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Ecology and conservation of the northern hopping-mouse (*Notomys aquilo*)

Rebecca L. Diete^{A,E}, Paul D. Meek^{B,C}, Christopher R. Dickman^D and Luke K.-P. Leung^A

Abstract. The northern hopping-mouse (*Notomys aquilo*) is a cryptic and enigmatic rodent endemic to Australia's monsoonal tropics. Focusing on the insular population on Groote Eylandt, Northern Territory, we present the first study to successfully use live traps, camera traps and radio-tracking to document the ecology of *N. aquilo*. Searches for signs of the species, camera trapping, pitfall trapping and spotlighting were conducted across the island during 2012–15. These methods detected the species in three of the 32 locations surveyed. Pitfall traps captured 39 individuals over 7917 trap-nights. Females were significantly longer and heavier, and had better body condition, than males. Breeding occurred throughout the year; however, the greatest influx of juveniles into the population occurred early in the dry season in June and July. Nine individuals radio-tracked in woodland habitat utilised discrete home ranges of 0.39–23.95 ha. All individuals used open microhabitat proportionally more than was available, and there was a strong preference for eucalypt woodland on sandy substrate rather than for adjacent sandstone woodland or acacia shrubland. Camera trapping was more effective than live trapping at estimating abundance and, with the lower effort required to employ this technique, it is recommended for future sampling of the species. Groote Eylandt possibly contains the last populations of *N. aquilo*, but even there its abundance and distribution have decreased dramatically in surveys over the last several decades. Therefore, we recommend that the species' conservation status under the *Environment Protection and Biodiversity Conservation Act* 1999 be changed from 'vulnerable' to 'endangered'.

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Introduction

The northern hopping-mouse (*Notomys aguilo*) is regarded as a cryptic and enigmatic species. Within a genus of largely arid-dwelling rodents, N. aquilo is the only extant species in Australia's northern, monsoonal tropics. In determining the distribution, habitat requirements and conservation status of N. aquilo, Woinarski et al. (1999) relied solely on indirect signs of its presence because trapping, spotlighting and predator scat studies failed to detect it. The type specimen was reportedly collected from Cape York (Woinarski et al. 1999); however, the current species distribution includes only Groote Eylandt and a small area in coastal Arnhem Land (IUCN 2008). An absence of recent mainland records (Woinarski et al. 2014) suggests that Groote Eylandt possibly holds the last remaining extant populations of the species. N. aquilo is currently listed as 'vulnerable' under the Environment Protection and Biodiversity Conservation (EPBC) Act 1999 (Woinarski et al. 2014). However, on the IUCN Red List, it is regarded as 'endangered' because of its small and declining distributional range as well

as declines in the quality and extent of its habitat (Woinarski 2008).

An important behaviour of hopping-mice is the construction of 'spoil heaps', which are piles of soil that mark the original entrance to their burrow systems. This initial entry point is subsequently back-filled from inside, and straight, narrow shafts known as pop holes are then used as the permanent entrances. Spoil heaps are visible for many months after burrow completion (Diete *et al.* 2014) and have been used in surveys as the sole indicator of the presence and abundance of *N. aquilo* (Firth 2008; Smith 2009; Ward 2009). However, recent evidence confirmed that spoil heaps are also made by the sympatric delicate mouse (*Pseudomys delicatulus*) and these cannot be reliably distinguished from those of *N. aquilo* (Diete *et al.* 2015a). These findings have cast uncertainty on all previous records of this species based on spoil heap surveys.

Other types of sign have been used in surveys of *N. aquilo*, including burrow pop holes and identification of the unique bipedal tracks left by the animals (Woinarski *et al.* 1999).

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Recently, camera traps were used successfully to record *N. aquilo* during burrow construction (Diete *et al.* 2014) and were demonstrated to be a viable alternative to indirect signs for detecting the species (Diete *et al.* 2015*b*). The species is known to be trap-shy towards box-style traps such as Elliott traps (Woinarski *et al.* 1999); however, other *Notomys* spp. can be captured in deep (600 mm or greater) pitfall traps (Predavec 1994; Moseby *et al.* 2006; Read *et al.* 2015). This type of pitfall trap has not yet been used on Groote Eylandt and an agreed standardised survey method for *N. aquilo* has yet to be developed.

The life history of *N. aquilo* has mostly been determined from the observations of Thomson (in Dixon and Huxley 1985), who kept a captive colony of the species in the 1940s. Moreover, understanding of the species' ecology is largely inferred from ecological and biological extrapolations of some of its better-studied congeners such as the spinifex hoppingmouse (*N. alexis*). It is unknown to what extent *N. aquilo* may be ecologically and behaviourally divergent from its relatives given the vastly different climatic and habitat characteristics of the distributional range of *N. aquilo* compared with those of other hopping-mice. Poor understanding of *N. aquilo* and conflicting data due to the confused identification of signs have hampered conservation efforts for this species.

This study aimed to fill key gaps in our knowledge of the ecology and conservation of *N. aquilo* by: (1) determining whether the species can be effectively sampled by pitfall trapping and/or camera trapping; (2) describing life-history traits such as breeding events, seasonal activity and morphology; and (3) describing the home range, habitat selection and burrow use of the species. The new methodological and ecological knowledge gained from this study are used to recommend more refined and appropriate conservation strategies for *N. aquilo*. Importantly, we review the current conservation status of this species on the basis of our findings.

