

Environmental factors influence nest excavation by foxes

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Abstract

Predation rates of freshwater turtle nests can vary markedly, suggesting that in addition to different suites of predators present, environmental factors (e.g. vegetation characteristics, distance to water/clearing and rainfall) also influence survival of turtle nests. Understanding the influence of environmental factors on nest success can aid turtle conservation through successful management of nesting habitat. This study simultaneously investigated the effect of multiple factors on artificial nest survivorship at a site where oblong turtles *Chelodina colliei* were present. Over a 12-month period, we monitored the fate of 580 artificial nests installed as 145 nest-sets, each having one of four treatments: (1) two chicken eggs (E) buried and the ground sprayed with water from a turtle pond (S) (E+S+), (2) two chicken eggs (E+S-), (3) nest sprayed (E-S+) and (4) hole dug and refilled (E-S-). Seven environmental factors were recorded for each site and nests. Nests were monitored for 2 months after installation, by which time, 46% of the nests had been excavated. Depredation rate was affected by both the presence of eggs ($P < 0.001$) and being sprayed with turtle pond water ($P = 0.005$); but we found that even 38% of empty nests (holes simply dug and refilled) were excavated. Nest excavation was more likely for more obvious nests located in areas with more sparse vegetation ($P < 0.05$) closer to the shoreline ($P < 0.01$). Excavation rates were highest immediately after installation, but continued for the duration of the monitoring period. The introduced red fox *Vulpes vulpes* was identified as the only predator observed on cameras for a subset of 60 nest-sets. In conclusion, foxes use both visual and olfactory cues to locate nests, and environmental conditions at the nest site significantly influence the fate of the nests.

Introduction

Chelonians are subject to high mortality during the early life stages (Congdon *et al.*, 1987; Shine & Iverson, 1995). The leading cause of nest mortality in many turtle species is nest predation (Tinkle, Congdon & Rosen, 1981; Congdon *et al.*, 1983; Thompson, 1983). For example, Thompson (1983) reported that 96% of turtle nests in the Murray River were depredated; 93% of which was by red foxes *Vulpes vulpes*.

While numerous studies record the identity of nest predators, fewer studies have examined factors that potentially influence the rate of nest depredation. A review of the literature reveals common trends across previous studies: nest depredation can be influenced by vegetation cover, distance to the shore, as well as age of the nest (Table 1). However, most previous studies have measured only a few environmental factors simultaneously, or have monitored nests over a relatively short duration (compared with the potential length of time of turtle egg incubation) (Table 1).

While artificial nests do not indicate depredation rate of real nests (Wilson, Brittingham & Goodrich, 1998), they allow manipulation of olfactory and visual characteristics to inves-

tigate cues used by predators in locating nests (Marchand *et al.*, 2002; Spencer, 2002; Burke, Schneider & Dolinger, 2005). In this study, we simultaneously investigated the effect of seven environmental measures (e.g. vegetation characteristics, distance to water/clearing) on the fate of four artificial nest treatments over 1 year. This is the largest, longest-term study of depredation of artificial nests, as well as examining multiple factors within a single experimental design. We also identified the predator species responsible for excavation of these artificial nests using cameras positioned over a subset of 60 nest-sets.

Methods

Study site and study species

This project investigated excavation of artificial nests at Chittering Lakes Nature Reserve (241.2 ha), in the area surrounding Lakes Chittering and Needonga, Bindoon, Western Australia. Study sites were arranged over ~10 km of the shoreline around these lakes at sites, and were accessed by private

Table 1 A summary of studies that have examined the factors affecting predation of freshwater turtle nests

Reference	Species	Location	Predator	Vegetation	Distance to water	Human disturbance	Rainfall	Age of nest	Nest density	Edge effect
Present study	Artificial nests in <i>Chelodina colliei</i> sites	Australia	Red fox (introduced)	✓	✓		✗	✓		
Wilhoft <i>et al.</i> (1979)	Artificial nests	America	Raccoon				✓		✗	
Burke <i>et al.</i> (2005)	Artificial nests	America	Raccoon					✓		
Marchand & Litvaitis (2004)	Artificial nests	America	Raccoon		✓	✓ ^a			✓	✗
Marchand <i>et al.</i> (2002)	Artificial nests	America	Raccoon, fox		✓			✓	✓	
Hamilton <i>et al.</i> (2002)	Artificial nests	America	Raccoon, opossum	✓		✓ ^b				✓
Spencer (2002)	<i>Emydura macquarii</i>	Australia	Red fox (introduced)		✓					
Doody <i>et al.</i> (2006)	<i>Carettochelys insculpta</i>	Australia	Yellow spotted monitor			✓ ^c				
Baldwin <i>et al.</i> (2004)	<i>Chrysemys picta</i>	America	Raccoon		✓					✓
Burger (1977)	<i>Malaclemys terrapin</i>	America	Fox, raccoon, gull, crow	✓				✓		
Burke <i>et al.</i> (1998)	<i>Kinosternon subrubrum</i> , <i>Pseudemys concinna floridana</i> , <i>Trachemys scripta</i>	America	Raccoons						✗	
Congdon <i>et al.</i> (1983)	<i>Emydoidea blandingii</i>	America	Raccoons, fox		✗			✓		
Congdon <i>et al.</i> (1987)	<i>Chelydra serpentina</i>	America	Raccoon		✓			✓		
Escalona & Fa (1998)	<i>Podocnemis unifilis</i>	Venezuela	–	✓						✓
Christens & Bider (1987)	<i>Chrysemys picta marginata</i>	Canada	Raccoon		✓			✓		

✓, found a significant effect; ✗, found no effect. ^aPredation was reduced in close proximity to roads. ^bThe installation of artificial deer feeders (known to subsidize predators) was found to increase nest predation. ^cThe introduction of the cane toad *Bufo marinus* caused a decrease in the population of yellow spotted monitor *Varanus panoptes*, the primary predator of *Carettochelys insculpta*.

property (permission granted by landowners) or public walk trail [nest sites were placed a sufficient distance (>20 m) from this trail to avoid interference from public and pets using the walking trail].

