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Cover image: A white-rumped vulture *Gyps bengalensis* sitting out a rain shower in Siem Pang Wildlife Sanctuary, June 2020 (© Jeremy Holden). Legrand *et al*. describe the discovery of a breeding colony of this Critically Endangered species on the Cambodia/Laos border on pp. 12–16 of this issue.

News

New Bachelor of Science in Agroecology at the National University of Battambang

The Cambodian economy is dependent on agriculture and most of the country's population is engaged in the sector. One third of the kingdom's population is aged between 14 and 30 years old, making these and the educational system an important entry point to extend target technologies to farming families. To ensure the country's future competitiveness, skilled graduates in STEM (science, technology, engineering and maths) fields and agricultural technologies are crucial to transition the national economy to higher-skilled and more productive industries.

A new curriculum on Agroecology has been developed by the National University of Battambang through a higher education improvement project, with technical support from the University of Sydney (Australia) and a variety of experts from France, USA and Cambodia. The BSc in Agroecology is the only academic programme of its kind in Cambodia and offers comprehensive education and training on sustainable farming methods that work with nature.

The BSc in Agroecology is future-focused to support the development of sustainable farming practices that integrate with natural systems and reflect important ecological principles and applied agriculture concepts. The program resonates strongly with the UN Sustainability Goals and One-Health principles in championing:

- farming practices that reduce emissions, recycle resources and prioritise local supply chains;
- biodiversity conservation, aiming to increase reliance on approaches such as integrative pest management and practices that improve soil health including the microbiome; and
- approaches led by local people and adaptive agricultural techniques to suit local contexts i.e. providing solutions for specific social, environmental and economic conditions.

The programme is designed to meet the needs of the labour market in Cambodia and stakeholder feedback was employed to define its intended learning outcomes. The BSc prepares students to become agroecologists, with comprehensive and contemporary knowledge of food systems which are critical for employment in private enterprises, government institutions and non-government organizations. These have an important role to play in moving Cambodian agriculture from mono-cropping to agrobiodiversity practices to improve the yields of staple crops and environmental health in the country. Further information can be obtained by contacting the first author below.

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Short Communication

What's in the water: using environmental DNA metabarcoding to detect fish biodiversity in the Cambodian Mekong

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Globally, freshwater biodiversity faces increasing threats (Dudgeon *et al*., 2006) and the fish community of the Lower Mekong Basin (LMB) exemplifies this trend. Climate change, hydroelectric development, and an increasing human population are major stressors in the basin (Lauri *et al*., 2012; Pokhrel *et al*., 2018; Yoshida *et al*., 2020), which covers approximately 571,000 km^2 across Cambodia, Thailand, Laos and Vietnam. In Cambodia, the Mekong enters the country at its border with Laos and flows 480 km south to the border with Vietnam. As it flows southward it is fed by large tributaries, including the Sekong, Sesan and Sre Pok (3S) rivers that drain southern Laos, central Vietnam and northeastern Cambodia. In addition, Southeast Asia's largest lake— the Tonle Sap—lies in western Cambodia. As a tropical watershed with a recurring wet season and associated flooding of expansive wetlands, the lake supports a productive and diverse community of over 1,000 fish species (Hortle, 2009a; Rainboth *et al*., 2012). The seasonal flood pulse drives fish abundance in the basin, as species are adapted to utilize seasonally available, highly productive habitats (Poulsen *et al*., 2002). This ecosystem supports an annual fish harvest in excess of two million tons and consisting of hundreds of species, providing food security for over 70 million people (Hortle, 2009b; FAO, 2020). This means that monitoring the fish community is essential, both as an early warning system for biodiversity losses and for evaluating impacts of conservation measures. However, Cambodia exemplifies challenges common to biodiversity monitoring in large, tropical river systems, including difficulties with accessing remote areas, extreme seasonal conditions, resource-limited management agencies, and a need for numerous gears and significant expertise to capture and identify hundreds of species. Given the growing stressors in the region, developing approaches for effective biodiversity monitoring is particularly critical.

Collection of genetic material from the environment (eDNA) is a non-invasive and increasingly applied approach to characterize biodiversity in freshwater systems and monitor for endangered species (Deiner *et al*., 2016; Evans & Lamberti, 2018; Doi *et al*., 2021; Laporte *et al*., 2021; Yao *et al*., 2022). Notably, eDNA metabarcoding is highly sensitive and has the potential to detect greater numbers of species compared to traditional capture methods (McColl-Gausden *et al*., 2021). Although applications of metabarcoding in tropical rivers have been challenged by a lack of reference sequences (Jerde *et al*., 2021), the technology is beginning to be applied in Southeast Asia. For example, metabarcoding has been used in the Chao Phraya Basin of Thailand to detect patterns in biodiversity to inform conservation efforts (Blackman *et al*., 2021), and it has been used in the LMB to distinguish patterns in fish diversity across ecological gradients and evaluate hypothesized barriers to fish dispersal (Durand *et al*., 2022).

We performed a pilot evaluation of the feasibility of metabarcoding for quantifying fish diversity in the Cambodian Mekong through the collection of water-

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borne eDNA. Sampling occurred between February and April 2022 throughout the Mekong River; the 3S rivers; and the Tonle Sap (Fig. 1, Table 1). Collection of eDNA samples was permitted by the Cambodian Fisheries Administration and conducted with support from the Inland Fisheries Research and Development Institute.

Samples were collected with single-use aquatic eDNA kits (Jonah Ventures, Boulder, Colorado, USA), which included nitrile gloves, a 60 ml syringe, a 5 μm filter cartridge, and a 1 ml syringe of Longmire's solution to stabilize DNA for storage and transport. These kits were selected because no special equipment is required

