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Evading the Ghost of Extinction: A Case Study for the Reintroduction of Ghost Bats (*Macroderma gigas*)

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Abstract: The ghost bat *Macroderma gigas* (Megadermatidae) is native to Australia. Its current distribution has dramatically contracted northwards in the past 150 years and the extant populations are scattered and isolated due to climatic and anthropogenic factors. To investigate the potential for reintroductions of wild ghost bats into suitable habitats, we examined the potential for refuges located in the southern-most parts of the species' former range to once again support populations. We identified Drovers Cave, located in Drovers Cave National Park in Western Australia, as a potential reintroduction site and used thermo-hygrochron iButtons to demonstrate that the microclimate conditions within the cave are suitable for ghost bats with temperatures close to 20 °C and 99% humidity throughout a ten-month period (December to September). We also showed that in the Pilbara region, ghost bats are opportunistic and flexible predators relying on small birds and mammal species. After comparing these data with the local fauna species list reported from Drovers Cave National Park, we concluded that the habitat at the proposed reintroduction site could provide sufficient diversity of potential prey species in terms of species richness, but we had no data on their relative abundance.

Keywords: ghost bat; *Macroderma gigas*; reintroduction; cave; microclimate; diet; Pilbara; Western Australia

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1. Introduction

The ghost bat, *Macroderma gigas* (Dobson, 1880) (Megadermatidae), is the largest microchiropteran bat native to Australia, with an average weight of 150 g [1]. This top-order predator once had a wide geographical range across the northern and central parts of the Australian continent [2] but its current distribution is patchy and in decline. The global population is represented by less than 10,000 mature individuals restricted to northern Queensland (NQ), the Northern Territory (NT), and the Pilbara and Kimberley regions of Western Australia. McKenzie and Hall [3] predicted that the species would decline by more than 10% within 25 years of that date, and Woinarski et al. [4] also estimated that the population was <10,000 individuals with an estimated continuing decline of >10% in 24 years (three generations). The reduction in the species' distribution has been most pronounced in southern and central Australia [5,6]. In Western Australia, ghost bats are present in the Pilbara and Kimberley regions. Recent environmental surveys carried out during the last decade in the Pilbara region [7] have indicated a population size of approximately 1850 ghost bats. McKenzie and Hall [3] estimated a total population of up to 4000 ghost bats in the Kimberley region. Nevertheless, this total population continues to decline in Western Australia, with the decline attributed to fewer suitable diurnal roost sites, human disturbance, habitat loss caused by mining activity, collisions with barbed wire fences erected to contain domestic cattle, geographical isolation among populations,

and the threat posed from ingesting cane toads (*Rhinella marinaris*) [8–10]. Ghost bats use diurnal refuges formed in natural rock formations and abandoned mine adits. Some refuges in old mining operations have been destroyed and others have begun to collapse [9,11]. Pastoral activities for beef cattle also have the potential to alter vegetation communities and the native fauna that they support, restricting prey diversity and abundance in the areas surrounding ghost bat roosts. Climate change within the next 100 years might create more suitable ambient conditions, but it is unclear whether ghost bats will be able to benefit from these changes [12]. As a result of this complex and worrying scenario, *M. gigas* is listed as a vulnerable species on the IUCN Red List, under Australian national environment legislation and under Western Australian, Queensland, and South Australian state legislation. Attempts have been made to construct artificial refuges for ghost bats in several parts of the Pilbara region [7], and ghost bats have made use of them within a short period of their construction, but there are no recorded cases of maternal roosts being established in these structures yet.

Management options to improve the conservation status of threatened species include increasing the number of populations and enhancing genetic diversity and resilience among extant populations [13]. When physical geographical barriers preclude the establishment of adequate corridors of connectivity, reintroductions, meaning 're-establishing a species in part of its historic range' [14], can be an effective conservation tool for the recovery of native threatened species [13]. Australia has a long history of reintroductions [14], with more than 380 translocations conducted for vertebrate species since 1880 [15], and with 233 of those conducted in Western Australia by 2016 [16]. However, to our knowledge, only one of those translocations has involved a bat species. In 2003, around 3000 grey-headed flying foxes (*Pteropus poliocephalus*: Megachiroptera) were relocated from the Melbourne Botanical Gardens (Victoria, Australia) to prevent further damage to the valuable flora of the gardens [17]. The translocated bats subsequently dispersed away from the release site. Although globally almost one in four bat species is facing the risk of extinction [18], to date the global number of bat translocations remains extremely low [19–21], and only a single case involving hand-reared orphan pipistrelle bats (*Pipistrellus* spp.) could be considered partly successful in the long term [22].

This study sets out the type of process that would likely be required to inform any decision to reintroduce ghost bats (or any other species of microbat). This planning process requires a sound knowledge of the behavioral ecology of the species along with a detailed assessment of the proposed release site(s) to ensure that adequate shelter and food are available, and that the underlying factors that contributed to the local extinction in the first place have been mitigated.

We proposed Drovers Cave, located within Drovers Cave National Park (DCNP) near Jurien Bay, on the mid-west coast of Western Australia (30°14′ S 115°05′ E), as a release site. The former range of ghost bats included this region but the question(s) as to when and why ghost bats disappeared have remained unclear for many years [23]. Some researchers claimed that climate change in the late Holocene increased aridity in the southern regions, which led to the contraction of the species' range [5,8,24]. However, others suggested an alternative interpretation of the impact of past climate: aridity would have suited ghost bats [25] and climate in the late Holocene in southern Western Australia was wetter; this would have increased vegetation density with more shelter for small prey leading to decreased food accessibility, as opposed to abundance, for the ghost bat [23,26,27]. Nevertheless, fossil and sub-fossil evidence indicate that the ghost bat was present in several caves in the area around Jurien Bay until quite recently. Guano piles and skeletal remains of ghost bats have been found at all depths, right up to the surface, in Drovers Cave, Moorba Cave, and Hastings Cave [27–29] (Figure 1). In addition, an observation of a living specimen was reported in the 1930s by F. Grigson, of Cockleshell Gully Farm (Western Australian Museum vertebrate paleontological collection registration number 67.4.37). This evidence suggests that the local extinction of the ghost bat in this district is relatively recent, and while the decline of many Australian mammals might have started under the influence

of climate change in the late Holocene, it may well have been expedited after Europeans arrived in Australia 200 years ago, given the habitat-specific nature of ghost bats [30,31]. The introduced European fox (*Vulpes vulpes*) is largely absent from the Kimberley region of Western Australia [32] and sparse in the Pilbara region [33] but foxes and feral cats (*Felis catus*) are common in southern parts of Western Australia where increased competition for prey [34,35] could also have decreased food supply for the ghost bat colonies. Mining of bat guano deposits in some of the larger caves in and around Drovers Cave [29] also may have resulted in unacceptable levels of human disturbance to ghost bat maternity roosts.