Vegetation surrounding the Chittering lakes is highly modified and fragmented because of proximity to both agricultural lands and a major highway. The vegetation buffer around the two lakes is a narrow (averaging ~40 m, broadening to >300 m in some places) strip of native vegetation dominated by *Melaleuca* and *Eucalyptus* tree species. Outside the reserve, the native vegetation has been cleared for paddocks, cropping or orchards, with little or no ground cover or canopy.

The lakes support a population of *Chelodina colliei*. These freshwater turtles inhabit rivers, lakes and swamps throughout the suburban, agricultural and natural areas across the region (Burbidge, 1967; Bush *et al.*, 1995). *Chelodina colliei* nests between September and January, and females may lay twice in the same year (Clay, 1981). Nesting is associated with specific weather conditions: seasonal rain-bearing low-pressure systems, falling barometric pressure and an ambient air temperature >17°C (Clay, 1981). The incubation period ranges from 210 to 222 days (Clay, 1981), with hatchlings emerging in mid-August (Clay, 1981; McCutcheon, 1985).

While the period over which artificial nests were installed did not explicitly correspond with the natural nesting period (although they overlapped), turtle eggs/embryos are potentially present at these sites year-round (Table 2) and predators may therefore benefit from digging for nests year-round.

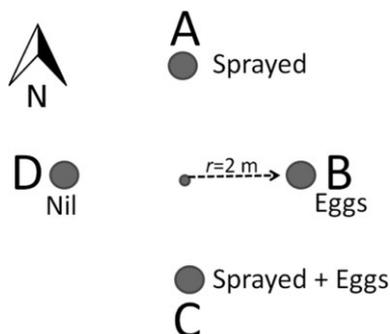
Female *C. colliei* excavate a nest around 150 mm deep using their hind limbs, with a chamber at the bottom for the eggs (Clay, 1981). After oviposition, the soil is replaced and the nest is compacted by the female raising her body and letting it fall on the nest (Clay, 1981). The female's nest distance to water varies from 20 (Clay, 1981) to 500 m (Burbidge, 1967), although sightings of turtles by landowners around the Chittering lakes (Densley, C., pers. comm.) suggest that this distance may be up to 1000 m. Clay (1981) reported that females generally chose areas free from thick vegetation for nest sites.

Artificial nests

Artificial nests were placed out between March 2012 and February 2013 (Table 2). Suitable sites ($n = 145$; relatively level areas clear of vegetation other than grass, with a diameter of ~5 m, to ensure consistency of conditions among nest-sets) were identified and marked with a metal peg and flagging tape.

Table 2 Timeline of *Chelodina colliei* nesting, turtle eggs present in the ground and the numbers of artificial nest-sets installed

	J	F	M	A	M	J	J	A	S	O	N	D
<i>Chelodina colliei</i> nesting	Y								Y	Y	Y	Y
Turtle eggs/embryos present	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y
Number of artificial nest-sets installed	6		18	27	17		30	21				26

**Figure 1** The layout of a nest set, indicating orientation and treatments: combinations of chicken eggs present (E+) or absent (E-); sprayed with turtle pond water (S+) or not (S-). r = radius.

Each site had four artificial nests ($n = 580$ individual nests in total) installed 2 m to the north, east, south and west of the central peg (Fig. 1). Each hole was approximately 150 mm deep; we based this depth on our observations of excavated (depredated) *C. colliei* nests at this site. Each hole was assigned one of four treatments, with each nest-set including all combinations of the presence and absence of chicken eggs and turtle pond water (Fig. 1). Two supermarket-sourced chicken eggs were placed at the bottom of two of the nests. Water collected from a turtle pond (containing female *C. colliei*; Armadale Reptile Centre, Perth, Australia) was sprayed into two nests: five pumps (or ~3 mL from a hand-held spray pump bottle) into the bottom of the open hole and on the surface of the filled hole. A similar method has been used for other nest depredation studies (Hamilton, Freedman & Franz, 2002; Marchand *et al.*, 2002). After closing, the surface of the artificial nest was lightly brushed by (gloved) hand to replace any disturbed grass/compost. To minimize contamination with human scent, only the trowel (used for digging), the researcher's shoes, knees and gloved hands touched the ground.

For each nest-set, seven environmental factors were recorded (Table 3). Data were transformed to achieve sphericity of data distribution (Shapiro–Wilk's W -test; Table 3). Weather data were sourced from the Bureau of Meteorology (www.bom.gov.au, accessed on 22 March 2013) using weather stations located at Bindoon (local) for rainfall and Gingin (19 km west northwest of study site) for daily minimum and maximum temperatures.

Artificial nest sites were checked regularly (every 2–3 days) up to 3 weeks after installation, after which checking was performed weekly. Checking the nest-sets was carried out on

Table 3 The environmental variables recorded for each nest in the study

Measure	Definition
Distance to water (m) (log-transformed)	For the majority of nests, measurement (± 1 m) was paced out. For 10% of locations (distance was too great, or vegetation too dense), this was measured using ArcMap from GPS coordinates to the edge of the vegetation around the lake (unless the water was visible) (Landsat imagery ^a). Only permanent water was included for distance to water measurements.
Distance to nearest clearing (log-transformed)	For the majority of nests, measurement (± 1 m) was paced out. For 10% of locations (distance was too great, or vegetation too dense), this was measured using ArcMap from GPS coordinates to the edge of the vegetation around the clearing.
Distance to nearest trunk (m) (square root-transformed)	Measurement (± 1 m) was paced out. Only included woody trunks >100 mm in diameter.
Canopy cover (%) (arsine-square root-transformed)	Measured using a convex spherical densitometer (Lemmon, 1956). Four readings were taken, facing north, east, south and west of the central marker peg; these were averaged.
Number of trunks within 10-m radius	Count of the number of woody trunks (>100 mm diameter) within 10-m radius of nest set.
Grass cover	Qualitatively ranked on a scale of 0 to 3, where 0 = bare ground, 3 = dense cover >100 mm in height.
Crypsis	Qualitatively ranked on a scale of 1 to 3, where 1 = obvious disturbance (discoloration was evident for areas, even for sites with no grass or compost to cover the digging, or the substrate was so hard that the filled hole was not continuous with the surrounding substrate), 3 = no sign of disturbance.

