. larval food plant *Triodia bitextura*. 8–11. adult butterfly being eaten by preying mantis. (Michael Braby)
Braby & Westaway (according to extent of wing wear) was observed just after midday feeding on the flowers of *Spermacoce* sp. on 14 February 2015. The specimen was observed only briefly before it was disturbed; it rapidly flew off and could not be relocated despite extensive searching. However, another adult was subsequently located nearby and photographed; however, this particular individual was being eaten by a preying mantis (Figs 8–11). After this insect predator had completed devouring its meal, two of the wings of the butterfly were recovered and retained as vouchers (accession number MFBC 00934, Australian National Insect Collection). The identity of the butterfly can be clearly discerned by the uniform brown ground colour to the wings and the presence of a series of eight large white spots on the underside of the hindwing (Fig. 8). The habitat in which these observations were made comprised an open disturbed area of pioneer plants adjacent to the railway line. There were no signs of the putative larval food plant (*Triodia bitextura*) growing in the immediate vicinity.

Two months after these initial field observations, we searched the adjacent woodland for presence of the larval food plant and likely breeding areas of the butterfly on 26 April 2015. On Middle Arm Rd, approximately 6 km north-north-east of Berry Springs and 6.4 km west-south-west of Noonamah (12.6456°S, 131.0233°E), we found *Triodia bitextura* (voucher JOW 4802, Darwin Northern Australia Herbarium) growing in abundance in open-woodland with a dense grassy understorey in sandy soil derived from well-drained laterite on relatively flat terrain (Fig. 6). On one particular tussock (Fig. 7), two old pupal shelters of *P. polysema* were recorded and collected, confirming the presence of an extant breeding site. This site was located approximately 1.5 km east of the site where adults were initially recorded earlier in February 2015. The breeding habitat was located approximately 28 km south-east of Darwin.

**Discussion**

Our discovery of *Proeidosa polysema* from the Darwin area near Noonamah and Berry Springs validates the historical records from 'Port Darwin' by Dodd as being reliable. Moreover, a distribution map of the larval food plant *Triodia bitextura* for the Darwin area (Fig. 12) shows that this plant is widespread but uncommon in the Darwin rural area, extending as far

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Fig. 12. Distribution map of *Triodia bitextura* in the Darwin area, together with the two extant sites of *Proeidosa polysema* (★). Symbols for plant data are as follows: • vouchered herbarium specimens, ▲ survey observations. (Flora and Fauna Division)
Spinifex Sand-skipper rediscovered

north as Gunn Point. In our experience, *T. bitextura* has a scattered occurrence and in some areas it may be locally abundant. *Triodia bitextura* (Fig. 7) does not exhibit the large hummock forming habit typical of ‘hard’ spinifex species, and as such it is less conspicuous. Moreover, it is not confined to sandstone outcrops, which may explain the belief of Meyer et al. (2006) that the butterfly does not occur in the Darwin area due to lack of suitable habitat. At the Noonamah-Berry Springs site the butterfly was breeding on plants growing on sandy soil derived from laterite in contrast to the general tendency of *P. polyxena* to breed on sand dunes and sand derived from rocky sandstone. The spatial distribution of *T. bitextura* suggests the butterfly may well occur in other locations near Darwin, particularly in the rural area to the south and south-east of Palmerston (Fig. 12). However, little habitat now remains closer to the city of Darwin, but it seems highly plausible that in 1909 Dodd collected the original specimens not far from his base camp in Parap (see also Braby & Nielsen 2011 for an account of Dodd’s collecting sites in the Darwin region), rather than along the railway line to the south of Adelaide River.

**References**


When I was asked to review Dr Grahame Webb's (2015) book *Wildlife Conservation: In the Belly of the Beast*, I was a bit uneasy. Cecil, the tame old lion at Hwange National Park in Matabeleland North, Zimbabwe, had just met an untimely end and I had heard Dr Webb’s name associated with trophy hunting. But having come to grips with the book, I must say it’s a valuable contribution to conservation and the issues that surround it.

Dr Webb writes that if his book “stimulates others to think in more depth about conservation, or helps them better understand and appreciate how bio-politics can enhance or constrain conservation” then his main goal in writing the book will be achieved.

Well I think he can rest assured on that point – the book contains a plethora of examples of rational science being ignored in favour of ‘bio-politics’ – the rarely-mentioned wheeling and dealing that goes hand in hand with a lot of conservation decisions.