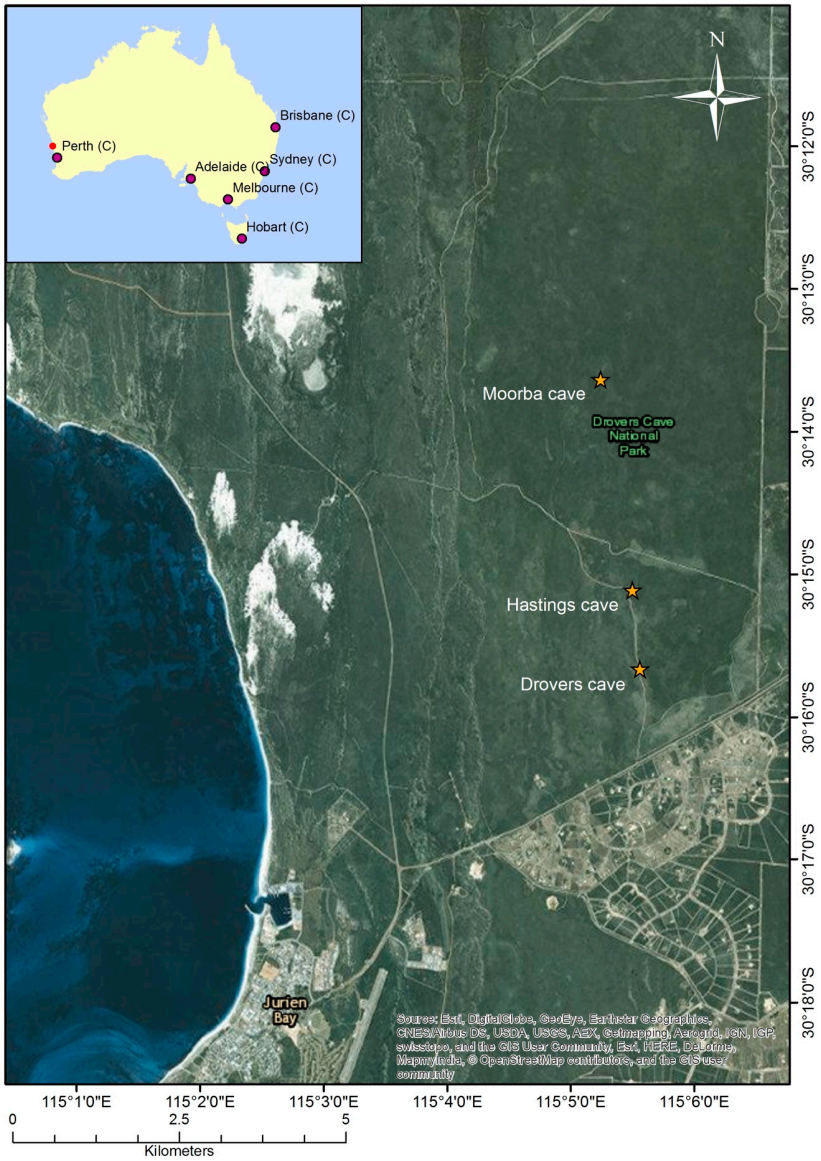


Figure 1. Map of Drovers Cave National Park, Western Australia. The red dot in the map of Australia indicates the location of Drovers Cave National Park and the stars in the larger map show the location of Drovers, Hastings, and Moorba Caves.

Drovers Cave National Park is managed by the Western Australian Department of Biodiversity, Conservation, and Attractions (DBCA), which carries out fire patch burning within the park. Feral cat and fox control have been advocated for this park but not implemented because of financial constraints. In 1969 the park authorities of the day erected a solid steel security door at the main entrance of Drovers Cave to prevent vandalism and the destruction of the geological formations within the cave [36]. Human access restriction has been shown to improve recovery of wild ghost bat populations [37]. In late 2015, DBCA

modified the security door to include a 300 mm-wide by 200 mm-high opening for the bats up to the size of ghost bats, which had been determined to be the smallest aperture through which ghost bats will fly [38].

Based on the recent history of occupation of caves within DCNP by ghost bats, we considered that Drovers Cave could meet all the reintroduction criteria. For this cave to be considered a suitable release site for the reintroduction of ghost bats we would need to demonstrate the following: (i) that the cave micro-climate was still within the temperature and humidity range of ghost bats; (ii) that the local fauna in the area around the cave was diverse and abundant enough to sustain a reintroduced population; and (iii) that the threatening processes that lead to the original local extinction were either no longer in effect or could be adequately mitigated.