^aChittering_2135_Nov_2010_Mosaic.Ecw, 1:80000.

foot, visually inspecting the area for disturbance and taking efforts to minimize any further disturbance to the area. The fates of nests were recorded at each inspection (excavated or intact) and the minimum time to excavation recorded.

Identification of nest predators

A subset of 60 nest-sets was also monitored by motion sensor camera. Appropriate sites (i.e. secure from human disturbance, sufficient clearing for the camera field of view) had one of 27 motion sensing cameras (Reconyx Hyperfire™, Reconyx, Holmen, WI, USA) placed to include the full nest-set within the field of view (approximately 5–10 m from metal peg and 1–2 m above ground level). Cameras were set to rapid-fire (five photos after every stimuli detected). Cameras were checked regularly (weekly or fortnightly). Cameras were set at the same time as nest installation and were left in position until all nests in the nest-set were excavated. Over 100 000 images were analysed over the course of this study to identify the nest predators (excavating nests), to improve accuracy of measures of time until excavation and to indicate the order in which the nests were excavated. We also carried out an activity index of all potential predators, scoring the number of passes in front of the cameras (including when animals triggered the camera but did not stop or show any interest in nest site). Separate passes were identified as passes by the same species separated by >1 h to take into account multiple trigger events.

Depredated turtle nests

During installation and monitoring of the artificial nest-sets, we searched the surrounding vegetation for evidence of depredated turtle nests (intact nests are not evident without disturbance). Depredated turtle nests were recorded and marked with a Global Positioning System. Only diggings containing remains of turtle eggs were included to avoid inclusion of unsuccessful digs. All excavations were similar to previous descriptions for *C. colliei* (e.g. Giles, 2001). Environmental variables for depredated turtle nests were recorded in the same fashion as for artificial nests (with the exception of crypsis at installation).

Data analyses

Nest environmental factors were analysed using a mixed-model analysis of variance (ANOVA) with the fate of the nest (excavated or intact at 2 months from installation) as the dependent variable. Nest-set ID was a random (blocking) factor, nest treatments (egg presence or absence; turtle pond water presence or absence) were fixed factors and the seven environmental values (Table 3) were covariates.

Effect of treatment on nest age at the time of predation (days) was analysed by Kruskal–Wallis ANOVA. Friedman ANOVA was used to compare the relative order of predation for 22 nest-sets where all the nests were excavated but we had some data on the order of predation (either through camera observation or where the nests were excavated over multiple nest checking intervals).

Spearman's rank order correlation (r_s) was used to examine the relationship between the number of nests excavated over each night (dependent variable) and rainfall (previous 24 h to 09:00 am and sum of the previous 7 days) and temperature (minimum and maximum for the previous 24 h) as independent factors.

Environmental data for depredated turtle nests were compared with those for excavated and intact artificial nest data using multidimensional scaling with a Bray–Curtis distance measure (PAST version 2.16, Hammer, Harper & Ryan, 2001) for differences between the three nest categories (excavated artificial nests, intact artificial nests, depredated turtle nests) using one-way analysis of similarity (ANOSIM). We tested all environmental factors (Table 3) except for crypsis at installation in our analysis (as this could not be measured for the turtle nests) and ran similarity percentage (SIMPER) analysis to determine factors that contributed to differences between the groups of nests.

Data are presented as means \pm 1 SD. Statistical significance was accepted at $\alpha < 0.05$. Analyses were carried out using Statistica 8.0 (StatSoft Inc., 2007) except where indicated.

Results

Fate of artificial nests

Forty-six per cent of the 580 artificial nests were excavated within 2 months of installation; the majority (89%) of which were within 30 days of installation (Fig. 2a). Factors influencing the fate of nests (excavated or intact after 2 months) are summarized in Table 4. Nest-set ID was a significant factor in determining the fate of nests ($F_{137,433} = 9.91$, $P < 0.001$), suggesting that there was a high degree of spatial autocorrelation.

The presence of chicken eggs ($F_{1,433} = 30.02$, $P < 0.001$) and turtle pond water ($F_{1,433} = 8.03$, $P = 0.005$) had a significant effect on the fate of the artificial nests. From highest to lowest excavation rates: E+S+ (53%), E+S– (48%), E–S+ (43%) and E–S– (38%).

Although there was no significant effect of nest treatment on the overall time until excavation (Kruskal–Wallis ANOVA $H_{3,n=355} = 3.27$, $P < 0.351$) (Fig. 2b), this may reflect the observation that foxes would often excavate all nests within a nest-set within a short period of time. Sixty-one per cent of individual nests excavated were part of a 'whole nest-set predation', where all four treatments were excavated, and 69% of these events had all four treatments excavated within 24 h of each other. These data therefore did not contribute to a difference in survival time between treatments. For 22 whole nest-set excavations, where we had sufficient information to allow us to determine the order of excavation, the order was significantly different between treatments (Friedman ANOVA $\chi^2_{d.f.3} = 20.91$ $P < 0.001$); from earliest excavated to last:

E+S-	E+S+	E-S+	E-S-
	=====	=====	

where underlining links treatments that were not significantly different in terms of order of excavation (pairwise Friedman analyses).