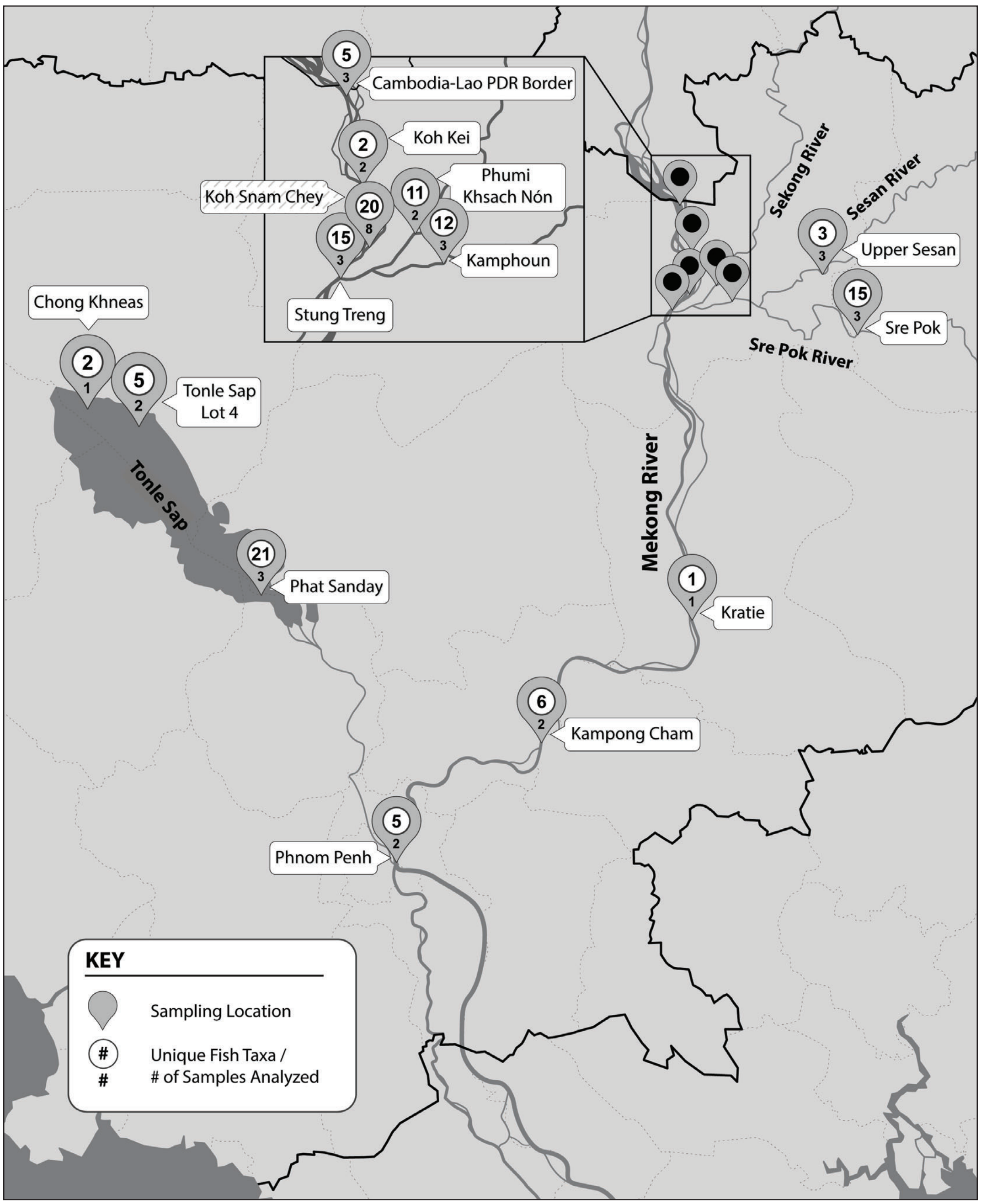

Fig. 1 The Lower Mekong Basin, Cambodia and locations where eDNA samples were collected. Each location is labelled with the total number of taxa detected in metabarcoding analysis, as well as the total number of samples from that location which yielded results.

Table 1 Locations, filter volumes and biodiversity detected in samples. Samples in which no DNA sequences were detected or for which no sequences could be assigned to fish taxa are excluded from the summaries of volume and taxa detections.

Site	# Samples Yielding Results / # Samples Collected	River	Mean Volume Filtered per Sample Kit (ml)	Total Volume Filtered at Site (ml)	Unique Fish Taxa	Families	Genera	Species
Border	3/9	Mekong	180	540	5	2	5	5
Koh Kei	2/3	Mekong	175	350	2	2	2	2
Koh Snam Chey	8/9	Mekong	150	1200	20	7	17	19[1]
Stung Treng [2]	0/3	Mekong	-	-	-	-	-	-
Kratie	1/3	Mekong	180	180	1	1	1	1
Kampong Cham	2/3	Mekong	120	240	6	4	5	5[1]
Phnom Penh	2/3	Mekong	120	240	5	5	5	5[1]
Stung Treng	3/3	3S (Sekong)	120	360	15	8	13	14
Phumi Khsach Nón	2/3	3S (Sekong)	120	240	11	9	10	11[1]
Kamphoun	3/3	3S (Sesan)	100	300	12	6	11	11[1]
Upper Sesan	3/3	3S (Sesan)	125	375	3	2	3	3
Sre Pok	3/3	3S (Sre Pok)	120	360	15	7	12	15[1]
Chong Khneas	1/2	Tonle Sap	30	30	2	2	2	2
Tonle Sap Lot 4	2/3	Tonle Sap	38	75	5	3	3	4
Phat Sanday	3/3	Tonle Sap	20	60	21	11	17	21[1]
Totals	38/56			4.55 l	123	69	106	31

[1] Exact sequence variants that could only be assigned to genus level were detected in these sites. Each of these is included in the species count as a single species for the site where it was detected. Fish taxa at the family and order level are not included in the genera or species counts. See Table 2 for details.

[2] Samples collected from the mainstem Mekong River in Stung Treng city failed to generate any sequence reads. This site is not included in Fig. 1, and the only Stung Treng site shown is that on the Sekong River above the confluence in Stung Treng city.

and samples stabilized with Longmire's solution do not require refrigeration in the field. Further, the filters are enclosed in a cartridge that reduces potential for contamination, and the single-use nature of the kits eliminates the need for decontamination of equipment. As an additional precaution to reduce contamination, field staff were instructed to collect upstream of where they were wading or of the boat used to reach the sample location. At most locations, a sample was collected from the right bank, middle of the channel, and the left bank, for a total of three samples per location. Samples were collected approximately five centimetres beneath the water's surface in all locations except for the site on the Mekong River near Koh Snam Chey (shaded label in Fig. 1 inset), where three samples apiece were collected at depths of 1, 35 and 60 m (Table 1).