2. Materials and Methods

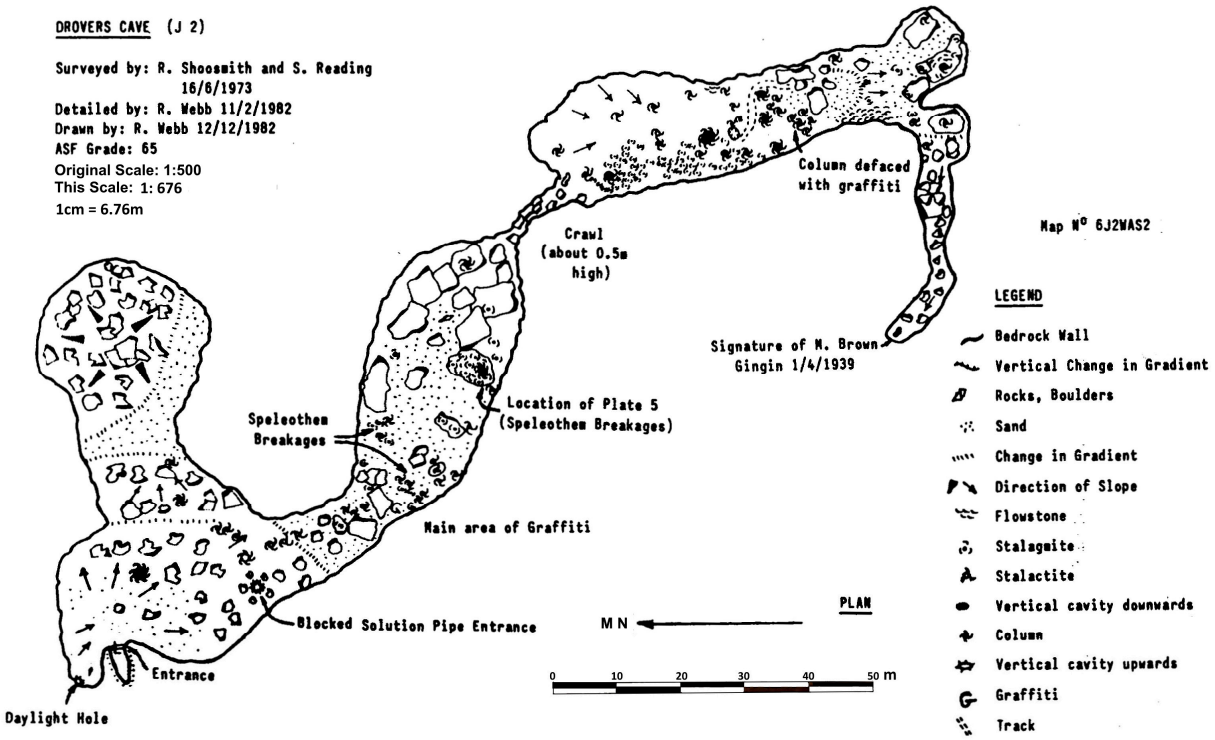
2.1. Microclimate Conditions at Drovers Cave

Drovers Cave National Park is located on the mid-west coast of Australia (Reserve No. 31302, 30°14′ S 115°5′ E; Figure 1). It covers an area of 2681 hectares of complex geological features [39,40] and broad extensions of sandplain. The native flora is highly diverse and composed mainly of proteaceous shrublands and woodlands [41,42] (Figure 2). This national park includes a series of limestone karst caves, with Drovers Cave being one of the larger at approximately 150 m long and 15 m deep, and with three chambers [36].

(**a**)

(**b**)

Figure 2. *Cont.*



(**c**)

Figure 2. (**a**) Main entrance to Drovers Cave; (**b**) sandplain plant community of the area composed mainly of proteaceous shrublands on sandy soils; (**c**) internal map of Drovers Cave (reproduced from Webb [36]).

Inside this cave we deployed 13 thermo-hygrochron iButtons (DS1923, Maxim Integrated Products, Inc., San Jose, CA, USA). Each of them was suspended from the cave wall ca. 200 mm from the point of attachment to simulate the position a ghost bat would occupy. We placed six iButtons in the first chamber and five in the second around the internal perimeter. Two iButtons were set on either side of the entrance and 5 m back from it. All iButtons were synchronized for time and set to measure temperature and relative humidity (RH) hourly from 16 December 2015 to 4 September 2016. It was intended that the iButtons would record data for a full 12-month period, but none of them either functioned for that long or remained undisturbed by members of the public illegally entering the cave. We calculated monthly temperature and relative humidity per iButton (from December 2015 to September 2016), as hourly changes were almost negligible. We then used a linear trend analysis to test for temporal and spatial correlation separately [43]. These linear models helped us to determine whether the temporal variable (months) and the spatial variable (chambers) had any effect on Drovers Cave's microclimate. Finally, post hoc tests (Bonferroni and Tukey tests) were used to detect which chambers or months differed significantly from each other to test for microclimatic stability. We also compared our data set (monthly mean $\pm$ S.E., maximum mean and minimum mean values) with the conditions outside of the cave to study its buffering capacity. Lastly, we determined the suitability of the cave's microclimate by comparing these mean values with the published records from more northerly ghost bat roost sites.

2.2. Food Supply at Drovers Cave National Park

To determine if Drovers Cave could once again support a colony of ghost bats, we aimed to examine the potential food supply in this habitat. We first analyzed old dried prey items collected from beneath ghost bat roosts located in five abandoned mines in the Pilbara region. Subsequently, we established a comparison between this data with the

NatureMap species database of DBCA [44] based on prey size. Since no recent surveys had been conducted in this national park, we were unable to obtain more accurate current data on the relative abundance of potential prey items. Former employees of the DBCA (AN Start and SV Leeuwen) donated unsorted dried prey remains collected from beneath ghost bat roosts located in the Pilbara region of Western Australia for this research. The samples were taken from dis-used copper (n = 3), gold (n = 1), and iron ore (n = 1) mines. Two of the copper mines were located west of Mt Newman (23°11′ S 119°25′ E; 23°20′ S 119°30′ E) with samples collected in 2005 and 2008, respectively, and the third mine west of the Maitland River (21°04′ S 116°57′ E) was sampled in 1986 and 1987. Material was also collected from an adit in the now-destroyed bulk sample area at the Marandoo iron ore mine (22°38′ S 118°06′ E) in 1990 and from the Klondyke Queen gold mine (21°20′ S; 119°53′ E) in 1994. The food remains included mainly body bones, rostra and mandibles, feet, feathers, and tails of prey species. Due to the spatial and temporal range in the sample sites, the prey remains were treated as a single pooled sample and not analyzed to determine any differences in prey species richness and diversity between source sites. Following our analysis, these remains were lodged with the Western Australian Museum.