Materials and methods

Study site

Groote Eylandt is Australia's fourth largest island (2378 km²) and is located in the Gulf of Carpentaria, Northern Territory. Rainfall is strongly seasonal, with 95% of the annual precipitation falling between November and April (Bureau of Meteorology 2015). The island is an Indigenous Protected Area with a part of it being leased for the exploration and mining of manganese. Groote Eylandt is a unique environment for Australian wildlife research due to the absence of most exotic vertebrates such as ungulates, lagomorphs, foxes and cane toads, as well as large parts of the island being undisturbed by agriculture and urban development. The island and the surrounding smaller islands are, therefore, significant habitat for the preservation of up to 12 threatened animal and plant species, many of which have declined on the adjacent mainland (Department of Natural Resources, Environment, the Arts and Sport, undated). There is no road access to much of the island, which limits accessibility for field research.

Areas surveyed using the methods described below were plotted on a habitat map created by collating substrate and habitat information from Crase and Hempel (2005), satellite imagery (Google Earth 2013) and ground truthing (Fig. 1). This

identified that large areas of the island's interior are either on laterite soils or sandstone (Fig. 1). Although laterite is an ambiguous term, it is characterised on Groote Eylandt by 20–30 cm of iron-rich gravel (Crase and Hempel 2005). The remaining habitat could not be accurately mapped using these methods, but it consists of a combination of woodland, shrubland, mangroves, heath, swamp, dunes and closed vine forest on either sandy, loam or clay soils.

Searches for signs, opportunistic camera trapping and spotlighting

Searches for signs of N. aquilo were conducted in potential habitats (those with a sandy topsoil that allows digging and burrowing) across as much of the island as could be practically accessed from May 2012 to December 2014, and in July 2015. A basic search involved two people either in a slow-moving $(\sim 10 \,\mathrm{km}\,\mathrm{h}^{-1})$ vehicle, or on foot, looking for spoil heaps, which were defined as piles of soil 30 cm or more in diameter with no obvious opening whence the soil had been displaced. Vehicle searches were conducted in areas of very open, low ground cover, otherwise they were conducted by walking through potential habitat looking for signs. When spoil heaps were identified, we attempted to locate pop holes and tracks of N. aquilo, using the descriptions of Woinarski and Flannery (2008). When possible signs of the species were found, camera trapping was conducted to determine whether N. aquilo was present. Scoutguard 560DF, Scoutguard 565F or Reconyx HC550 (all with white flash illumination) camera traps were set within 3 m of the potential signs, or, if signs were prolific, in a transect through the area for 2-7 nights. Depending on the abundance of signs, 2-8 camera traps were used at any time, set horizontally on a tree or star picket 20 cm from ground level and baited with sunflower seeds placed 1.5 m from the camera trap.

Spotlighting in potential habitat was carried out at night by two people searching either from a slow-moving (15– $20\,\mathrm{km\,h^{-1}}$) vehicle or on foot using a \geq 200-lm torch and/or the vehicle headlights while watching the track. Spotlighting was conducted for two or three consecutive nights. The distance sampled depended on the size of the area where potential hopping-mouse signs were found during the day. Line transects ranged from 1 km to 15 km in length with 20–60 min spent at each site per night. In areas where trapping grids were established, this occurred on four occasions throughout 2014; otherwise, it was usually conducted once, when land could be accessed throughout the study.

Live trapping

Live trapping was first conducted in April 2013 at the Cave Paintings area (CP) (13.9705°S, 136.5025°E), where the presence of *N. aquilo* was confirmed by spotlighting and camera traps. A preliminary trial was conducted by setting 12 Sherman 'Type A' traps (H.B. Sherman Traps Inc., Florida) around three burrows with fresh spoil heaps (four traps per burrow). These were baited with either sunflower seeds or peanut butter and oats in an alternating allocation. After only one night of trapping, an adult female *N. aquilo* was captured in a trap baited with sunflower seeds. However, all subsequent attempts

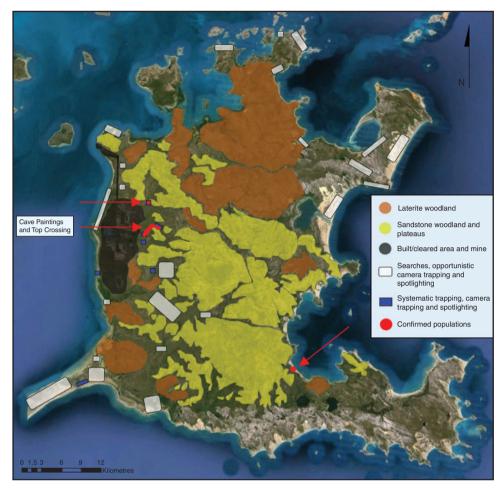


Fig. 1. Map of Groote Eylandt indicating areas where *N. aquilo* was confirmed as present as well as areas searched and trapped. Black, orange and yellow shading indicate areas inherently unavailable to *N. aquilo* as habitat. Searches were conducted in one site that was later cleared for mining.

with these traps failed to capture any more *N. aquilo* and, therefore, this trapping method was discontinued.

In May 2013, at CP, 90 pitfall traps were installed in a grid 660 m long and 120 m wide (8 ha). Three different-sized pitfall traps were trialled. Two of these were constructed from PVC storm-water pipe, 65 cm deep, with diameters of 22.5 cm and 15 cm respectively. The third pitfall type was a 20-L plastic bucket (40 cm deep, 29 cm wide). The traps were set 30 m apart, alternating by trap type, each with a 10-m-long, 40-cm-high drift fence made of aluminium insect screening. After trapping in this configuration twice, the 30 bucket traps were removed and the remaining 60 PVC pitfalls were used in five more trap sessions. In August 2013, another grid in the original configuration of 90 pitfall traps was set in coastal grassland on the south-west peninsula of the island. In April and May 2014, five smaller trapping grids of 30 PVC pitfall traps (15 of each type) were set with traps 30 m apart in rows of three or four. These were trapped 1-4 times until December 2014. The trapping was conducted for 5-7 consecutive nights. Pitfall traps were checked every morning at dawn. Captured N. aquilo were weighed and measured for the length of the combined head and body, hind foot, ear and tail. Sex and breeding data recorded

were scrotal width, lactation, pregnancy (via palpation of the abdomen) and the vagina was scored as either imperforate, perforate or open. To mitigate for the variation in measurements that can occur between researchers, all *N. aquilo* were processed by the same person (RLD). Each individual was uniquely marked using ear notches and released at the point of capture.