A significant effect for distance to water (Fig. 3a; $F_{1,137} = 9.70$, $P = 0.002$) indicated more predation closer to the shoreline. Three measures of vegetation reflect a significant negative effect of vegetation density on excavation rate: dis-

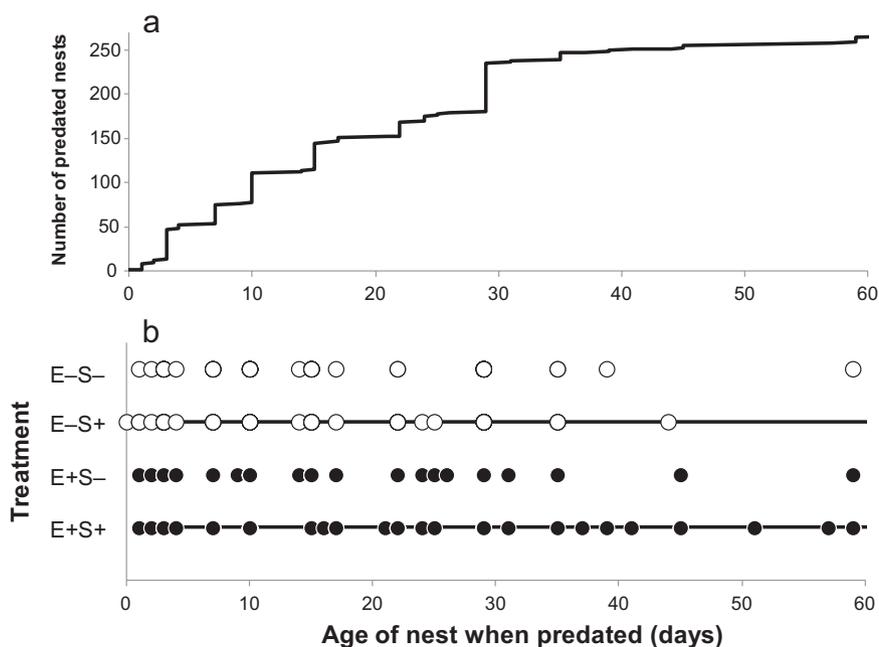


Figure 2 The effects of time on predation of artificial nests. (a) shows cumulative nest predation over time, (b) shows age of nest at the time of predation for each of the four treatments (filled symbols indicate the presence of eggs, horizontal lines indicate the presence of turtle pond water).

Table 4 Summary of the mixed-model ANOVA testing factors affecting the fate of artificial nests (excavated or intact at the end of the study)

	Effect	d.f.	<i>F</i>	<i>P</i>	
Nest-set ID	Random	137,433	9.91	<0.001	***
Eggs (presence/absence)	Fixed categorical	1,433	30.02	<0.001	***
Pond water (sprayed/not sprayed)	Fixed categorical	1,433	8.03	0.005	**
Distance to water	Covariate	1,137	9.70	0.002	**
Distance to clearing	Covariate	1,137	1.06	0.304	
Distance to nearest trunk	Covariate	1,137	26.23	<0.001	***
Canopy cover	Covariate	1,137	14.01	<0.001	***
Number trunks in 10-m radius	Covariate	1,137	4.20	0.042	*
Grass cover	Covariate	1,137	2.71	0.102	
Crypsis	Covariate	1,137	58.17	<0.001	***

Statistical significance is indicated with asterisks (* $P < 0.05$, ** $P < 0.01$ and *** $P < 0.001$).

tance to nearest trunk (Fig. 3b; $F_{1,137} = 26.23$, $P < 0.001$), canopy cover (Fig. 3c; $F_{1,137} = 14.01$, $P < 0.001$) and number of trunks within a 10-m radius (Fig. 3d; $F_{1,137} = 4.20$, $P = 0.042$). Excavated nests were also scored as more obvious (i.e. less cryptic) during installation compared with those that were intact at the end of the study ($F_{1,137} = 58.17$, $P < 0.001$). The effect of grass cover ($F_{1,137} = 2.71$, $P = 0.102$) and distance to nearest clearing ($F_{1,137} = 1.06$, $P = 0.304$) were not statistically significant.

The number of artificial nests excavated in a day was not significantly correlated with rainfall in the preceding 24 h ($r_s = 0.024$, $P > 0.05$) or the previous 7 days ($r_s = -0.022$, $P > 0.05$), nor the daily minimum ($r_s = 0.036$, $P > 0.05$) or daily maximum ($r_s = 0.028$, $P > 0.05$) temperatures.

Comparison with depredated turtle nests

Thirty-four depredated turtle nests were located in this study. The habitat variables for these depredated turtle nests did not

differ significantly from excavated artificial nests (one-way ANOSIM: $R = -0.075$, $P = 0.949$), but both were significantly different from intact artificial nests (comparison with excavated artificial nests: $R = 0.097$, $P < 0.001$ and depredated turtle nests $R = 0.129$, $P = 0.008$) (Fig. 4). The number of trunks in a 10-m radius accounted for 82.5% of the difference between intact and excavated artificial nests and 81.2% of the difference from depredated turtle nests (SIMPER).

Artificial nest predator identification

Six species of potential nest predators were identified on camera traps (Table 5). Foxes were the most common nest predator seen in the area (94% of all sightings). Ninety-three per cent of cameras placed out during this study recorded at least one fox, indicating that foxes were present throughout the study area. Only foxes were witnessed excavating artificial nests (26 separate camera events) (Fig. 5a). At one site, a raven