DNA metabarcoding employed MiFish primers (Miya *et al*., 2015), which target the 12S region of the mitochondrial genome and are known to provide genetic resolution to the species level. PCR amplification was performed in replicates of six, none of which were pooled. Each round of PCR included a non-template control to identify any laboratory cross-contamination. Metabarcoding produced hundreds of thousands of sequences, which were processed using a custom bioinformatics pipeline that summarized the number of unique exact sequence variants (ESV) amplified in each water sample. ESV assignments were based on percent similarity to reference sequences from GenBank (release 248), plus five unpublished sequences from specimens sequenced by Jonah Ventures (*Clupeoides borneensis*, *Henicorhynchus entmema*, *Puntioplites falcifer*, *Trichopodus trichopterus* and *Ompok siluroides*). A full description of laboratory methodology can be found in the supplemental materials of Campbell *et al*. (2022). A recursive matching algorithm assigned ESVs to known species according to sequence similarity, and if below a similarity threshold ESVs were assigned to higher taxonomic levels. In most cases, ESVs were designated to species, but genus, family, and order level assignments did occur. We then used R statistical

Table 2 Fish species detected in 38 eDNA samples collected throughout the Lower Mekong Basin. Two exact sequence variants (ESVs) were only assigned to Siluriformes, six were only assigned to Pangasiidae, and four were only assigned to Cyprinidae and are not explicitly included in this table, although these orders and families are represented at each site by other genus and species level assignments. For taxa identified only to the genus level, the number of unique ESVs assigned to that genus is provided in the species column. The number of ESVs assigned to each species are given in parentheses after the species name. VU=Vulnerable, EN=Endangered, CR=Critically Endangered, per IUCN (2022). Scientific names are based on NNEF (2021).

Species (# of ESVs)	Mainstem Mekong						3S Basin					Tonle Sap		
	Border	**Koh Kei**	**Koh Snam Chey**	**Kratie**	**Kampong Cham**	**Phnom Penh**	**Stung Treng**	**Phumi Ksach Nón**	**Kamphoun**	**Upper Sesan**	**Sre Pok**	**Chong Khneas**	**Tonle Sap Lot 4**	**Phat Sanday**
Osteoglossiformes														
Notopteridae														
Chitala ornata (2)					X									
Notopterus notopterus (3)	X								X					X
Clupeiformes														
Clupeidae														
Clupeichthys aesarnensis (5)									X	X				
Clupeoides borneensis (1)												X	X	
Sardinops sp. (1)										X				
Engraulidae														
Lycothrissa crocodilus (2)						X								
Cypriniformes														
Cyprinidae														
Amblyrhynchichthys micracanthus[1] (2)											X			
Barbonymus altus (7)			X				X		X		X			
Barbonymus gonionotus (10)	X		X					X			X			X
Catlocarpio siamensis[2] CR (2)													X	
Cirrhinus microlepis VU (5)	X		X						X					
Cosmochilus harmandi (1)			X											
Cyclocheilichthys apogon (1)														X
Cycloheilos enoplos (5)		X							X					
Epalzeorhynchos sp. (2)											X			
Hampala dispar (2)			X											
Henicorhynchus entmema (2)							X							
Henicorhynchus siamensis (3)					X		X							X
Hypsibarbus malcolmi (2)	X													
Labeo chrysophekadion (3)											X			
Labiobarbus leptocheilus (1)				X			X							
Labiobarbus sp. (2)														X
Mystacoleucus marginatus (3)			X					X	X					
Onychostoma meridionale (4)			X						X					
Osteochilus vittatus (4)														X

Table 2 Cont'd.

Species (# of ESVs)	Mainstem Mekong						3S Basin					Tonle Sap		
	Border	**Koh Kei**	**Koh Snam Chey**	**Kratie**	**Kampong Cham**	**Phnom Penh**	**Stung Treng**	**Phumi Ksach Nón**	**Kamphoun**	**Upper Sesan**	**Sre Pok**	**Chong Khneas**	**Tonle Sap Lot 4**	**Phat Sanday**
Osteochilus melanopleurus (1)														X
Osteochilus microcephalus (1)					X									
Puntioplites falcifer (4)			X				X		X		X			
Puntioplites proctozystron (1)														X
Puntioplites sp. (1)											X			
Scaphognathops bandanensis VU (3)	X		X				X							
Sikukia gudgeri (11)			X				X				X			
Danionidae														
Raiamas guttatus (3)			X						X					
Rasbora dusonensis (1)			X					X			X			
Rasbora sp. (1)							X							
Xenocyprinidae														
Paralaubuca typus (2)						X			X					
Botiidae														
Yasuhikotakia eos (5)			X					X			X			
Yasuhikotakia lecontei (3)			X								X			
Cobitidae														
Acantopsis dinema (4)							X	X	X					
Nemacheilidae														
Nemacheilus platiceps (1)			X											
Schistura sp. (1)			X											
Siluriformes														
Ailiidae														
Laides longibarbis (1)			X					X						
Bagridae														
Hemibagrus sp. (3)					X			X						
Hemibagrus spilopterus (1)								X						X
Mystus atrifasciatus (1)														X
Mystus sp. (3)						X								X
Clariidae														
Clarias macrocephalus (1)						X								
Pangasiidae														
Pangasianodon gigas[2] CR (5)													X	
Pangasianodon hypophthalmus[2] EN (1)													X	
Pangasius macronema (4)			X		X	X					X			X
Pseudolais pleurotaenia (2)							X	X						

Table 2 Cont'd.

Species (# of ESVs)	Mainstem Mekong						3S Basin					Tonle Sap		
	Border	Koh Kei	Koh Snam Chey	Kratie	Kampong Cham	Phnom Penh	Stung Treng	Phumi Ksach Nón	Kamphoun	Upper Sesan	Sre Pok	Chong Khneas	Tonle Sap Lot 4	Phat Sanday
Siluridae														
Ompok siluroides (1)														X
Beloniformes														
Belonidae														
Xenentodon cancila (2)											X			X
Zenarchopteridae														
Dermogenys siamensis (1)														X
Dermogenys sp. (1)														X
Perciformes														
Ambassidae														
Parambassis sp.[3] (1)							X							
Anabantiformes														
Anabatidae														
Anabas testudineus (1)														X
Channidae														
Channa micropeltes (1)														X
Channa striata (5)							X	X				X		X
Nanidae														
Pristolepis fasciata[4] (4)							X	X		X	X			X
Osphronemidae														
Trichopodus trichopterus (2)														X
Gobiiformes														
Gobiidae														
Gobiopterus sp.[3] (1)											X			
Papuligobius ocellatus (2)		X	X				X							

[1] ESVs were assigned to *A. truncatus*, however this formerly monotypic genus has been split into two species: *A. truncatus* in the Sundaland region and *A. micracanthus* in the northern Indochinese region (Ng & Kottelat, 2004). We therefore presume that the species detected was in fact *A. micracanthus*.

[2] These species were detected at Tonle Sap Lot 4, where they were released prior to eDNA sample collection as part of a mark-recapture study.

[3] Sequences were assigned to species in these genera, but because these species are not known to occur in Cambodia, they are presumed to be incorrect assignments based on poor resolution of the MiFish primers for species within these genera, incomplete genetic reference libraries for the region, or both. As such, the sequence assignments were retained at the genus level, as they were presumed to represent actual diversity at the sites where they were detected.