Standardised camera trapping

Four camera traps were set 50 m apart along a straight line through each live-trapping grid for 14 consecutive days starting at the end of each live trapping session. At each camera trap station, a Reconyx HC550 (LED white flash) camera trap was mounted in a vertical, overhead orientation using a bookshelf bracket attached to a star picket that was driven into the ground so that the camera trap was 1.5 m from the bait holder positioned directly below. Each station was baited with sesame oil soaked into cotton balls, placed inside a stainless steel tea infuser pegged to the ground. This method proved effective at detecting the species in preliminary trials (Diete *et al.* 2015*b*). The camera trap's field of view (~1 m) was cleared of vegetation and leaf litter to increase visibility of animals and to reduce false triggers.

Camera traps were set on fast shutter, one image per trigger with no delay between triggers. The bait was renewed after seven days.

All animals recorded in camera trap images were identified to species when possible. As *N. aquilo* was rarely photographed more than 5 min after the previous detection of the species on the same night, the delineator between independent detections ('events') was set at 5 min, meaning that successive photos of the species less than five minutes apart were classed as one event.

Radio-tracking

Nine adult *N. aquilo* (five females and four males) captured in pitfall traps at CP were fitted with radio-transmitting collars and tracked for 9–19 consecutive days (mean = 14 days). A single-stage VHF transmitter with a 10-cm whip antenna was attached to each animal's neck using a zip-tie collar (model ZV1C 105, Sirtrack Ltd, Havelock North, New Zealand). A small piece of reflective tape was attached to the transmitter to aid visibility of the animals by torch light. The collar and transmitter configuration weighed between 1.6 and 1.9 g and was less than 5% of the animal's body weight. The battery life of each transmitter was governed by a predetermined programmed pulse rate of 40 ppm for ~30 days.

All collared animals were adults in breeding condition, which was defined as previously mated (perforate vagina) for females or the testes being scrotal for males. Three different individuals were tracked during each trap session in May, August and December 2013. Animals were collected from traps in the morning, collared during the day, and released at the point of capture in the evening. Low capture rates and the larger body weight required prevented radio-tracking more animals concurrently. Radio-tracking commenced at least 2h after animal release, with only one fix on the release night, and continued each night between 19:00 and 06:00 hours. Collared animals were located on foot with a folding, three-element, Yagi antenna and an Ultra® (Sirtrack Ltd) narrow-band receiver. Radio-tracking commenced at different times each night to ensure that the whole nocturnal period was sampled evenly. Three or four fixes were obtained per animal per night with each fix ~1-2 h apart. Additionally, one fix was obtained during daylight hours on most days to determine the location of burrows.

When animals or burrows were located, the positions were recorded with a hand-held GPS, and microhabitats were scored. An average value for each microhabitat characteristic was visually approximated for a 3×3 m area around the animal or burrow. Ground characteristics assessed were ground cover, woody debris, bare ground and leaf litter. Each characteristic was allocated a proportion so that the total of these characteristics was 100% on each occasion. Bare ground and leaf litter were classed as 'open' microhabitat while ground cover and woody debris were classed as 'closed' microhabitat. Ground cover height, canopy cover (%), shrub canopy cover (%) and shrub height were also assessed but not subsequently used in the analysis of data as they appeared to have little explanatory or predictive power for presence of N. aguilo. Animals could usually be detected directly by sight, sound or by pinpointing to a structure such as a log. When observed directly, we noted

whether it was using a 'closed' (under/amongst ground cover or woody debris in a way that visually obscured its body) or 'open' (moving or resting in the open, not using cover or structures) feature of that microhabitat. Removal of radio-collars was attempted at the end of each tracking session using a butterfly net to catch the animals. Five of the nine individuals were captured with this method. Two animals could not be recaptured and two collars became unattached for two other individuals.

Burrow measurements

Some burrows could be determined as belonging to *N. aquilo* either by camera trapping during construction (Diete *et al.* 2014), or by radio-tracking animals to the burrow. At each burrow, we recorded the length, width and height of the spoil heap, the number and diameter of visible pop holes and distance from the centre of the spoil heap to each pop hole.

Data analysis

Individuals were classified as adult if they equalled or exceeded the head and body length of the smallest scrotal male or perforate female depending on the sex of the individual. Body weight (pregnant females omitted), head and body length, hind foot length, tail length and ear length measurements for adult N. aquilo were compared between the sexes using two-sample t-tests. Adults were considered to be in breeding condition if they had descended testes (scrotal) (males) or if they were pregnant, lactating or had an open vagina (females). Data from 2013 and 2014 were too few for seasonal comparisons and were therefore pooled across the two years. Fisher's Exact Tests were used to compare the proportion of adult males and females in breeding condition in the wet and dry season as well as the proportion of juveniles in the population for each season. Wet season was defined as the months of November-April, and dry season May-October.

A body condition index was calculated for all individuals except pregnant females by dividing the ln(body weight) by the ln(head and body length). This index is a good predictor of body fat in rodents (Labocha *et al.* 2014). A general linear model was used to determine the best predictors of body condition. Factors selected in the analysis were a combined age and breeding class of juvenile, non-breeding adult or adult in breeding condition as well as sex (male/female) and season (wet/dry). Interactions between these factors were tested but none were significant and hence were dropped from the model. Tukey pairwise comparisons were used for *post hoc* tests. The ratio of measurements on the natural logarithmic scale could not be back-transformed to the original scale for presentation.