For instance, the ‘sea turtle conservation community’ estimated the total Caribbean Hawksbill Turtle population as 5000, even though Cuba harvested 5000 adult turtles every year. Armed with reasonable science showing the harvest was sustainable, Cuba applied to CITES (the Convention on International Trade in Endangered Species) to export 500 turtle shells to Japan. It was blocked by the United States of America and conservation interests.

Dr Webb makes a compelling case that the United States was motivated by political factors like blockading Cuba and getting the votes of expatriate Cubans living in the United States. Of more concern is the conclusion that conservation groups were desperate to continue using the Hawksbill as one of their iconic fundraising species – something that would be less than convincing if it were admitted that Hawksbills were thriving in Cuba as part of a sustainable use program.

You might argue that legal trade in secure Cuban Hawksbills would be the best cover for illegal trade in endangered populations from elsewhere, but prohibition on trade in wildlife hasn’t exactly been successful – both Tigers and Black Rhinoceros have come much closer to extinction after international trade was banned.

Cuba eventually caved in to the pressure and banned the Hawkshill harvest – bringing a 500 year old tradition to an end. The World Wildlife Fund is providing the turtle fishing communities with “sustainable economic alternatives” (http://www.wwf.ca/about_us/successes/hawksbill/). I can’t help wondering if they will support the communities in the long term.
One of the most iconic conservation issues is that of elephants, and the ivory they produce. Dr Webb points out that “given the opportunity, wild elephants will continually multiply until they ultimately destroy the habitat in which they live.” I’ve seen a graphic demonstration of this in Kenya’s Amboseli National Park where a few remaining monkeys clung pathetically to tree stumps – the last remnants of a woodland eaten out by elephants.

Dr Webb argues that instant death from culling is far more humane than a slow, agonising end from starvation. He further points out that some of the culling can be done by controlled trophy hunting, with the resulting revenue benefiting local communities – who then value the animals as a source of income, rather than seeing them as an agricultural pest.

This may be a distasteful argument, but it also seems to be a compelling one – provided the controls on the program ensure a humane and sustainable harvest.

A similar point can be made with trophy hunting of crocodiles in the Northern Territory.

Dr Webb’s personal involvement in the commercial aspects of crocodiles gives him a valuable perspective on the sometimes perverse results of the costs of wildlife regulation.

I guess the government position is that any industry using a protected species should fund the regulatory regime that ensures the species is harvested sustainably and humanely. But in the case of crocodiles, the costs of CITES compliance could be having a perverse impact on the species.

For example, a crocodile has 66 teeth, each of which you can sell for between $5–$10, once you have cleaned, drilled and mounted it on a leather thong.

But to sell that crocodile’s teeth overseas, each tooth needs a separate export permit from Australia and in some cases, an import permit from the destination. Each permit takes about 40 working days and about $60 in fees and costs. So to sell the teeth from one farmed crocodile, could take nearly $8000 in permit fees. OK, the skin is the main product, but one impact of this cost structure is to favour agriculture over wildlife production – with consequent damage to natural ecosystems.

Another likely impact of this cost structure is to drive the trade underground – if it’s impossible to comply with the costs of CITES, then people will look for underground ways to sell their produce – especially in hard-pressed third world countries.

I can’t help thinking that at the heart of this book is the clash of the generations. On the one hand, Dr Webb provides compelling rational arguments for the controlled commercial use of wildlife, but you can hear his (mostly younger, urban) opponents saying how they hate the idea of killing anything and especially dislike firearms.

Perhaps this difference stems from today’s society being separated from the realities of despatching farm animals for food. I suspect that our farming grandfathers would find
current attitudes to wildlife rather inconsistent — at least for those of us who eat meat or fish.

And speaking of attitudes, one of the inescapable lessons from this book is that unfortunately, scientific facts frequently come a poor second to emotion when it comes to why people adhere to particular beliefs.

Emotional responses often come from experiences. So politicians probably won’t have any personal support for a cause unless they have had a relevant experience. And even when they have, they won’t take action unless it is OK with the electorate. Which is why using the media to get grass roots support for something is so important.

And also why the most prominent conservation and animal welfare organisations are the most expert at using the media.

In this controversial book, Dr Webb applies scientific method to the field of conservation and often finds it wanting.

The book is full of examples of conservation decisions taken for political reasons against scientific evidence — it’s a thought-provoking work — and a scary read! Everyone should have a look at it.

Reference

Advice to authors

The *Northern Territory Naturalist* publishes works concerning any aspect of the natural history and ecology of the Northern Territory or adjacent areas of northern Australia. It is a registered, peer-reviewed journal (ISSN 0155-4093) for original research. Contributors include a range of field naturalists and scientists who do not have to be members of the Northern Territory Field Naturalists' Club Inc.

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