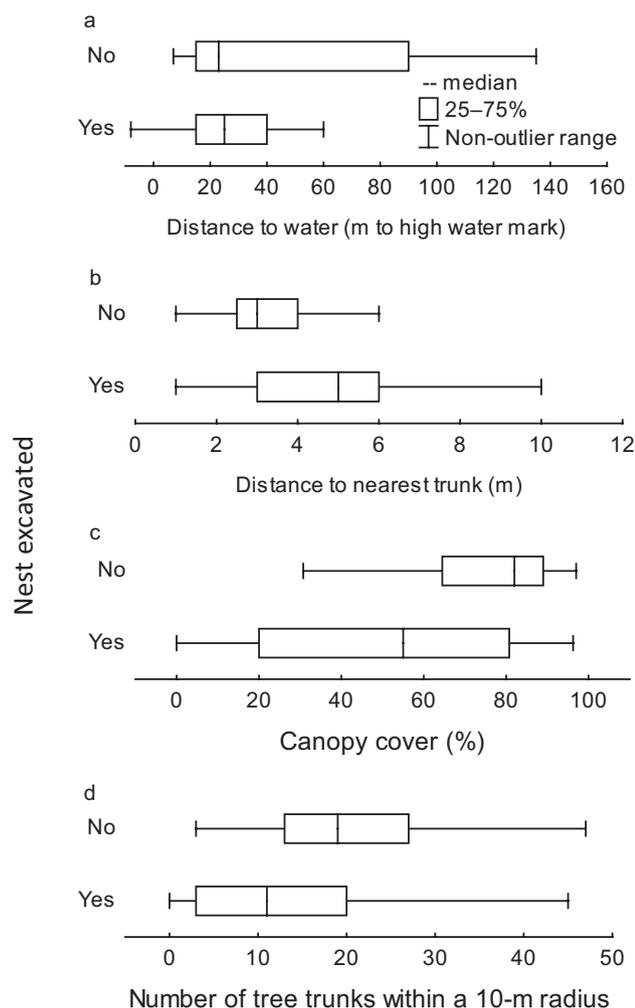


Figure 3 Environmental factors that had a significant influence on the fate of artificial nests (Table 4). Data shown are the non-transformed raw values and therefore non-parametric measures (median, quartiles) are shown.

Corvus coronoides was photographed investigating a newly installed artificial nest and then 2 and 4 days later (after the same nest had been excavated by a fox). Feral pigs *Sus scrofa*, feral cats *Felis catus* and rats *Rattus rattus* were photographed in the area; however, there was no evidence of excavation/investigation of the sites by these species. Domestic dogs *Canis familiaris* were once viewed on camera investigating an artificial nest but did not exhibit behaviour consistent with predation of a nest (i.e. was not digging). All diggings at sites where predation was confirmed were wide holes (>5 cm diameter), with a pile of earth moved to one side (Fig. 5b). All excavations of unconfirmed predator were of a similar nature to those confirmed.

Discussion

Nearly half of the artificial nests (46%) were excavated in the present study. Predation rates of artificial nests have been

recorded as low as 22% (Marchand *et al.*, 2002), but also as high as 80% (Spencer, 2002), 89% (Hamilton *et al.*, 2002) and 100% (Wilhoft, Del Baglivo & Del Baglivo, 1979). We identified the introduced red fox the major nest predator; no other species was identified excavating artificial nests. The presence of chicken eggs and turtle pond water increased the chance of excavation of artificial nests. We also identified various environmental factors (proximity to the shoreline, vegetation density) which were correlated with increased likelihood of a nest being excavated.

Environmental factors

Chelodina colliei selects nest sites that are relatively open (Clay, 1981), and we found that excavation was greater for artificial nests located in areas with less canopy cover and lower vegetation density. This result contrasts with studies of other turtle species that have recorded greater predation in more vegetated areas (Burger, 1977; Hamilton *et al.*, 2002). For example, Escalona & Fa (1998) report that terrapin nest predation by animals was greater closer to vegetation (while predation by humans was greater in more open areas), while Burger (1977) reported more avian predation (gull, crow) in open areas but more mammalian predation (fox, raccoon) in more vegetated areas. Our results suggest that foxes are more likely to find artificial nests placed in the open and differences between studies are likely to reflect the search patterns of likely predators.

Another common finding of past studies, which was supported by our data, is that predation is greater closer to the shore (Christens & Bider, 1987; Marchand *et al.*, 2002; Spencer, 2002; Baldwin, Marchand & Litvaitis, 2004; Marchand & Litvaitis, 2004; but see Congdon *et al.*, 1983). Nest predator species vary with distance to water (raccoons predated nests closer to water but foxes being the major predators for nests further from shore; Congdon *et al.*, 1987) and therefore the methods used by predators to search for nests are likely to be an important factor influencing nest predation rates. *Chelodina colliei* is known to make large journeys to nest (Clay, 1981; Guyot & Kuchling, 1998); however, studies report that the majority of nesting occurs only 25.4 ± 2.4 m from the shore in the December/January season, and 86.6 ± 10.0 m in the September/November season (Clay, 1981). Therefore, predators concentrating their searches in areas close to the water edge may do so to maximize their success.

Effect of nest age and season

In the present study, the majority of artificial nests were excavated within 30 days of installation. Previous studies similarly report that, for both real and artificial nests, predation is greatest close to the time of oviposition/installation (Burger, 1977; Wilhoft *et al.*, 1979; Tinkle *et al.*, 1981; Congdon *et al.*, 1983, 1987; Christens & Bider, 1987; Hamilton *et al.*, 2002; Spencer, 2002), with predation often occurring within hours of oviposition. The increased vulnerability for new nests is