[4] There is ongoing uncertainty regarding the taxonomic placement of this species. It was assigned to Anabantiformes and Pristolepidae by Kottelat *et al*. (1993), but placed in Nandidae within Perciformes by Nelson (2006). Though the genetic reference library assigned the species to Pristolepidae, we have applied its most latest assignment to Nanidae in Anabantiformes by NNEF (2021).

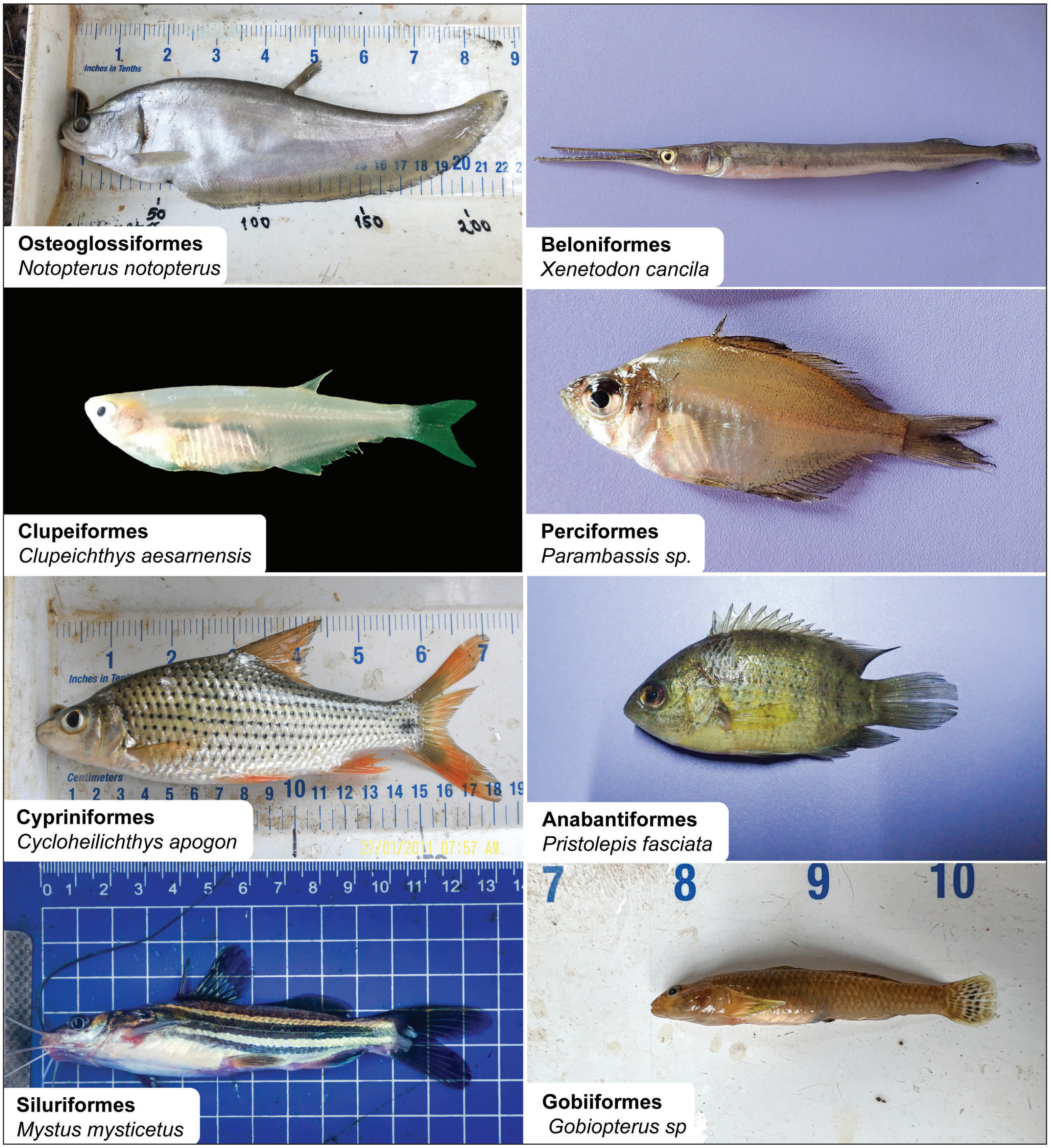

Fig. 2 Fish species among those detected in eight orders in the metabarcoding samples.

software (R Core Team, 2022) for subsequent filtering and analysis.

In total, 56 eDNA samples were collected. Of these, we excluded 11 for PCR failure, three for only having unidentifiable ESVs, and four that only contained mammalian DNA (e.g., *Bos* spp., *Sus scrofa* and *Homo sapiens*). This left 38 samples for analysis. Sequence and sampling data have been uploaded to the NCBI SRA database (BioProject ID: PRJNA1003506). Across these samples, a total of 161 fish ESVs were assigned to 63 fish taxa that represented eight orders, at least 23 families, at least 49 genera, and at least 55 species (Table 2). Two

ESVs could only be assigned to order (both Siluriformes) and ten were only assigned to family (four to Cyprinidae and six to Pangasiidae). A number of ESVs were assigned to genera, including three ESVs each assigned to *Mystus* and *Hemibagrus*, two ESVs each to *Labiobarbus* and *Epalzeorhynchos*, and one ESV each to *Dermogenys*, *Puntioplites*, *Rasbora*, *Sardinops* and *Schistura* (Table 2). Each of these genera assignments were also represented by species level assignments of other ESVs, except for *Epalzeorhynchos*, *Sardinops* and *Schistura*.

We compared all taxa identified in the samples to a list of species known to occur in the Mekong (Jerde *et al*., 2021). Two species from the Western Hemisphere were excluded from our final dataset due to possible laboratory cross-contamination. Another two taxa—*Parambassis ranga* and *Gobiopterus lacustris*—are not known to occur in Cambodia, but other species within these genera do. As *P. ranga* and *G. lacustris* were the sole representatives of these genera in the reference library, this indicates high sequence similarity with Cambodian representatives of these genera, or else the matching algorithm would have only assigned them to genus. We retained these two sequences as genus level taxonomic representatives at the sampled locations. This result, along with other assignments to genus or higher levels, highlights the need for mitochondrial DNA vouchers from all Cambodian fish species.

The detected taxa represented a wide variety of species (Fig. 2), including the Critically Endangered *Catlocarpio siamensis* and *Pangasianodon gigas*, and the Endangered *P. hypophthalmus* (IUCN, 2022). Detection of these iconic species was expected given their release at the project site in the Tonle Sap (Campbell *et al*., 2022), but these results confirm that metabarcoding is able to detect them in natural settings. Sequences belonging to *Cirrhinus microlepis* and *Scaphognathops bandanensis*—both listed as Vulnerable (IUCN, 2022)—were also detected. Further, we detected a sequence belonging to genus *Schistura*, which suggests that cryptic species like loaches may be effectively detected with eDNA, although more work is needed to improve genetic reference libraries for such diverse genera (Jerde *et al*., 2021). The *Schistura* detection occurred only in the sample collected at a depth of 35 m at Koh Snam Chey, suggesting that some species may not be detected from surface sampling alone.