To calculate diet diversity, we first classified all the dried material taxonomically by taking physical measurements using a Vernier caliper and observing their morphology with a stereomicroscope (OLYMPUS stereomicroscope LINE-UP SZX9, Magnification × 10–20; L.M.G. Scientific Services, Balcatta, WA, a division of James Optronics Pty Ltd., USA). We compared our samples with published information and specimen collections at the Western Australian Museum. In consultation with the curatorial staff, we were able to identify some of our dried material to the genus and species level. For dasyurid and rodent species, we looked at the size of the pre-molars and molars as well as the shape of the cusps. Bat species were identified from the skulls. We used wing feathers and foot morphology to identify bird species. For amphibians and reptiles, we determined the species based on skull morphology. Invertebrates, represented only by beetles, were identified to the genus level based on the morphology of the elytra. Diet diversity was calculated as the proportion of the number of prey species from one taxon in relation to the total number of prey species for pooled data across all sample sites.

It was not always possible to classify all dried material to the species level, so we calculated the relative abundance of prey to the next most appropriate, higher taxonomic level. We conservatively estimated the number of whole individuals from each taxonomic class present in the dried material samples (n = 215). Only mammals were divided into three orders: rodents, dasyurids, and bats. To avoid re-counting the same individuals, we considered only the largest number of a particular prey item from one specific taxon coming from the same location. For example, when analyzing birds collected from the first collection site, we counted either pairs of wing bones or legs, depending on which had the larger sample size. We calculated the minimum number of prey individuals in our samples by dividing the dried material according to the anatomy of the prey species. For instance, we divided by two the total number of insect elytra, feet, or wings of birds and bats. The total number of limbs of frogs, reptiles, dasyurids, and rodents were divided by four. Dis-articulated tails were not included in the data analysis, although they were numerous, since many were broken and we could not determine which fragments were part of the same tail. Detailed information on the full diet range identified from visual examination and subsequent metabarcoding examination are provided elsewhere [45,46].

2.3. Potential Prey Species at Drovers Cave National Park

We created a list of potential prey species that could be equivalent in size or body mass to the prey species from the Pilbara and recorded within a 10 km radius from Drovers Cave. Records from the NatureMap database [44] were collated for the period 1 January 1900 to 28 January 2016. We included records from such a long period of time due to the limited fauna survey effort that has been reported from the area around Drovers Cave. We classified the local species on this list according to simple categorical mass (g) and size (mm) ranges.

Data about body measurements were obtained mainly from published descriptions [47–49]. The mean mass was calculated by dividing the minimum and maximum value for females and males, when available, and then taking the mean of both sexes. The prey mass was ranked at three arbitrary levels: small (S), 1–33 g; medium (M), 34–67 g; and large (L), 67–100 g. We ranked amphibians and reptiles using snout–vent length instead of mass, because of limited data on mass in the literature, as small (S), 10–100 mm; medium (M), 101–200 mm; and large (L), 201–300 mm. These mass and size categories were chosen as they would likely relate to the capacity of ghost bats to detect prey, capture and subdue it, and then eat it in situ or to carry it back to their roost.

3. Results

3.1. Microclimate Conditions at Drovers Cave

We had a large but incomplete dataset due to stolen and failed iButtons, which created a wide variation in our sample size between months. This resulted in 6303 data record intervals and a total of 38,756 data points for temperature and 50,670 for RH. Fortunately, we had at least one iButton per chamber working for temperature and a minimum of two for RH. One of the two iButtons placed at the entrance stopped functioning after March and the other one after June 2016. Therefore, data comparisons between chambers should be interpreted with caution as they only refer to data collected until June 2016.

The average temperature and RH within Drovers Cave during the whole period was 19.41 °C and 98.86%, respectively. Our linear model demonstrated that the temporal variable (months) did not influence temperature and humidity values inside the cave (F_{Temp} = 1.763; df = 9; p = 0.098; F_{RH} = 0.384; df = 9; p = 0.939). Monthly mean temperature and RH remained constant, ranging only from 18.51 °C to 20.08 °C and from 97.35% to 99.64%, respectively (Table 1, Figure 3).