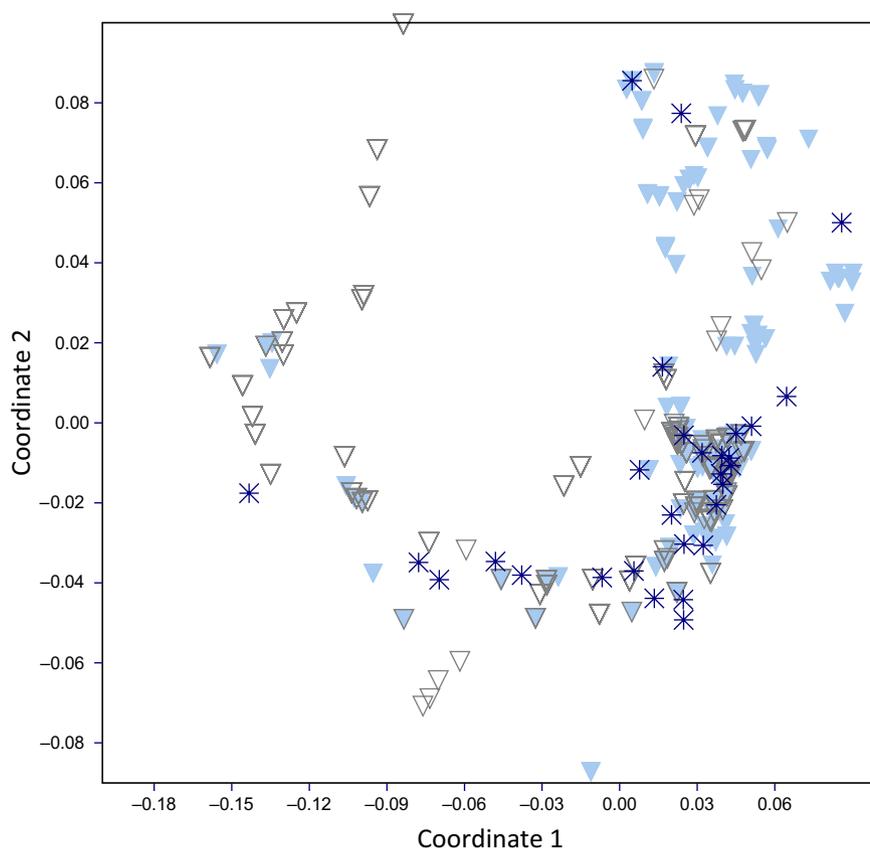


Figure 4 A multidimensional scaling plot indicating the environmental variables for 580 artificial nests (∇ 265 excavated; \blacktriangledown 315 nests that remained intact at the end of the study) and 34 depredated turtle nests (*). There is overlap in excavated and intact nest values where the four nest treatments may have had different outcomes (excavated or not).

Table 5 Values are the numbers of identified depredate events (artificial nest excavated) and the total activity indices of each potential predator species (number of separate passes in front of the cameras). Percentage values are for the total across all predator species

	Nest excavation			Total activity	
	Camera	Total	%	Camera	%
Fox <i>Vulpes vulpes</i>	26	26	96.3	466	93.4
Dog <i>Canis familiaris</i>	0	1 ^a	3.7	12 ^b	2.4
Raven <i>Corvus coronoides</i>	0	0	0	3	0.6
Feral pig <i>Sus scrofa</i>	0	0	0	3	0.6
Feral cat <i>Felis silvestris catus</i>	0	0	0	12	2.4
Rat <i>Rattus rattus</i>	0	0	0	3	0.6
Total	29	27		499	

^aIn the single event where a dog was photographed in the vicinity of an artificial nest; the behaviour of the dog did not conform with a nest predation event (i.e. it was not digging).

^bA resident dog (residence <50 m away from the artificial nest site) was captured on cameras, chasing foxes away from the nest sites.

thought to be a result of the cues (visual, olfactory) aiding in predator detection of nests fading with time (Congdon *et al.*, 1983).

Most previous studies have not monitored nests for long past installation. For example, Spencer (2002) considered a nest successful if it was not depredated within 28 days, while

Hamilton *et al.* (2002) monitored nests for only 21 days. Given that *C. colliei* incubate for 200–230 days (Burbidge, 1967), it is very possible that nests may be excavated for the full term of the nest and extending the length of nest monitoring may improve understanding of the effects of nest depredate.

Giles (2001) proposed that rainfall may increase predation rate of nests, a theory that was not supported by our data. Other studies have similarly found that rainfall did not decrease the chance of nest predation (Wilhoft *et al.*, 1979), while decreased predation of artificial bird nests after rain has been postulated to be due to reduced olfactory cues following rainfall (Whelan *et al.*, 1994). Turtle embryos move towards warmer regions of the egg to avoid dangerously high temperatures (Zhao *et al.*, 2013), and it is possible that rainfall may elicit similar movements within the egg that predators could be trained in to finding. Clearly, the mechanism that predators use to locate nests will be important in this regard.

Predator detection of nests

The red fox has applied significant and unsustainable predation pressure to many turtle populations, killing adults and depredating predating nests. Red foxes have been identified as

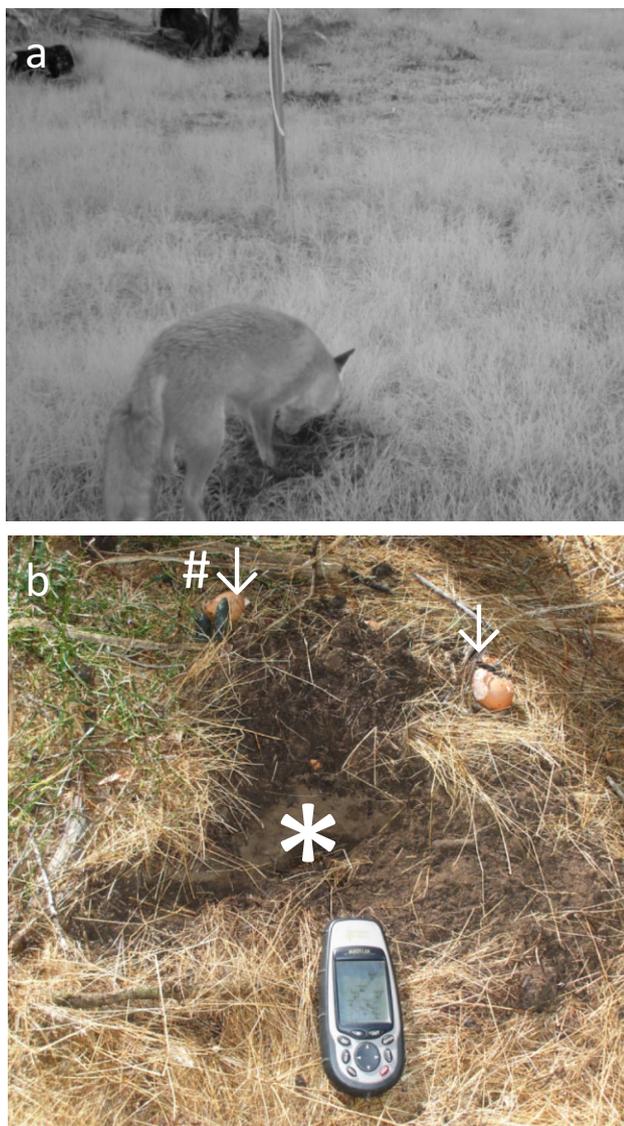


Figure 5 Only foxes were captured on camera excavating artificial nests (a). Evidence of predation at excavated nests (asterisk) included eggshell fragments (arrows) and the occasional fox scat (#).

the most prevalent predator of turtle nests across a number of studies and have been labelled a causative factor in Australian turtle population declines (Parmenter, 1976; Chessman, 1978; Thompson, 1983; Spencer & Thompson, 2003, 2006).