An average of 8.78 fish taxa were detected at each site, and varied from 21 at Phat Sanday to one at Kratie (Fig. 1, Table 1). We used taxa accumulation curves to evaluate whether the number or volume of samples adequately captured taxonomic richness at a regional scale, as well

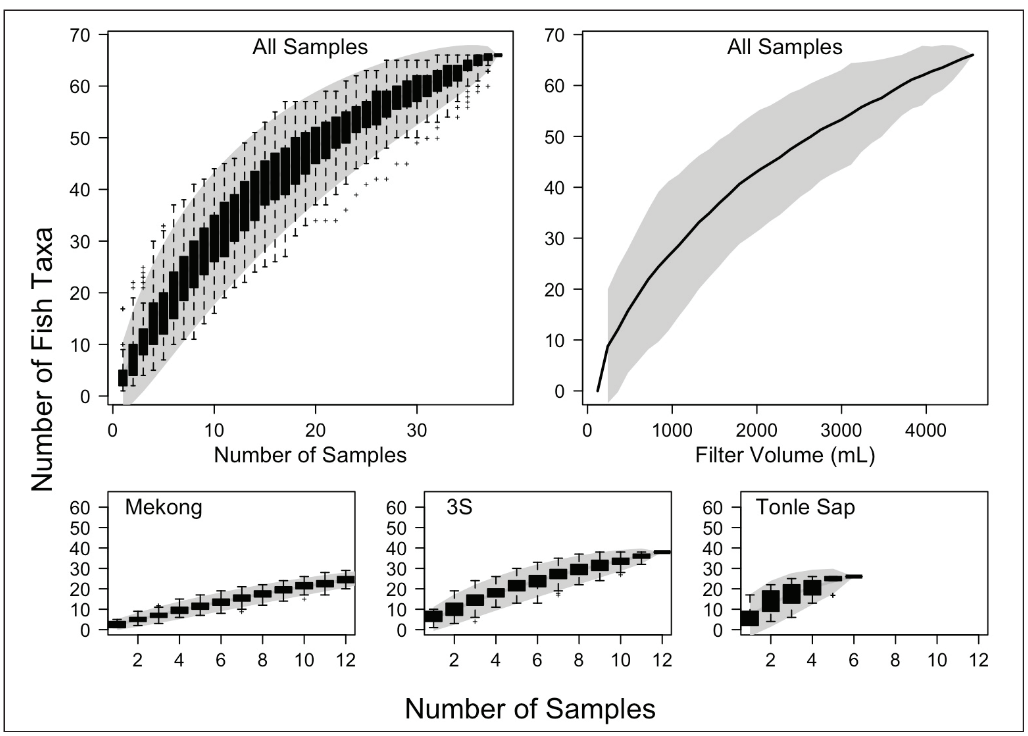

Fig. 3 Fish taxa accumulation curves based on the number of samples from all sites in the Lower Mekong Basin in Cambodia (top left), the Mekong mainstem, 3S basin and Tonle Sap sites. The top right panel is based on total filter volume for all samples.

as basin-wide using all samples. Accumulation curves increase with increasing number of samples and should reach an asymptote, at which point additional samples will not detect more species. The curves were constructed using the '*specaccum*' function available in the *vegan* package for R (Oksanen *et al*., 2022). For the number of samples collected in each region and across the entire basin, the number of species increased but did not reach an asymptote (Fig. 3). The same pattern applied to sample filter volume. The curves indicate that our limited sampling did not provide adequate taxonomic coverage in any region, therefore we cannot evaluate differences in regional levels of diversity.

Our pilot study illustrates some tradeoffs to consider when implementing eDNA metabarcoding studies in the LMB. High turbidity at sample locations precluded the ability to filter large volumes with the sample kits. Larger pore size filters may allow for filtration of greater volumes (e.g., Durand *et al*., 2022), but this carries greater risk of sample contamination with PCR inhibitors (Herder *et al*., 2014), which are abundant in turbid systems (Kumar *et al*., 2021). When filtering smaller volumes (e.g., < 500 ml), finer pore sizes capture significantly more DNA than filters with larger pore sizes (Jeunen *et al*., 2019). Therefore, the best option for the application of single-use kits is to increase the number of samples. Indeed, sample volumes as low as 100 ml collected with kits similar to those we used can effectively detect biodiversity in turbid, tropical systems if suitable replication is achieved (Blackman *et al*., 2021). Clearly, studies seeking to use eDNA to quantify biodiversity will require greater numbers of samples, and consequently greater volumes of water filtered to maximize the number of species detected. In addition, collecting samples from multiple depths may increase detection of benthic species. In the laboratory, increasing the number of PCR replicates may increase detection species with low DNA concentrations. However, this runs the risk of increased chances for cross-contamination. Finally, using eDNA in the most efficient and meaningful way possible in the LMB will require comparisons between the data from traditional sampling and metabarcoding data to better understand the strengths and limitations of a molecular approach. Studies seeking to do so are currently in progress.

Global loss of aquatic biodiversity not only threatens vital fisheries, but also imperils numerous other benefits that freshwater biodiversity provides to humanity (Lynch *et al*., 2023). In the LMB, eDNA is a valuable tool for monitoring biodiversity and informing conservation approaches. Significant benefits provided by eDNA metabarcoding compared to traditional sampling methodologies are its scalability and relatively lower cost and effort. Standardized eDNA sampling may be useful for detecting species diversity in areas that are not represented in current LMB monitoring programs (Halls *et al*. 2013). Pending refinement of best practices in sampling design and buildout of genetic reference libraries, eDNA may also be useful in rapid biodiversity inventory applications such as environmental impact assessments associated with dam development. Each of these potential benefits, when considered in the context of the urgent need for improved fish biodiversity monitoring throughout the LMB, suggest that applications of eDNA studies in the basin may provide important information for fisheries management, conservation efforts, and policy decisions in the region.