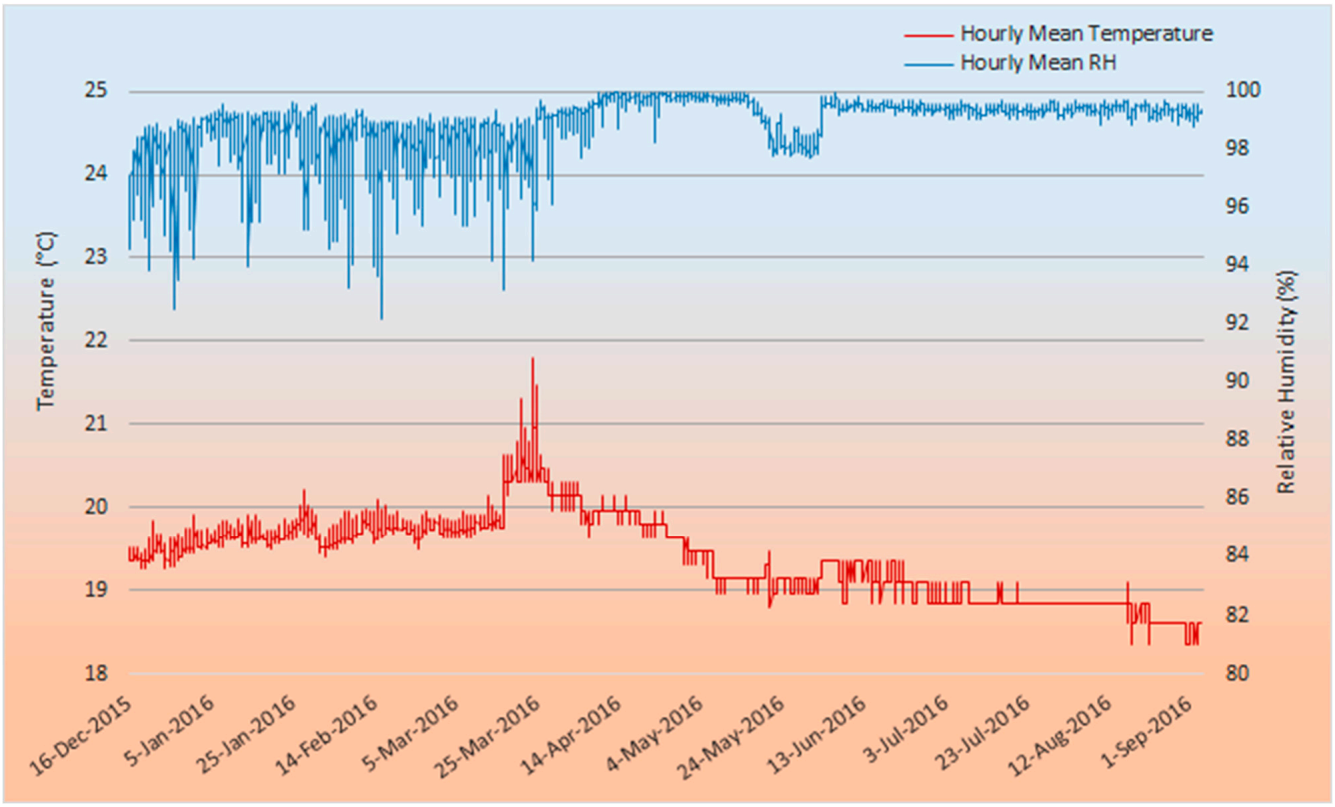


Figure 3. Hourly mean temperature (°C; red line) and relative humidity (%RH; blue line) from 13 iButtons placed in Drovers Cave from 16 December 2015 to 4 September 2016 (n = 2208). The timeline is divided into weekly periods.

Table 1. Monthly mean, maximum, and minimum temperature (°C) and relative humidity (%RH) for all the iButtons deployed at Drovers Cave. Ambient mean, maximum, and minimum values of temperatures and %RH at Jurien Bay (observations were drawn daily at 9 am from station 009131 Bureau of Meteorology (BOM)).

		December	January	February	March	April	May	June	July	August	September
Temperature (°C)	Mean ± S.E.	19.47 ± 0.006	19.69 ± 0.004	19.73 ± 0.004	20.08 ± 0.01	19.89 ± 0.01	19.18 ± 0.01	19.16 ± 0.01	18.88 ± 0.01	18.76 ± 0.01	18.51 ± 0.04
	Max–min	19.83–19.25	20.22–19.45	20.10–19.40	21.80–19.64	20.30–19.30	19.63–18.80	19.35–18.85	19.10–18.85	19.10–18.35	18.60–18.35
	Sample size	4836	9672	9048	6192	2160	2232	1490	1488	1488	150
	Ambient mean	23.9	24.8	25.8	24.5	20.1	16.0	13.3	13.0	13.3	14.9
	(Max–min)	(28.9–16.0)	(30.0–19.4)	(30.8–17.8)	(29.4–17.9)	(25.5–15.9)	(22.8–10.4)	(19.9–9.7)	(19.3–8.9)	(19.4–8.6)	(19.9–7.6)
RH (%)	Mean ± S.E.	97.35 ± 0.068	98.48 ± 0.036	97.86 ± 0.048	98.19 ± 0.06	99.64 ± 0.02	99.19 ± 0.03	99.41 ± 0.03	99.39 ± 0.02	99.40 ± 0.02	99.26 ± 0.08
	Max–min	99.02–92.52	99.62–93.95	99.36–92.18	99.73–93.16	100–97.73	100–97.69	99.95–97.83	99.70–99,05	99.70–98.83	99.60–98.80
	Sample size	4836	9672	9048	7584	5040	5208	3030	2976	2976	300
	Ambient mean	53	69	54	61	76	78	84	75	75	68

We detected significant differences in temperature and RH between the two chambers in Drovers Cave (F_{Temp} = 30.122; df = 2; $p < 0.001$; F_{RH} = 132.64; df = 2; $p < 0.001$). Bonferroni and Tukey tests showed that the average temperature of the cave measured at the entrance was around 1.7 °C warmer than the sections of the second chamber furthest from the entrance, and a difference of 0.73 °C between the first and the second chamber (p.bonferroni < 0.001; p.tukey < 0.001). For relative humidity, only the entrance differed significantly from the other two chambers, which were around 9% more humid (p.bonferroni < 0.001, p.tukey < 0.001).

The warmest conditions inside Drovers Cave were reached in March 2016 when the mean temperature inside the cave reached its maximum of 20 °C (Table 1). This increase was influenced by the iButtons placed closest to the entrance of Drovers Cave, which recorded temperatures above 25 °C. During March 2016, there were days when the ambient temperature at Jurien Bay exceeded 35 °C and the onshore wind speed was particularly strong and coming from the east-southeast [50]. Conversely, 18.5 °C was the minimum average temperature recorded at Drovers Cave throughout the month of September (Table 1), when the meteorological conditions at Jurien Bay were cold with a mean ambient temperature of approximately 15 °C [50]. In general, there was a small but gradual thermal decrease at Drovers Cave from December to September (Figure 3).

The maximum RH of 100% was often recorded by all the iButtons, except the two placed closest to the entrance. The most humid conditions in the cave, averaging over 99.6%, were detected during the month of April; however, maxima over 99% were recorded in all months (Table 1). The lowest RH mean recorded in Drovers Cave, of about 97%, was in December when the meteorological conditions at the nearest meteorological station located on the coast at Jurien Bay were much drier (RH 53%). Overall, Drovers Cave maintained high humidity for the whole study period, with monthly and hourly mean values no lower than 92% (Figure 3).

3.2. Food Supply at Drovers Cave National Park

Prey Species and Their Relative Abundance in the Pilbara Region

The ghost bat diet derived from the dry material collected from beneath diurnal roosts in the five caves/adits in the Pilbara region contained a high number of small mammals (15 species; 42% of total species consumed) and birds (11 species; 30.5%). Within the mammal remains, dasyurid species (17%) outnumbered rodent (14%) and bat (11%) species. Reptile, amphibian, and insect species were less represented, with only five species identified (Table 2).

Table 2. Dversity in ghost bats (*Macroderma gigas*) prey, expressed as the percentage of prey species in each taxonomic group, determined from regurgitated pellets collected from diurnal refugia in the Pilbara region of Western Australia.