The presence of chicken eggs and being sprayed with turtle pond water increased the likelihood of nests being excavated suggesting that olfactory cues may have also aided predator location of nests. Similarly, soil disturbance and ocean water increased predation of artificial nests by raccoons (Burke *et al.*, 2005), although Hamilton *et al.* (2002) found no effect of olfactory cues (water from a turtle enclosure) on nest predation rate.

We found that artificial nests which were more visually obvious (less 'cryptic') were more likely to be excavated, but

even 38% of empty 'nests' (E-S-; i.e. simply dug and refilled holes) were excavated. Predators also excavate artificial nests containing ping pong balls (Wilhoft *et al.*, 1979). These findings suggest that, in addition to olfaction, foxes use visual cues (soil disturbance) to locate nests. Similarly, Spencer (2002) reported that more disturbed nests (synonymous with less cryptic in our study) were more likely to be depredated by foxes although Hamilton *et al.* (2002) found no effect of hiding visual cues (spreading sand over the nest site to mask specific visual cues) on nest depredation rate.

Mass predation events

We recorded 61% of individual nests were excavated as part of 'whole nest-set predations', in which all four treatments were excavated. On three occasions, localized (encompassing several hundred metres of lake shoreline or ~2–5 ha) mass excavation events occurred overnight. Of the 126 nest-sets where predation was recorded, 81 (64%) had two or more nests excavated within 24 h, and a nest's fate was confounded by the chance of the nearest nest being excavated (evident as a significant random factor – nest ID – effect in the statistical analyses). Mass predation events may be aided by the small distance between treatments in this study: the four nests in each nest-set were placed 2 m from a central point (~2.8 m between nests). Because 'clumped' nests are more likely to be depredated than scattered nests (Burger, 1977; Marchand *et al.*, 2002), previous studies have installed artificial nests >30 m (Hamilton *et al.*, 2002) or >50 m (Spencer, 2002) apart to avoid density-dependant predation. We elected to place nests closer than these previous studies in order to match the environmental conditions across all four experimental treatments. Evidence of the order in which nests were excavated is informative in guiding the likely cues that were used to find the nest-sets, and we advise that future projects also consider methods of monitoring nest predation order to counter this issue. Due to their highly selective nesting habits, a similar outcome of mass predation is likely for turtle nests.

Depredated turtle nests

We found that the environmental factors around depredated turtle nests were not different from those of excavated artificial nests, indicating that artificial nests can be considered useful surrogates for the effects of environmental conditions on turtle nest survival. The number of predations of turtle nests recorded in this study ($n = 34$) is very likely a vast underestimate as only digs with egg remains were included, but foxes do not always leave eggs at the site (only 16.6% of excavations of artificial nests with eggs, E+, actually had egg remains left behind). Many other diggings were found across the study area which resembled the excavation pattern of depredated nests (100–150 mm deep, spoil pile to one side), but were excluded from analyses because there were no egg remains.

Methodology

Wilson *et al.* (1998) cautioned regarding the use of artificial nests as a surrogate for real nests for various reasons. In this

study, we used supermarket-sourced chicken eggs, however, infertile chicken eggs will produce a different olfactory signal to turtle eggs and therefore their use as a proxy for turtle eggs requires some caution (Burger, 1977). Furthermore, unfertilized chicken eggs will eventually rot, which would change olfactory cues over time (thus we terminated nest monitoring at 2 months post-installation). Biases may be associated with the use of artificial nests (Marchand *et al.*, 2002), such as a scent trail left by the researchers while checking or installing artificial nests (paradoxically, studies have shown that marking nests with human scent reduced depredation rate, Burke *et al.*, 2005). Human scent deposited by researchers during installation of artificial nests may 'cloud' olfactory cues, effectively removing the difference between treatments. Finally, foxes are intelligent predators, and while studies have shown that placing flags around turtle nests had no effect on the likelihood of predation by raccoons (Burke *et al.*, 2005), it is possible that the foxes learn to associate flagging with nests situated nearby. As all artificial nests were marked by a centrally placed marker, biases associated with flagged nests are unlikely to affect the relative patterns of predation (Marchand *et al.*, 2002).

In conclusion, although there are caveats around the use of artificial nests to understand nest depredation rates, such studies are helpful in revealing environmental factors that can influence nest depredation. We demonstrated that excavation of artificial nests was greater in more open areas closer to water. All predations were either confirmed as foxes through camera trapping (26 occasions), or were consistent with the description of fox predation, both from this and previous studies (e.g. Thompson, 1983). The nesting of *C. colliei* corresponds with specific weather patterns (occurrence of rain-bearing low-pressure systems, falling barometric pressure and air temperature >17°C, Clay, 1981), which means that many turtles nest over a small period of time. Furthermore, females also show strong habitat selectivity. The specific timing and localization of turtle nesting will contribute to increased vulnerability to fox predation, as foxes show evidence of developing search patterns based on visual and olfactory cues, and can excavate many nests in a small amount of time. Methods for mitigating the effect of fox predation on these freshwater turtles therefore needs to consider the long period over which fox depredation is acting (long-term solutions are required) as well as the habitat factors (e.g. soil type, weed control) that could make nests more vulnerable to excavation.