Standardised camera trapping was implemented in 2014 after methodological trials. Because of the short sample period, these data were not analysed for seasonal changes. A two-sample *t*-test was conducted to determine whether the mean number of events differed between the two sites where *N. aquilo* were captured. The number of camera trap-nights at these two sites were identical. A Pearson correlation was computed to determine whether the number of camera trap events per 100 trap-nights correlated with the minimum number of *N. aquilo* known to be alive (MNKA) from pitfall trapping the same area. For CP, where *N. aquilo* was trapped consistently, this analysis was

conducted for each successive day of camera trapping to determine the number of days after which the correlation became significant.

Home ranges for all radio-tracked individuals were estimated using Ranges 9 (Kenward et al. 2014). Incremental area analyses of location fixes showed that the range area used by each individual stabilised at ~20 fixes, thus demonstrating that the data collected were sufficient to draw conclusions on home range (Kenward et al. 2014). Individuals were noted to spend all of the day and a substantial portion of the night at unpredictable times in their burrows. Therefore we used only one fix per burrow, ignoring subsequent fixes when the animal was below ground in the same burrow to prevent home ranges being skewed by periods of inactivity (Kenward 1985, 2001). We termed the subsequent range estimation the 'active home range'. Fixes at burrows when the animals were underground were used to calculate the proportion of the nocturnal period spent in burrows.

Kernel contours at 50% and 95% were constructed in Ranges 9. We found that using the default fixed smoothing multiplier of 1.0 led to overestimates of active home ranges in areas where animals were not observed (sandstone woodland: Fig. 2). Reducing the smoothing factor to 0.88 gave tighter contours around the rocks without breaking the 95% contours into separate areas. We investigated differences in active home-range size and proportion of nocturnal period spent in burrows between the sexes using Kruskal–Wallis tests.

To determine whether animals were using open or closed microhabitat features in proportion to their availability, or were selecting some features over others, we calculated the proportion of open microhabitat within each individual's home range. This was achieved by summing the average leaf litter and bare ground percentages from the microhabitat analyses for each individual. Using the ratio of proportion of open habitat used to that

available, we calculated how many times the individual ratio was greater than 1.0; that is, how many individuals used proportionally more open habitat than available. This was analysed with a test for one proportion. A Kruskal–Wallis test was conducted to determine whether these proportions differed between the sexes. Except for home-range analyses, all statistical analyses were conducted in Minitab® (Minitab Inc. 2015).

Results

Searches, spotlighting and opportunistic camera trapping

Approximately 224 search hours, 132 spotlighting hours and 870 opportunistic camera-trap-nights were completed in 32 separate locations across Groote Eylandt during 2012-15. A greater number of sites were searched in coastal grassland habitat; however, search and trap effort in sandy woodland habitat was approximately twice that of grassland habitat (Table 1). N. aquilo was confirmed to be present in three of these locations (Fig. 1). In one location, the species was first detected by spotlighting with vehicle headlights and then with camera traps. In the two other locations, likely signs of N. aquilo were detected during area searches followed by confirmation with camera traps. In areas where the species' presence was confirmed, straight, deep pop holes >2.5 cm in diameter were also found associated with spoil heaps. Pop holes of this size were not located in areas where camera trapping was unsuccessful. Camera traps next to six inhabited burrows (as determined by radio-tracking or recording burrow construction) successfully detected N. aquilo within four days. All signs of N. aquilo activity as well as the active home ranges (see below) were on sandy soils (Figs 1, 2).

Live trapping

Pitfall trap-nights totalled 7917, including 6703 trap-nights with tube-type pitfall traps that were effective in trapping

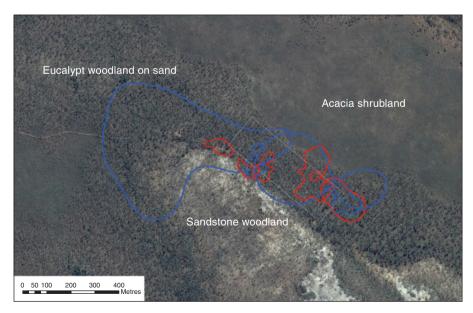


Fig. 2. An aerial image of the Cave Paintings study site showing the trapping grid (white rectangle) and the 95% estimated home range of male (blue) and female (red) *N. aquilo*. The proximity of the three distinctly different broad habitat types illustrates the selection of eucalypt woodland on sand by *N. aquilo*.

Table 1. Summary of total survey effort for *N. aquilo* across Groote Eylandt in broad locations and habitats

Searches for signs and spotlighting effort are presented in person-hours (h), opportunistic and standardised camera trapping in camera trap-nights (CTN) and pitfall trap effort in trap-nights (TN)

Habitat	Location	Sites (n)	Search (h)	Spotlight (h)	Opportunistic (CTN)	Standard (CTN)	Pitfall (TN)
Coastal grassland	North	12	45	16	32	0	0
Coastal grassland	South	5	33	20	96	120	1770
Coastal grassland	West	2	8	6	157	168	570
Sandy woodland	Interior	13	138	90	585	1088	5577

Table 2. Morphometrics of adult female and male N. aquilo captured during the study