Class	***Insecta***	***Amphibia***	***Reptilia***	***Aves***	***Mammalia***			**Total Species No.**
Order	1	1	1	4		3		
					Dasyuridae	*Rodentia*	*Chiroptera*	
Family	1	2	4	9	1	1	3	
Genus	4	2	4	10	4	4	3	
Species	4	2	4	11	6	5	4	36
Percentage (%)	11	5.5	11	30.5	17	14	11	100

In terms of abundance, mammals represented more than two thirds (69.7%) of the total number of prey items identified, and birds nearly a quarter (24.2%). Within mammals, rodents were the most abundant prey (44.7%) followed by bats (19.1%) and dasyurids (5.6%). Insects had a slightly higher relative abundance (3.7%) compared to amphibians (0.9%) and reptiles (1.9%) (Figure 4).

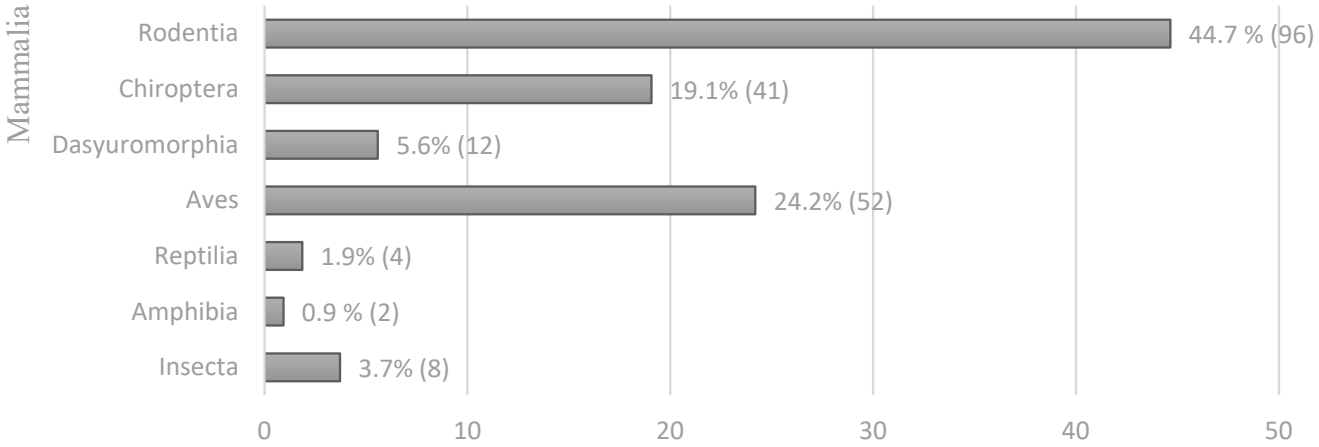


Figure 4. Prey abundance determined from material collected beneath ghost bat (*Macroderma gigas*) roosts at five mines/adits in the Pilbara region. The bar graph shows the proportion (as a % of the total number of prey items; n = 215) with the sample sizes in brackets.

3.3. Potential Prey Species at Drovers Cave National Park

A comparison between the abundance of different-sized prey from the Pilbara and DNCP showed that ghost bats in the Pilbara have a preference for small amphibians that weigh less than 34 g, which was also the only size range of amphibians found in DCNP and was represented by seven species (Table 3). With respect to reptiles, ghost bats in the Pilbara have a preference for medium-sized species (75% of recorded species), and at DCNP there are mainly medium- (55%) and small- (32%) sized reptiles (Table 3).

Table 3. Relative abundance of ghost bat (*Macroderma gigas*) prey species according to their size in the Pilbara, based on the remains of prey species retrieved from roosts, and Drovers Cave National Park, based on the NatureMap database [43]. The prey mass was ranked from small (S) (1–33 g), to medium (M) (34–67 g), to large (L) (67–100 g). Amphibians and reptiles were classified using snout–vent length as small (S): 10–100 mm; medium (M): 101–200 mm; or large (L): 201–300 mm.

Taxa	Location	Ranked Mass/Length	No. Species	No. Records	Percentage of Total Individuals
Insects	Pilbara	S	4	8	100
		M	0	0	0
		L	0	0	0
	Drovers Cave NP	S	12	18	100
		M	0	0	0
		L	0	0	0
Amphibians	Pilbara	S	2	2	100
		M	0	0	0
		L	0	0	0
	Drovers Cave NP	S	7	48	100
		M	0	0	0
		L	0	0	0
Reptiles	Pilbara	S	0	0	0
		M	3	3	75
		L	1	1	25
	Drovers Cave NP	S	9	160	32
		M	11	279	55
		L	2	65	13

Table 3. *Cont.*

Taxa	Location	Ranked Mass/Length	No. Species	No. Records	Percentage of Total Individuals
Birds	Pilbara	S	6	24	58
		M	3	15	37
		L	2	2	5
	Drovers Cave NP	S	25	220	92
		M	3	5	2
		L	1	14	6
Mammals	Pilbara	S	11	87	70
		M	4	38	30
		L	0	0	0
	Drovers Cave NP	S	6	317	100
		M	0	0	0
		L	0	0	0

In the Pilbara region, small perching bird species such as budgerigars (*Melopsittacus undulatus*) were the most abundant (58%) prey species, and they outnumbered medium-sized birds in terms of species and numbers (37%). Only two large bird species weighing more than 66 g were found in the dried material we examined; these were the grey-crowned babbler (*Pomatostomus temporalis*) and the magpie-lark (*Grallina cyanoleuca*). At DCNP, a large proportion of bird species (92%) recorded in the area are in the small mass/size range, and there are only a few medium-sized species, such as some of the larger honeyeater species (*Anthochaera* spp.) and the brown songlark (*Megalurus cruralis*) (Table 3). Ghost bats in the Pilbara consumed 70% small-sized mammals and 30% medium-sized mammals. At DCNP, we found a large number of small-sized mammal species, especially rodents (233 records), but also one bat species (34 records), and possibly three other species (lesser long-eared bat (*Nyctophilus geoffroyi*), Gould's wattled bat (*C. gouldii*), and Southern forest bat (*Eptesicus regulus*)). None of the vertebrate species recorded from the area around DCNP are currently listed as threatened species (https://www.dbca.wa.gov.au/wildlife-and-ecosystems/animals/list-threatened-and-priority-fauna; accessed on 4 July 2024) or has suffered notable declines in abundance during the 100-year time frame from which the records were drawn.