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Short Communication

A first description of a breeding colony of white-rumped vultures *Gyps bengalensis* on the Cambodia/Laos border

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In 1985, white-rumped vultures *Gyps bengalensis* were described as "possibly the most abundant large bird of prey in the world", with a global population almost certainly numbering several million individuals at that time (Houston, 1985; BirdLife International, 2021). In South Asia, populations of the species then underwent a catastrophic crash due mostly to veterinary use of diclofenac, a non-steroidal anti-inflammatory drug (NSAID) with lethal consequences for vultures (Oaks *et al*., 2004). The toxicity of diclofenac when ingested by vultures is such that it has reduced the global population of white-rumped vultures by over 99% in the past 30 years (Prakash *et al*., 2007). The global population of the species is currently estimated to be ca. 6,000–9,000 individuals, equating to 4,000–6,000 mature individuals (BirdLife International, 2021, 2023), most of which are in India and Nepal (DNPWC, 2015; Prakash *et al*., 2019). An isolated and disjunct population remains in Cambodia (Fig. 1), centred on the northern plains landscape, though vagrant individuals may cover a larger range (Gilbert *et al*., 2007; Botha *et al*., 2017).

In Cambodia, the status of populations of this Critically Endangered species remain precarious. Following a rapid decline in 2010–2016 from 201 to 109 individuals, which was associated with secondary poisoning (i.e. by carbamates) and a reduction in carrion availability (Clements *et al*., 2013; Loveridge *et al*., 2018), the national population has continued to decline at a slower rate and may now have stabilized around 70 individuals according to a national census (Broadis *et al*., 2021).

Thus far, the Cambodian population has avoided the NSAID poisoning epidemic because the drug has not been widely used in the country. Following discovery of diclofenac use in domestic veterinary practices in 2018, this was banned in Cambodia in July 2019. However, vulture populations in the country are threatened by exposure to pesticides and other chemicals (Loveridge *et al*., 2018), habitat degradation and limited food availability due to the collapse of wild ungulate populations it feeds on (Clements *et al*., 2013), which are listed as critical threats for vultures in the region (Botha *et al*., 2017).

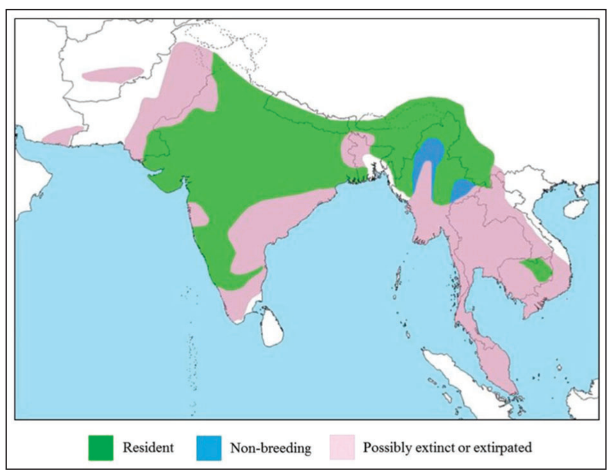

Fig. 1 Global distribution of white-rumped vultures *Gyps bengalensis*, including the range of the isolated population in Cambodia and Laos. Adapted from Botha *et al*. (2017).

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White-rumped vultures (WRVs) are a social species, dozens of individuals can be observed at roost sites and feeding on the same carrion. The species breeds in colonies in trees of varying height (Thakur, 2015; Sehgal & Kumar, 2022) and in northern India, WRVs also seem to favour nesting sites relatively close to feeding stations (Sehgal & Kumar, 2022). According to Naoroji (2006), nests are usually located at a height of 10–18 m throughout their range, and up to 30 m on tall trees, and sometimes as low as 4 to 5 m in protected areas or when larges trees are unavailable.

Because WRVs first breed at an age of 5.1 years and have a generation length (the average age of parents of the current cohort) of 11.2 years (Bird *et al*., 2020), reversing the ongoing population decline in Cambodia will require long-term efforts. Protection of breeding sites is of paramount importance. Well aware of the importance of feeding stations for the conservation management of the species (Gilbert *et al*., 2007), a vulture feeding station has been in existence at Siem Pang Wildlife Sanctuary (SPWS), since July 2004 (Birdlife International *et al*., 2005). Since August 2020, one cow or buffalo has been provided on a weekly basis at the station to ensure a reliable and regular supply of food for vultures.

Three species of vultures regularly visit the feeding station and nests of slender-billed vultures *G. tenuirostris* and red-headed vultures *Sarcogyps calvus* are recorded annually in SPWS. In 2022 for example, nine slender-billed vulture nests and two red-headed vulture nests were found in the wildlife sanctuary (Rising Phoenix, 2022). With only one nest found in 2015 and two in 2016, white-rumped vultures rarely nest in SPWS yet are the most commonly recorded vulture species at the SPWS feeding station, with an average monthly count of 48 birds in 2022 (Rising Phoenix, 2022). In addition, no WRV nests were recorded in Cambodia in 2020 and 2021 and only one (which failed) was recorded in 2022, a surprising and concerning outcome considering it is by far the most numerous vulture species in the country (Broadis *et al*., 2021). This situation contrasts markedly with records of up to 32 nests in 2005 (of which 22 were in Mondulkiri Province and 9 in Lomphat Wildlife Sanctuary) and 28 nests in 2006 (Goes, 2013). Neither of these areas have active WRV nests at present and only foraging individuals have been observed, in line with notion that individuals may cover large areas in search of food.

To resolve the mystery of their breeding locality and preferred nesting habitats, we captured an adult and an immature WRV at SPWS between 4 and 7 March 2020. Both birds were fitted with GPS trackers (OrniTrack-50 - solar powered GPS-GSM tracker) from Ornitela (Vilnius, Lithuania). These devices weigh 50 g, representing around 1.3% of the bird's bodyweight, which is less than the 3% usually deemed acceptable for tracking (Kenward, 2001; Barron *et al*., 2010). The GPS acquisition interval was set to every hour and GSM transmission attempt was set to every two hours during the day. The tracker quickly revealed that both individuals were returning to a single location on the Phou Kiou ridge on the southern boundary of Xe Pian National Park in Laos, just 2 km from the western boundary of SPWS (Fig. 2).

Located in Champasak Province of Laos, the Phou Kiou hills (14°05'08.0"N, 106°07'25.7"E) encompass a forested ridge that rises steeply from the surrounding plains (which are 90 m above mean sea level, AMSL) to an altitude of 220 to 290 m AMSL. The ridge extends approximately 30 km on a northwest to southeast axis, with slopes being steeper on the southern side. The hills are covered with dense semi-evergreen forest that is relatively untouched, although we observed selective logging of timber trees on the ridgetop.

During a first partial survey of the site on 14 April 2020, two nests and three chicks (including one coming from a third nest) were found. On 12 April 2022, a second survey successfully located six nests, including four with chicks and two nests which were not in use.