Measurement	Female				Male			t	d.f.
	n	Mean	s.d.	n	Mean	s.d.			
Weight (g)	14	40.19	3.97	14	31.72	4.61	< 0.000	4.93	22
Head and body (mm)	14	94.29	5.34	14	89.5	5.93	0.034	2.24	25
Hind foot (mm)	14	35.39	1.76	14	34.82	1.75	0.397	0.86	25
Tail (mm)	14	165.42	9.72	14	156.2	17.3	0.104	1.70	20
Ear (mm)	13	16.0	0.18	13	15.27	0.43	0.140	1.55	16

hopping-mice. In total, 50 captures of 39 individual *N. aquilo* were made from 3830 trap-nights at the CP and Top Crossing (TC) sites (Fig. 1), giving an overall trap success of 1.02 individuals per 100 trap-nights. The CP trapping grid was trapped in seven sessions in 2013 and 2014, while TC was trapped four times during 2014. Pitfall traps captured *N. aquilo* in all trap sessions at CP except the seventh; those at TC were successful only once. Wide PVC traps yielded 32 captures, narrow PVC traps 15 captures, and bucket traps captured three small juveniles on one occasion. No *N. aquilo* were captured on any other trapping grids. *P. delicatulus* and a variety of frogs and reptiles were commonly captured in all three pitfall trap types. Pitfall trapping capture data for all species will be analysed and presented separately.

Morphology

Adult female *N. aquilo* were significantly heavier and longer than adult males, but there were no differences in hind foot, tail and ear lengths between females and males (Table 2). Minimum head and body length of sexually mature females was 89 mm, although no females were captured that measured between 78 and 89 mm (Fig. 3*a*). Minimum head and body length of sexually mature males was 81 mm (Fig. 3*b*).

The proportion of adults breeding did not differ significantly between the wet and dry seasons for females (Z=-0.39, P=1.00) or males (Z=-1.77, P=0.282). However, there was a significant seasonal effect on the proportion of juveniles in the population, with more juveniles observed in the dry season (Z=3.16, P=0.015) (Fig. 4). Most juveniles were captured in June and July, with fewer in September and December. Pregnant females were captured in April, May and August.

Body condition index was significantly affected by the combined age and breeding status of individuals ($F_{2,32}$ =66.27, P<0.001), with breeding adults having the highest body condition scores followed by non-breeding adults and then juveniles (Fig. 5). Females had significantly higher body

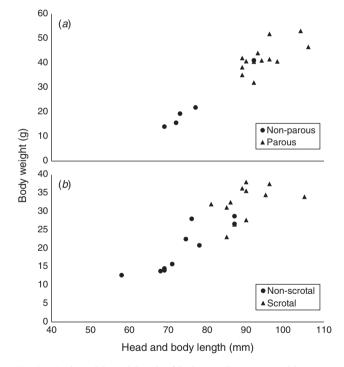


Fig. 3. Body weight and length of both sexually mature and immature (a) female and (b) male N. aquilo.

condition scores than males ($F_{1,32} = 20.48$, P < 0.001) (Fig. 6), while the effect of season bordered on significance ($F_{1,32} = 4.12$, P = 0.051), with slightly heavier individuals in the wet season.

Standardised camera trapping

Standardised camera trapping was conducted during 1376 trapnights in seven locations. As for pitfall trapping, *N. aquilo* was captured at standardised camera trap stations only in the CP and TC sites. Detection and the number of events varied between stations; however, at the two sites, the species was captured at a mean rate of 5.06 events per 100 trap-nights. Standardised camera trapping failed to detect the species in a trapping grid

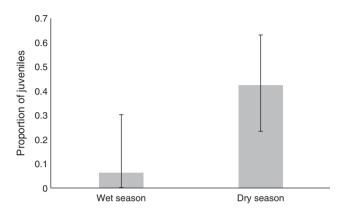


Fig. 4. Proportion of juveniles in the population as determined by pitfall trapping in the wet and dry season, with error bars indicating the 95% confidence interval.

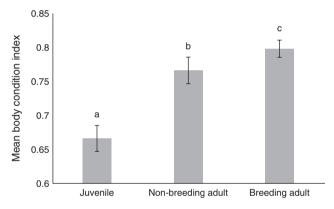


Fig. 5. Mean body condition index of combined age and breeding classes of *N. aquilo*. 'Breeding' refers to adults in breeding condition. Error bars indicate the 95% confidence interval and lettering indicates classes that are significantly different using Tukey pairwise comparisons.

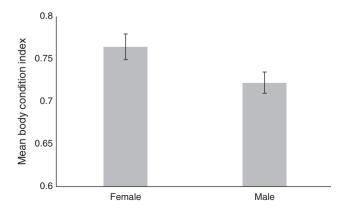


Fig. 6. Mean body condition index of male and female *N. aquilo*, with error bars indicating the 95% confidence interval.

north of these two locations in 2014; however, in 2015, opportunistic camera trapping near burrows detected it (Fig. 1). This area had been affected by fire during the 2014 trapping.

Mean number of events per 100 camera trap-nights did not differ significantly between CP and TC (t_5 =0.22, P=0.836). Number of events per 100 trap-nights correlated significantly with MNKA at CP (r=0.975, P=0.025) but not at TC (r=-0.229, P=0.771) (Fig. 7). The correlation at CP reached the critical value of 0.900 (d.f.=2, one-tailed test) after 10 consecutive days of camera trapping (Fig. 8).

Radio-tracking

The 95% active home-range size varied greatly between individuals, from 0.39 ha for a lactating female to 23.95 ha for a scrotal male (Table 3). The mean 95% core for all adults was 4.63 ha. Mean 95% core for lactating females was 0.45 ha and 1.61 ha for non-lactating females. Mean home-range size did not differ between the sexes for the 95% core (h_1 =2.16, P=0.142) or the 50% core (h_1 =2.16, P=0.142). On average,

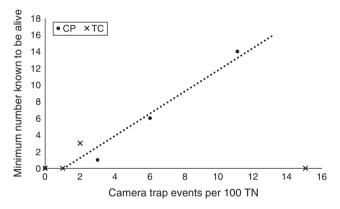


Fig. 7. The relationship between the number of *N. aquilo* camera-trap events per 100 trap-nights (TN) and the minimum number known to be alive at the Cave Paintings (CP) and Top Crossing (TC) trap sites. The dashed line shows the trend line for CP for which the correlation was significant.