4. Discussion

Many bat species around the world are categorized as threatened by the IUCN due to numerous and diverse threats [18]. Bats are sensitive mammals that rely on appropriate conditions at their diurnal roost sites [51]. In Western Australia, the loss of suitable roost sites and habitat clearance with the resultant decrease in prey abundance have had a detrimental impact on ghost bat populations [9]. Here, we investigated the suitability of Drovers Cave, a site that supported an extant population until the 1920s, as a potential reintroduction site.

Our results show that the thermal buffering capacity of Drovers Cave is remarkably efficient. When the main rainfall period (late May to August) at Drovers Cave commenced, the rainfall readily moved through the shallow aeolian soils and limestone karst and into the cave system leading to a reduction in variability in the recorded RH during the austral winter months, despite the declining number of operating iButtons. We also did not want to risk losing all remaining iButtons that retained data. While our data values are lower than the 23–27 °C reported in the literature for ghost bats [37,51,52], thermo-physiological studies have demonstrated that ghost bats can maintain their body temperature over 35 °C when external temperatures range from 15–37 °C [51], although they have a wider tolerance, from 0–38 °C [53]. This capability to tolerate lower ambient temperatures is further supported by the observations made by [54] of ghost bats foraging at night for

varying lengths of time up to all night when minimum ambient temperatures ranged from 10–15 °C and relative humidity in the dry season was as low as 8% (measured at 0900 Hr). All the published data for ghost bat roost temperatures come from locations in the tropical north of Australia and are understandably higher than what might be expected at the southern limits of the species' former and recent distribution. In addition, their adult survival seems not to be affected when the ambient temperature drops below 10 °C during winter [55]. In their experimental studies, Leitner and Nelson [53] observed that ghost bats displayed physiological responses to compensate for heat loss below 20 °C; however, bats did not enter torpor or decrease their body temperature. It must be noted that bats used in that study were kept alone with no chance to form colonies or select warmer areas of their roost, as they would do under conditions such as those at Drovers Cave where the mean temperature at the entrance can be above 20 °C. In addition, all Leitner and Nelson's [53] experiments were performed using a circulating air system of dry room air, which might have enhanced heat loss. Drovers Cave, with its high RH (ca. 99%), could ameliorate evaporative water and heat loss, and therefore, ghost bats might be able to maintain a high body temperature even at lower ambient temperatures [37].

An important advantage of the thermal values we obtained is the capacity to restrict the distributional expansion of the cane toad (*Rhinella marina*), as recent studies consider the consumption of this toxic species a significant factor in the decline of northern populations of ghost bats [10,56]. Cane toads experience physiological constraints in locomotion under cold climates of $\leq$20 °C and the risk of desiccation [57,58] and thus are not likely to survive the conditions recorded in the DCNP. The lack of any permanent surface water is also likely to prevent cane toads from occupying the area around Drovers Cave.

Baudinette et al. [51] highlighted the importance of water balance and regulation, and how this physiological dependency can restrict the number of suitable roosting sites that bat species occupy. Given that ghost bats have been recorded from habitats with a measured RH of ca. 80% [52], Drovers Cave should provide a suitable microclimate for them. High humidity (>90%) was maintained inside the cave, even when external local summer conditions at Jurien Bay were extremely dry (ca. RH 20%). This high RH recorded within Drovers Cave could ameliorate evaporative water loss, which will reduce the compensatory need for metabolic water production. At times, there is limited free water on the floor of Drovers Cave (typically in winter) or water flowing over the external surfaces of stalactites. Therefore, any reintroduced bats would require less water intake, which might be an advantage during summer when rainfall is minimal in the area.

Both Hastings Cave and Moorba Cave (Figure 1) have been recorded as important roost sites for large numbers of ghost bats in the past, as evidenced by their substantial guano deposits [27–29]. Ghost bats can disperse considerable distances [37,54] and are able to fly 21–22 km/h [54,59]. Therefore, reintroduced ghost bats could travel the short distance (<5 km) to these other caves (Figure 1), and the length of the entire national park (2681 ha) could be flown in less than 2 h.

The results of our study confirmed that ghost bats capture a broad range of prey items, as also reported by other researchers [9,23,37,44,45,56,59,60]. A recent satellite tracking study of ghost bats in the Pilbara has indicated that this species can forage over areas up to 105 $\pm$ 133 Ha (range 20–420 Ha) in size [54]. Ghost bats have also been shown to hunt alone and in groups, and that once caught, the smaller prey items are carried back to their roosting sites before being eaten [23], which might explain the size range of prey items recovered beneath roosts.

The examination of the dry prey material revealed that ghost bats fed on mammals, birds, reptiles, amphibians, and insects, which indicates their importance as a top-order predator in the ecosystem. With specific reference to vertebrate prey species, ghost bats showed a clear preference for mammals (>40% of species identified), with dasyurid species outnumbering rodent and bat species, but it is not clear if this result represents hunting preferences of the bats or the structure of the available prey community and that part that could be transported back to the roosts. However, in terms of prey abundance, rodents

and bats were more frequently hunted than dasyurids (45%, 19%, and 5%, respectively). From the identifiable species, the introduced house mouse (*Mus musculus*) was the most abundant, being twice as common as the next species, the common rock-rat (*Zyzomys argurus*), followed closely by the bat species *Taphozous* spp. and the spinifex hopping-mouse (*Notomys alexis*). Start et al. [45] identified another four species of bats from the same midden material based on long wing bones, but they did not provide any information on the relative abundance of any of those additional species. That study increased to nine the number of bat species preyed upon by ghost bats in the Pilbara.

Birds made up almost one quarter of all the identified prey species. The little button-quail (*Turnix velox*) was the most frequently recorded species, followed by the budgerigar, singing honeyeater (*Gavicalis virescens*), and Horsfield's bushlark (*Mirafra javanica*). These findings are consistent with those of Boles [60], who recorded that ghost bats feed on more than 50 different avian species, and other studies [56,61].