On returning to the site on 27 April 2023, we recorded 15 nests along a 4 km transect on the ridge line (Fig. 2). Twelve of these were active and nine chicks were counted in the nests. Two nests per tree were observed on six occasions (Fig. 3). Though we explored the ridge, it is possible that we may have missed some nests and likely that undiscovered nests exist to the west of where we searched on the ridge.

The Phou Kiou white-rumped vulture colony is a new and important discovery. To the best of our knowledge, it is the first report of a breeding site for WRV in Laos in peer-reviewed literature (Timmins & Vongkhamheng, 1996; Thewlis *et al*., 1998; Evans *et al*., 2000; Duckworth & Tizard, 2003; Pain *et al*., 2003; Duckworth & Timmins, 2015). As such, Laos should be added to the list of breeding range states for WRV in the next edition of the multi-species action plan to conserve African-Eurasian vultures.

Birds that breed on Phou Kiou ridge feed at the feeding station in SPWS which is located 25 km to the east. It is not known whether the establishment of this colony predates carcass provisioning at SPWS. The nests were found in *Anisoptera costata* with a height of at least 20 m, which is within the known range for WRV (Naoroji, 2006). Tall trees with strong forks to support nests have been identified as preferred nesting sites for the species elsewhere (Gilbert *et al*., 2002; Baral *et al*., 2005; Thakur &

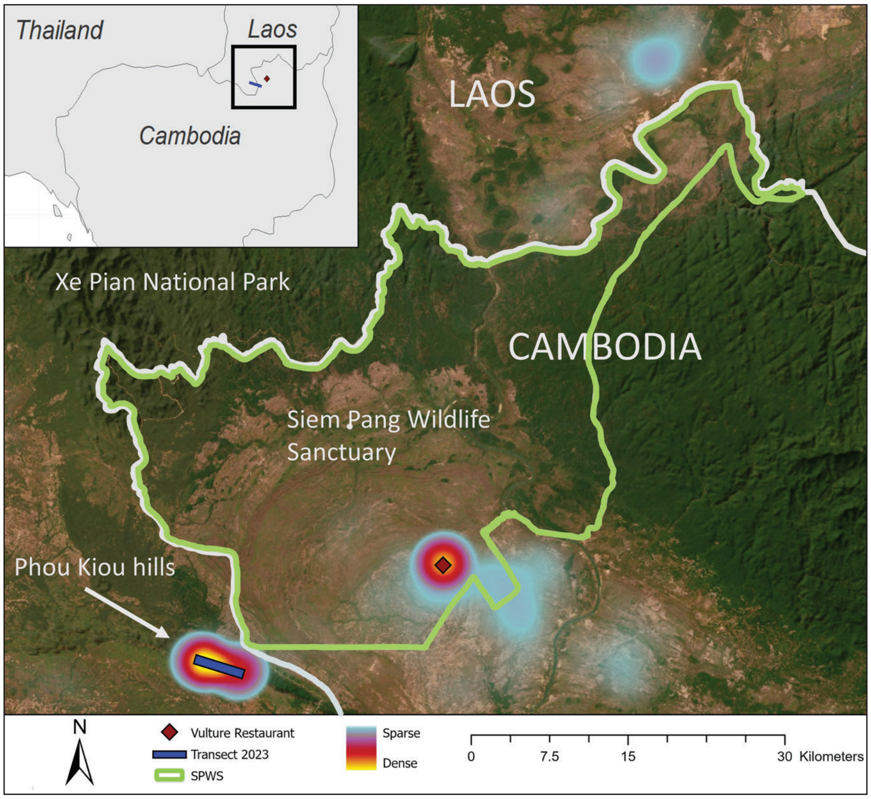

Fig. 2 Heatmap of recorded locations of one white-rumped vulture tagged in 2020. Two hotspots were observed, one centred on the vulture restaurant in Siem Pang Wildlife Sanctuary and one in the Phou Kiou hills in Champasak Province, Laos.

Fig. 3 Nests (yellow circles) of white-rumped vultures photographed in April 2023. One young is resting in the lower nest while the other is perching in the far left of the image.

Narang, 2012; Sehgal & Kumar, 2022). A study by Murn *et al*. (2015) indicates that the spatial pattern of nests relies on both the distribution of trees and their ability to support more than one nest. These results highlight that the preservation of larger nest trees and the sustainable management of timber resources are essential components for conservation management (Botha *et al*., 2017).

We recommend that annual surveys be undertaken of the colony and that use of drones (Junda *et al*., 2015; Santangeli *et al*., 2020; Zink *et al*., 2023) or examination of high-resolution satellite images (Hughes *et al*., 2011; Fretwell *et al*., 2017) be considered. We also recommend satellite tagging of additional WRVs to determine the location of additional nests or colonies. To this end, we captured and tagged two further adult WRV on 24–25 April 2023 and also recaptured an immature WRV tagged in 2020, which allowed us to ensure that device was still functioning and fitting properly.

During our short surveys, we observed logging of timber trees in semi-evergreen forests in the immediate vicinity of the colony on top of the ridge, and the widespread destruction of deciduous dipterocarp forests at the foot of the ridge. We also observed people clearing and burning forest to plant cassava within Xe Pian National Park.

Although diclofenac has been banned in Cambodia for veterinary use since 2019, it is unknown whether it is used in Laos. The same is true of the presence and use of other vulture-toxic drugs such as ketoprofen, flunixin and aceclofenac in both countries. Potential risks posed by these drugs to vultures should be assessed, along with other possible threats such as electrocution from power lines or reduced food availability.

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Short Communication

First specimen-based record of blackskin catfish *Clarias meladerma* Bleeker, 1846 (Siluriformes: Clariidae) from the Mekong River basin

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Members of the Old World catfish family Clariidae naturally occur in freshwater bodies throughout Africa and Asia (Ng, 1999; Teugels *et al*., 2001; Ferraris, 2007; Ng & Kottelat, 2008, 2014; Ng *et al*., 2011; Ng & Hadiaty, 2011; Nelson *et al*., 2016). Clariid catfishes have a slender body with long-based dorsal and anal fins without an adipose fin, except for *Heterobranchus*, *Dinotopterus* and *Bathyclarias* species and *Clarias ngamensis* (with a short adipose fin), and they possess subtruncate and truncate, rounded caudal fins and four pairs of barbels (Rainboth, 1996; Anseaume & Teugels, 1999; Ng, 1999, 2004; Skelton, 2001; Nelson *et al*., 2016). They are commonly known as "walking catfish" or "air-breathing catfish" (Taki *et al*., 2021) and have air-breathing organs that allow them to tolerate low-oxygen level environments and survive out of water for considerable time (Ng & Kottelat, 2014; Taki *et al*., 2021).