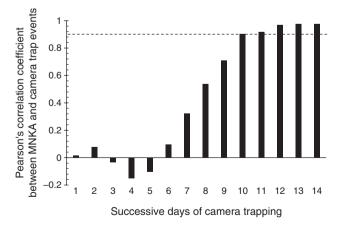


Fig. 8. The relationship between the number of successive days of camera trapping and the minimum number of *N. aquilo* known to be alive (MNKA) at the Cave Paintings trap site. The critical value (dashed line) of 0.900 was reached at Day 10.

Tracking period	ID	Sex	Breeding status	No. of days tracked	Total no. of fixes	95 HR (ha)	50 HR (ha)	No. of burrows	% night time in burrows	Open habitat used: open available
May	4	F	Lactating	14	43	0.39	0.12	1	28	1.05
May	5	F	Parous	17	52	0.6	0.18	4	23	1.07
May	6	M	Scrotal	14	34	10.03	2.44	3	41	1.38
August	1	F	Lactating	16	49	0.51	0.2	1	43	1.10
August	8	M	Scrotal	19	63	23.95	4.78	3	28	1.19
August	10	F	Pregnant	18	56	2.13	0.61	1	38	1.61
December	15	M	Scrotal	9	33	1.15	0.38	4	24	1.38
December	17	M	Scrotal	10	41	0.76	0.26	2	31	1.21
December	18	F	Parous	9	31	2.1	0.53	3	26	1.24

Table 3. Reproductive attributes, radio-tracking effort and results for the nine *N. aquilo* tracked during the study 95 HR, estimate of the 95% core of the active home-range; 50 HR, estimate of the 50% core of the active home-range

individuals spent 31% (\sim 4 h) of the nocturnal period in burrows and this did not differ significantly between the sexes ($h_1 = 0.06$, P = 0.806).

All individuals used open microhabitats proportionally more than what was available (Table 3) (n=9, P=0.004), with no difference between the sexes $(h_1=0.96, P=0.327)$. One female was frequently observed using a vehicle track to travel from her burrow to the area where she most often foraged. Individuals were rarely observed using woody debris or rocks as shelter, but sometimes crouched within grass tussocks or under shrubs. However, most often they were travelling or foraging in open areas.

The two lactating females and one pregnant female used only one burrow each throughout their respective tracking periods, while males and parous females all used 2–4 burrows each (Table 3). At least some overlap occurred between home ranges of males and females in each tracking session and the home ranges of two females also overlapped in one session (Fig. 9). A male in Session 2 was found using the same burrow as a male in Session 3. In the third tracking session, a male and female were found to be sharing two different burrows (Fig. 9). This female had previously mated but was not pregnant or lactating.

The radio-tracked animals frequently moved out of the trapping area, which was entirely within eucalypt woodland on sand. However, they rarely moved into the two other habitat types adjacent to the trapping grid: sandstone woodland and acacia shrubland (Fig. 2).

Burrow measurements

2.8

All animals radio-tracked during the study used burrows with visible spoil heaps; however, one of four burrows used by one male had a spoil heap obscured by bandicoot diggings. Radiotracking and camera trapping on burrows allowed a total of 24 confirmed *N. aquilo* burrow spoil heaps to be measured. Mean spoil heap length was 81.9 cm but ranged from 51 to 110 cm (Table 4). Mean width was 61.4 cm and mean height was 6.38 cm.

The number of pop holes found ranged from 0 to 4. Pop holes were far less conspicuous than spoil heaps, but radio-tracking aided their discovery as the collared animal was often located beneath the ground close to a pop hole. Pop holes that *N. aquilo* were observed using at night were never plugged during the day.

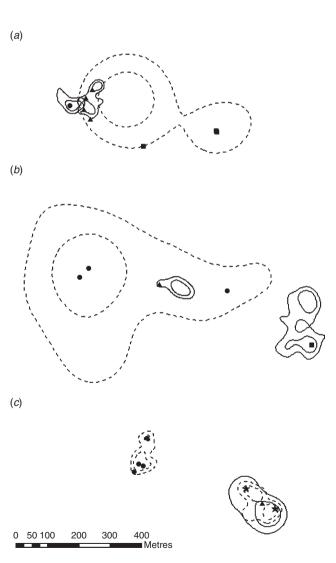


Fig. 9. The 95% and 50% estimated home range cores for female (solid lines) and male (dashed lines) *N. aquilo* radio-tracked in (*a*) May, (*b*) August and (*c*) December 2013. The burrows used by individuals in each session are differentiated by circles, triangles or squares with the stars in (*c*) indicating shared burrows by a male and female.

Burrow characteristic Mean Range s.d. Spoil length 24 81.9 cm 51-110 cm 13.01 Spoil width 24 61.4 cm 40-91 cm 12.72 Spoil height 24 6.38 cm $0-17 \, \text{cm}$ 4.16 No. of pop holes 18 1.5 0-40.96 Pop hole diameter 28 2.91 cm 2.1-4.0 cm 0.40 Distance from pop hole to centre of spoil 280.37 cm 120.39 2.7 91-535 cm

Table 4. Measurements of burrow characteristics of confirmed N. aquilo burrows

Visible tracks of the species were virtually non-existent in the woodland habitat due to the leaf litter cover.