Reptiles, amphibians, and insect species were less common in the prey items retrieved from the remains collected from the Pilbara roosts, and our findings are similar to other published studies [37,62]. The quantity of scat material we identified containing these classes of prey was small; therefore, data should be interpreted with caution due to the small sample sizes (n = 8 insects, n = 2 amphibians, n = 4 reptiles). There are some possible explanations for these results. Firstly, ghost bats are opportunistic predators [37,59] that hunt whatever is most readily available in terms of both abundance and accessibility. In the Pilbara region where small rodents and birds are common, ghost bats might have no need to hunt insects with their lower nutritional value. The lack of water in many parts of the Pilbara during most of the year might simply mean that frog populations are small or restricted to habitats remote from ghost bat roost sites. Lastly, ghost bats have powerful jaws and large teeth (for a microbat) that could easily mill thinner bones such as those of frogs and small reptiles, allowing them to eat the whole prey item without discarding any remains.

Most of the classified specimens fell within the small-sized category, possibly due to the increased effort required to lift larger prey items and fly back to the roosts with them. Boles [60] showed that 70% of the 50 avian species hunted by ghost bats weighed less than 35 g. In the same way, Johnston et al. [61] reported the Australian owlet-nightjar (*Aegotheles cristatus*), with a weight of 46 g, was the largest bird species preyed on in Karijini National Park (Western Australia). In the Northern Territory, avian species as large as the dollar bird (*Eurystomus orientalis*), with a mass up to 125 g, have been reported to be prey items of ghost bats [62]. In our study, we recorded two large avian prey species (67–100 g): the grey-crowned babbler and the magpie-lark. This is consistent with laboratory studies performed with live prey demonstrating that ghost bats can lift 60 g mice and drag 100 g rats which represent up to 80% of the ghost bat's body mass [63].

When comparing this dietary information with species present at DCNP, we had to create a new list of potential prey species for ghost bats in this area. In Western Australia, potential prey species distribution varies on both spatial and temporal scales. None of the potential prey species identified are currently listed as threatened species [64]. There is considerable turnover in species between the Pilbara and Geraldton Sandplains of Western Australia, and there are seasonal variations in both presence and absence of some prey species due to migratory movements for birds, and behavioral changes in response to rainfall and declining ambient temperatures for frogs and reptiles. Given that the Pilbara and Drovers Cave are separated by more than 1300 km and a substantial desert, it is not surprising that the species we found in the Pilbara differed from those likely to be available at DCNP. We consider that at Drovers Cave seven amphibian species, 22 reptile species, 29 bird species, and six mammal species could be potential prey for reintroduced ghost bats, with the majority falling in the small (1–33 g) size/weight range. The chocolate wattled bat (*Chalinolobus morio*) was the fourth most commonly (34 records out of a total of 317 mammal records from within 10 km of Drovers Cave) reported mammal species in DCNP [43], and given that Start et al. [45] recorded four species of micro-bat in the diet of

ghost bats in the Pilbara, chocolate wattled bats could provide an important food source for the ghost bats. We identified 25 small avian species (220 records) that could also provide a prey source, including species such as the New Holland (*Phylidoniris novaehollandiae*) and brown (*Lichmera indistincta*) honeyeaters, the white-browed scrubwren (*Sericornis frontalis*), the silvereye (*Zosterops lateralis*), and the welcome swallow (*Hirundo neoxena*). The little button-quail, which was a very common prey item in the Pilbara, was not reported in the Birds Australia Atlas database from the vicinity of Drovers Cave, and Johnstone and Storr [48] also indicate that it is absent from coastal mesic parts of the Geraldton Sandplains. The presence of suitable prey species may not mean they are able to provide food for ghost bats, as the shrubby kwongan heath vegetation [41] present at Drovers Cave may not afford ghost bats the same hunting opportunities as those present in the more arid Pilbara locations.

DCNP supports large numbers of small-sized mammal species, especially rodents like the ash-grey mouse (*Pseudomys albocinereus*) and house mouse. No medium/large mammal species remain extant in the Drovers Cave area, which is largely a consequence of population declines/extinctions associated with the introduction of foxes and cats to the region and possibly the limited field surveys conducted in the area in recent times.

Given that more than 100 years have passed since ghost bats were last recorded in Drovers Cave and that we could not provide any data on prey abundance, it would be incumbent on any proposer for a ghost bat reintroduction to demonstrate in the requisite Translocation Proposal that their re-establishment at this site would not pose a threat to any prey species.

In order to establish the first colony of reintroduced ghost bats in Western Australia and guarantee its long-term viability, it is necessary for a management plan to be developed for DCNP that addresses a number of key issues, including fire management, control of introduced predators/competitors for prey, and effective management of human access to caves that are suitable habitats for ghost bats. This reserve area is situated in a semiarid region of Western Australia, which is prone to wildfire. Wildfires are unlikely to affect ghost bats directly in their roosts; rather, they will indirectly affect the bats through changes in the vegetation and prey population dynamics [65]. Researchers have investigated the effect of fires on small mammals in semiarid regions of Western Australia [66] and demonstrated that, contrary to what might be expected, rodents preferred to forage in recently burnt areas, and that fire regimes of approximately 10-year cycles help to conserve both plant and rodent communities. Proteaceous vegetation communities such as the one found in DCNP typically recover to pre-fire status in 11–15 years [67,68]. However, it is important to note that none of the vegetation communities studied in these studies are represented at DCNP. The only published study to examine the impact of any burning regime on ghost bats suggested that ghost bats tended to return to burnt areas as soon as the habitat and the vegetation have been regenerated [69]. The DBCA has carefully managed prescribed fires within DCNP in recent years (2003, 2007, 2011, 2014) and a fire management plan for the whole district of Jurien is under development (L. Strumpher, pers. comm.). Another key element of park management that needs to be implemented as soon as possible is feral predator (fox and cat) control to reduce competition for prey species important to ghost bats. Illegal access to the cave by humans will also need to be adequately managed given the demonstrated sensitivity of ghost bats to disturbance.

5. Conclusions

Our studies have shown that Drovers Cave has suitably humid conditions for the ghost bat and that its thermal tolerance would likely allow it to cope with the lower mean temperature in the cave. We have demonstrated that there is likely adequate richness in prey species within DCNP, but there are insufficient data at present to determine whether prey species abundance is adequate. This study represents the first steps taken towards a strategic action aimed to prevent the continued decline of this unique bat species. This

study also provides a working template for other bat researchers to consider and adapt to support conservation actions involving reintroduction programs of other micro-bat species.

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