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References

- Baldwin, E.A., Marchand, M.N. & Litvaitis, J.A. (2004). Terrestrial habitat use by nesting painted turtles in landscapes with different levels of fragmentation. *Northeastern Naturalist* **11**, 41–48.
- Burbidge, A.A. (1967). *The biology of south-western Australian tortoises*. Perth: University of Western Australia.
- Burger, J. (1977). Determinants of hatching success in diamondback terrapin, *Malaclemys terrapin*. *Am. Midl. Nat.* **97**, 444–464.
- Burke, R.L., Schneider, C.M. & Dolinger, M.T. (2005). Cues used by raccoons to find turtle nests: effects of flags, human scent, and diamond-backed terrapin sign. *J. Herpetol.* **39**, 312–315.
- Burke, V.J., Rathbun, S.L., Bodie, J.R. & Gibbons, J.W. (1998). Effect of density on predation rate for turtle nests in complex landscapes. *Oikos* **83**, 3–11.
- Bush, B., Maryan, B., Browne-Cooper, R. & Robinson, D. (1995). *A guide to the reptile and frogs of the Perth region*: Nedlands: University of Western Australia Press.
- Chessman, B.C. (1978). *Ecological studies of freshwater turtles in south-eastern Australia*: Melbourne: Monash University.
- Christens, E. & Bider, J.R. (1987). Nesting activity and hatching success of the painted turtle (*Chrysemys picta marginata*) in southwestern Quebec. *Herpetologica* **43**, 55–65.
- Clay, B.T. (1981). Observations on the breeding biology and behaviour of the long-necked tortoise, *Chelodina oblonga*. *J. Roy. Soc. WA* **4**, 27–32.
- Congdon, J.D., Tinkle, D.W., Breitenbach, G.L. & van Loben Sels, R.C. (1983). Nesting ecology and hatching success in the turtle *Emydoidea blandingi*. *Herpetologica* **39**, 417–429.
- Congdon, J.D., Breitenbach, G.L., van Loben Sels, R.C. & Tinkle, D.W. (1987). Reproduction and nesting ecology of snapping turtles (*Chelydra serpentina*) in southeastern Michigan. *Herpetologica* **43**, 39–54.
- Doody, J.S., Green, B., Sims, R., Rhind, D., West, P. & Steer, D. (2006). Indirect impacts of invasive cane toads (*Bufo marinus*) on nest predation in pig-nosed turtles (*Carettochelys insculpta*). *Wildl. Res.* **33**, 349–354.
- Escalona, T. & Fa, J.E. (1998). Survival of nests of the terecay turtle (*Podocnemis unifilis*) in the Nichare-Tawadu Rivers, Venezuela. *J. Zool. (Lond.)* **244**, 303–312.
- Giles, J., (2001) The impacts of roads on the population dynamics and ecology of the Oblong Turtle (*Chelodina oblonga*) at Blue Gum, Booragoon and Piney Lakes. Honours Thesis, School of Environmental Science, Murdoch University, Perth, Western Australia.
- Guyot, G. & Kuchling, G. (1998). Some ecological aspects of populations of oblong turtles *Chelodina oblonga* in the suburbs of Perth (Western Australia). In *Le Bourget Du Lac*: 173–181. Miaud, C. & Guyétant, R. (Eds). France: Societas Europaea Herpetologica.

- Hamilton, A.M., Freedman, A.H. & Franz, R. (2002). Effects of deer feeders, habitat, and sensory cues on predation rate on artificial turtle nests. *Am. Midl. Nat.* **147**, 123–134.
- Hammer, Ø., Harper, D.A.T. & Ryan, P.D. (2001). PAST: paleontological statistics software package for education and data analysis. *Palaeontologia Electronica* **4**, 1–9.
- Lemmon, P.E. (1956). A spherical densiometer for estimating forest overstory density. *Forest Sci.* **2**, 314–320.
- Marchand, M.N. & Litvaitis, J.A. (2004). Effects of landscape composition, habitat features, and nest distribution on predation rates of simulated turtle nests. *Biol. Conserv.* **117**, 243–251.
- Marchand, M.N., Litvaitis, J.A., Maier, T.J. & DeGraaf, R.M. (2002). Use of artificial nests to investigate predation on freshwater turtle nests. *Wildlife Soc. B.* **30**, 1092–1098.
- McCutcheon, R.H.J. (1985). Observation on the breeding of the oblong turtle (*Chelodina oblonga*). *WA Nat.* **16**, 40.
- Parmenter, C.J. (1976). *The natural history of the Australian freshwater turtle Chelodina longicollis Shaw (Testudinata, Chelidae)*: Armidale: University of New England.
- Shine, R. & Iverson, J.B. (1995). Patterns of survival, growth and maturation in Turtles. *Oikos* **72**, 343–348.
- Spencer, R.-J. (2002). Experimentally testing nest site selection: fitness trade-offs and predation risk in turtle. *Ecology* **83**, 2136–2144.
- Spencer, R.-J. & Thompson, M.B. (2003). The significance of predation in nest site selection of turtles: an experimental consideration of macro- and microhabitat preferences. *Oikos* **102**, 592–600.
- Spencer, R.-J. & Thompson, M.B. (2006). Experimental analysis of the impact of foxes on freshwater turtle populations. *Conserv. Biol.* **19**, 845–854.
- StatSoft Inc. (2007) Statistica (data analysis software system), version 8.0. Retrieved from www.statsoft.com. Tulsa OK, USA.
- Thompson, M.B. (1983). Populations of the Murray River Tortoise, *Emydura* (Chelodina): the effect of egg predation by the red fox, *Vulpes vulpes*. *Wildl. Res.* **10**, 363–371.
- Tinkle, D.W., Congdon, J.D. & Rosen, P.C. (1981). Nesting frequency and success: implications for the demography of painted turtles. *Ecology* **62**, 1426–1432.
- Whelan, C.J., Dilger, M.L., Robson, D., Hallyn, N. & Dilger, S. (1994). Effects of olfactory cues on artificial-nest experiments. *The Auk* **111**, 945–952.
- Wilhoft, D.C., Del Baglivo, M.G. & Del Baglivo, M.D. (1979). Observations on mammalian predation of snapping turtle nests (Reptilia, Testudines, Chelydridae). *J. Herpetol.* **13**, 435–438.
- Wilson, G.R., Brittingham, M.C. & Goodrich, L.J. (1998). How well do artificial nests estimate success of real nests. *The Condor* **100**, 357–364.
- Zhao, B., Li, T., Shine, R. & Du, W.-G. (2013). Turtle embryos move to optimal thermal environments within the egg. *Biol. Lett.* **9**, 20130337.