Discussion

Intensive survey effort over three years in ostensibly suitable habitats for N. aquilo revealed only three locations where the species was detected. From this, it is evident that both the abundance and distribution of N. aguilo on Groote Eylandt have declined significantly since observations of the species last century. Thomson noted that hopping-mouse tracks were observable 'in incredible numbers' in the south-west corner of the island (in Dixon and Huxley 1985) while Johnson (1964) found himself unwittingly amongst a 'flourishing colony' of the species in the north-east where their burrows and tracks were numerous. Woinarski et al. (1999) later described the species as locally common with preferred habitats including shrublands and coastal grasslands. Despite prolonged and concerted effort, no such observations were achieved during our study. In areas where signs were located but subsequent camera trapping could not detect the species, it is likely that these signs (spoil heaps) were those of P. delicatulus and that N. aquilo was either absent or present but in low, undetectable numbers. Unfortunately, search efforts for the species across the island could not be standardised from the onset of the study because no effective methodology existed previously. Nonetheless, the decline of N. aquilo at the study site, particularly in coastal areas, is demonstrable from our results. Documenting a decline in eucalypt woodland is more problematic as there are no historical records of the abundance of the species in this habitat type.

Large areas of the interior of the island are inherently unavailable to N. aquilo due to the hard substrates of sandstone and laterite in which it would not be possible for the animals to dig burrows. In the remaining areas, the species' distribution is apparently patchy due to factors that could not be explained by this study. However, at the two study sites where we captured N. aquilo, the habitat could be characterised by deep sandy soils with a floristically diverse shrub and ground layer, with ground cover averaging 5-30% across the site. The area had been patchily burnt in the previous five years and during the study. Cool patch burning has been demonstrated to have a small positive or negligible effect on the abundance of N. alexis (Letnic 2003; Letnic and Dickman 2005), although numbers of this species fall dramatically in the wake of hot wildfires (Pastro et al. 2011). Some areas on Groote Eylandt, particularly those near roads and local indigenous communities, experience frequent hot fires one or more times each year; as with N. alexis, these fires could potentially reduce habitat suitability

for *N. aquilo* and drive its numbers to low levels. The only coastal area where the presence of *N. aquilo* could be confirmed was in the south-east of the island. This habitat was sand plain—coastal grassland with ground cover averaging 25%, consisting predominantly of the tussock grasses *Chrysopogon oliganthus*, *Sarga plumosum* and *Triodia microstachya*. This area was similar in both plant species composition and structure to several other areas that we searched unsuccessfully. It remains unknown why *N. aquilo* persists in the south-eastern site, apparently in relatively high numbers, but was absent from other areas that appeared to be little different.

Prior to this study, there were merely three reported captures of N. aquilo in live traps (Woinarski et al. 1999; Mahney et al. 2009). Our capture of 39 individuals has demonstrated that the species is more trappable than once suggested (Johnson 1964) and that, with suitable methods, animals can be obtained for ecological studies without collecting them from burrows that are ultimately destroyed in the process. However, it is clear that for occupancy and abundance studies, camera trapping is both more effective, less labour intensive and likely less expensive in the longer term. Overall capture success from camera trapping was five times higher than for pitfall trapping and detected the species during two additional sampling periods. The most plausible explanation for the correlation of number of events and MNKA at CP and not at TC is the larger number of pitfall traps at CP as the camera trapping effort was equal at both sites. The result is less likely due to differences in abundance and/or activity levels between the two sites as the mean number of events were not significantly different. Camera trapping also has lower ethical risks and, regrettably, three young N. aquilo were killed in a pitfall trap by a predator during the study (Diete et al. 2016). Due to these considerations, we recommend that the camera trapping methods outlined here be adopted for future monitoring of the species. Camera trapping for less than 10 consecutive days did not correlate with MNKA; therefore, 14 days or more is suggested, as abundance of the species may be low, potentially affecting detection probability.

The body condition of individuals improved with both age and breeding status, with adults in breeding condition being the heaviest relative to body length. Female *N. aquilo* were longer, heavier and had higher body condition scores than males. Sexual size dimorphism in favour of females has been noted in the closely related *N. alexis* (Breed 1983) but has otherwise not been reported in Australian murids. In most instances where sexual size dimorphism occurs in rodents, the bias is towards larger males (Schulte-Hostedde 2007). This disparity tends to occur in polygynous mating systems with intense male—male physical competition, where large body size increases male

mating success (Schulte-Hostedde 2007). Explanations for female-biased sexual size dimorphism are less consistent between species but, in reviews of its occurrence in mammals, has been attributed to increased competition between females (Ralls 1976) or reduced competition pressure between males (Isaac 2005). However, in many cases, multiple factors are likely to be responsible (Bondrup-Nielsen and Ims 1990; Isaac 2005). Breed (1983) suggested that *N. alexis* females may be the more aggressive sex on the basis of intra- and intersex interactions between captive animals, confirming earlier observations of captive animals by Stanley (1971). Thomson (in Dixon and Huxley 1985) noted very low levels of aggression in either sex in captive *N. aquilo* and found no fixed arrangement of sexes when excavating burrows of free-ranging animals.

30

In the present study, we recorded overlap of the home ranges of females. Females were also observed taking refuge after disturbance by researchers in burrows that they were not observed using during the day and were likely to have belonged to other females, who are the predominant builder of burrows (Diete et al. 2014). These observations suggest, therefore, that female territoriality and aggression may not be important drivers of sexual size dimorphism in this species. In a study of several genera of microtine rodents, Bondrup-Nielsen and Ims (1990) found very large home-range sizes of males to be the strongest predictor of female-biased sexual size dimorphism. They argued that in species for which mobility was more important than defence, selection would favour smaller males because total energetic costs increase with size. A similar mechanism is possibly influencing male size in N. aquilo. Mobility is clearly important for reproductively successful individuals given the tendency of animals to select for open microhabitats, leaving them potentially more exposed to predation risks. More agile and mobile males would likely be better at foraging, avoiding predators and be able to visit more burrows of females and therefore increase their opportunities for being selected for mating by the larger sex. It does not appear that male N. aquilo invest in their offspring in the form of parental care which is an intuitive, though frequently non-substantiated, consequence of female-biased sexual size dimorphism (Ralls 1976). Radio-collared lactating female N. aquilo were never observed foraging with other individuals, as was sometimes observed with non-lactating females and males. A camera trap placed next to the burrow of a lactating female captured only the collared individual and no other. This supports early observations that females with young are generally alone in burrows (Dixon and Huxley 1985), indicating that mothers raise their young unaided.

Thomson (in Dixon and Huxley 1985) captured wild juvenile *N. aquilo* in January, March, May, June and September. Our live trapping results also indicate that breeding is not strictly seasonal in this species, with breeding adults and juveniles captured throughout the year. However, we were able to demonstrate a higher influx of juveniles into the population in the dry season with more juveniles captured in June and July. This possibly coincides with improved conditions for dispersing young, such as the seeding of certain grass species. One of the most abundant grass species at the main trapping site, *Eriachne stipacea*, seeds predominantly between March and August (Mallett 2005). However, dietary studies are required to support

an association between juvenile abundance, food availability and food use. On two occasions, three small juveniles were captured together in pitfall traps, supporting observations that the litter size is usually three (Dixon and Huxley 1985).

The two largest active home ranges recorded during the study belonged to males in the dry season. Two males radiotracked during the wet season in December had much smaller home ranges even though all animals were in breeding condition. Unfortunately, too few individuals were radio-tracked to adequately demonstrate seasonal and sex-related differences in home range. However, the smaller male home ranges at the end of the year did coincide with reduced breeding activity, as evidenced by reduced proportion of juveniles in the population. All radio-tracked N. aquilo utilised discrete home ranges in the short term, although some individuals were clearly more mobile than others. Although several radio-tracking studies of other hopping-mice have been conducted, most calculated mobility rather than home range (Letnic 2001; Moseby et al. 2006; Dickman et al. 2010); therefore, we cannot directly compare our home range results with those of arid-dwelling Notomvs. N. alexis can travel distances of over 10 km over time (Dickman et al. 2010). Periods of low food availability have been associated with increased mobility and decreased burrow fidelity in this species (Dickman et al. 1995, 2010). Food availability is less variable in tropical climates; therefore, the variation in home range of N. aquilo in our study is more likely to be related to seasonal breeding activity rather than food availability.

Bipedal rodents are common components of arid-zone faunal assemblages and their preferential selection of open microhabitats is well documented (Kotler and Brown 1988). This preference is likely due to their increased agility and capacity to avoid predators compared with their quadrupedal counterparts which select more sheltered microhabitats (Kotler and Brown 1988). Our results demonstrate that the preferred selection of open microhabitats is also true for a bipedal rodent utilising a tropical environment. This may decrease competition between N. aquilo and sympatric quadrupedal rodents such as P. delicatulus. A similar spatial partitioning of the habitat has been demonstrated for N. alexis and the sandy inland mouse (Pseudomys hermannsburgensis) in arid central Australia (Murray and Dickman 1994), with bipedal N. alexis selecting more open habitats than the quadrupedal P. hermannsburgensis and using escape rather than avoidance behaviour to reduce predation risk (Spencer et al. 2014). Conversely, Morris et al. (2015) found the dusky hopping-mouse (Notomys fuscus) to forage preferentially under cover. It was not clear whether other rodents occurred in this last study, but it is possible that hopping-mice may extend their activity to incorporate sheltered habitats if other rodent species are absent.

Almost all burrows observed during the study were associated with clearly visible spoil heaps and most had pop holes that could be found with a small amount of search effort. However, the mean number of pop holes of 1.5 per burrow is very likely an underestimate due to the inconspicuousness of some of these burrow entrances in woodland habitat. Contrary to observations by Johnson (1964), pop holes that radio-collared individuals were observed using at night were never found to be plugged during the day. The results suggest that these indirect

signs may still be useful in indicating the presence of N. aquilo regardless of the potential confusion with P. delicatulus signs (Diete et al. 2015a). However, confirmation should always be attained with an unequivocal method such as camera trapping. There was clear overlap in this study in the sizes of spoil heaps associated with N. aquilo burrows and the small number that could be confirmed as belonging to P. delicatulus (Diete et al. 2015a). Anecdotally, the spoil heaps of P. delicatulus appear to be larger in coastal areas with softer sand where this species can be very abundant (R. L. Diete, unpubl. data). There is potential for a greater degree of overlap in the sizes of spoil heaps of these species in these coastal habitats. As the minimum N. aquilo spoil heap length was 51 cm and the mean was 82 cm, spoil heaps measuring under 50 cm at the longest length can be disregarded as belonging to N. aquilo with reasonable confidence.

In this study we were able to make significant advances in the understanding of the ecology and conservation of *N. aquilo*, despite being initially constrained by confounding information on effective sampling techniques, low capture rates and difficulty of access at the study site. Future research and management should endeavour to elucidate drivers of decline and extirpation of this charismatic species to better inform management decisions and conservation. Because of the large reduction in the distribution and abundance of this species on Groote Eylandt, and the absence of any recent records of this species elsewhere, we recommend that the conservation status of this species be upgraded to 'endangered' under the *EPBC Act 1999*, which would reflect that of the IUCN Red List.

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