The Clariidae is a diverse family containing at least 113 species in 16 genera (Ferraris, 2007; Ng & Hadiaty, 2011; Ng & Kottelat, 2008, 2014). Within the family, the genus *Clarias* comprises an estimated 56 species, including 20 species in Southeast Asia (Ng *et al*., 2011; Kottelat, 2013; Ng & Kottelat, 2014) and is classified into two species complexes, the *C. nieuhofii* complex with an elongated body, and the *C. batrachus* complex with a rather shortened body (Ng *et al*., 2011; Ng & Kottelat, 2014). Although eight species of *Clarias* have been reported from the Mekong River basin (Rainboth, 1996; Taki *et al*., 2021), only four have been confirmed with voucher specimens (i.e., *C. batrachus*, *C. macrocephalus*, *C. serniosus* and introduced *C. gariepinus*) (Ng & Kottelat, 2014; So *et al*., 2018; Taki *et al*., 2021). Blackskin catfish *C. meladerma* Bleeker, 1846 has been recorded from short drainages on the western face of the Cardamom Mountains in the coastal areas of the Koh Kong and Preah Sihanouk provinces in

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Cambodia, but not from the Cambodian Mekong River basin (So *et al*., 2018; Taki *et al*., 2021).

On 26 March 2021, seven individuals of *Clarias* (four medium-sized and three juvenile specimens) morphologically resembling *C. meladerma* were collected from a small forest stream in the Cambodian Mekong basin in Sesan District, Stung Treng Province (Fig. 1). This site is situated in the Ochralang watershed which drains into the Mekong River at Tboung Khla Village, Omresh Commune, Siem Bok District, Stung Treng Province. Following careful examination of the seven specimens, we identify these as *C. meladerma* and report these as the first record from the Mekong River basin in Cambodia.

Clarias meladerma Bleeker, 1846

Material examined: Four specimens from Sesan District (13.361021°N, 106.219737°E) were preserved in 95% ethanol and deposited in the fish collection room of the Faculty of Fisheries and Aquaculture, Royal University of Agriculture (RUAFI), Cambodia. These were RUAFI-F00001 (175 mm SL [standard length]), RUAFI-F00002 (170 mm SL), RUAFI-F00003 (136 mm SL) and RUAFI-F00004 (135 mm SL). Comparative materials for *C. meladerma* comprised four specimens from south and southwest Cambodia deposited in the specimen room of the Inland Fisheries Research & Development Institute (IFReDI), Fisheries Administration, Cambodia, as follows: IFReDI-P06012 (235 mm SL), collected 21 November 2011, 10.645278°N, 103.643889°E, Kbal Chhay Canal, Srai Cham, Prey Nob District, Preah Sihanouk Province; IFReDI-P06478 (208 mm SL) & IFReDI-P06486 (190 mm SL), collected 5 February 2012, 10.630556°N, 103.630806°E, Peanichkam Market, Preah Sihanouk Town, Preah Sihanouk Province; IFReDI-P06550 (205 mm SL), collected 1 June 2012, 11.416239°N, 103.267175°E, Chhay Preik Brong Waterfall, Trapeang Roung Commune, Koh Kong District, Koh Kong Province. Finally, two specimens of *C. macrocephalus* caught in arrow-shaped traps and deposited in RUAFI were also examined for comparative purposes: RUAFI-F00120 (20.5 mm SL), collected 29 October 2022 and RUAFI-F00400 (239 mm SL), collected 18 November 2022, 13.233880°N, 103.862000°E, flooded forest, Chong Khneas Commune, Tonle Sap Lake, Siem Reap Province. Meristic and morphometric data were taken following Ng (1999).

Diagnosis: *Clarias meladerma* is characterised by a short and extremely rounded occipital process, a shorter distance between the occipital process and the first dorsal fin (3.67–4.51 % SL), a long and distinctly serrated (14–17) anterior edge of the pectoral spine and a smooth posterior edge of the pectoral spine (Table 1). The specimens preserved in 95% ethanol are dark brown and do not have small blotches on the sides of the body, head or fins. The number of dorsal fin-rays range from 66 to 68 whereas the number of anal fin-rays range from 55 to 56 (Fig. 2 & 3).

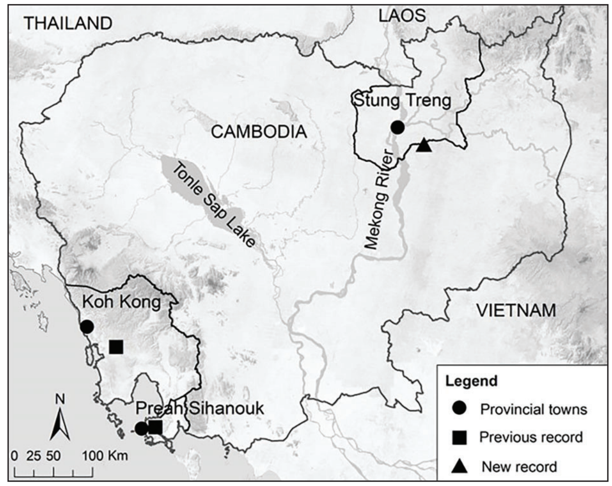

Fig. 1 Records of the blackskin catfish *Clarias meladerma* Bleeker, 1846 in Cambodia. Black squares are the past records in the Koh Kong and Preah Sihanouk provinces of So *et al*. (2018) and Taki *et al*. (2021), whereas the black triangle represents the first specimen-based record from the Mekong drainage in Sesan District, Stung Treng Province.

Description: Data on morphometric characters and meristic counts of the Mekong and Cardamom specimens is presented in Table 1. Additional characters include: head depressed, covered with hard bony plates that are firmly sutured. Eyes small, ovoid. Four pairs of barbels with thick fleshy bases that gradually taper toward tip. Frontal fontanelle is short and squat. Occipital fontanelle is small and oval-shaped. Occipital process is short and broadly rounded. Anterior margin of pectoral-fin spine with 14–18 strong serrae. Body dark brown, without dark blotches.

Habitat: Our four specimens of *C. meladerma* were collected from a small slowly-flowing stream in swampy forest. Juveniles of *C. meladerma* were also observed in a stream with slowly-flowing water and aquatic vegetation. The watershed drains into the Mekong River at Tboung Khla Village, Omresh Commune, Siem Bok District, Stung Treng Province. The species has also been reported from coastal streams draining the Cardamom Mountains in the Preah Sihanouk and Koh Kong provinces (So *et al*., 2018; Taki *et al*., 2021).

Remarks: Meristic and morphometric characters for specimens from the Mekong River basin agree closely with those of specimens from the Cardamom Moun-

Table 1 Morphometric and meristic data for specimens of *Clarias meladerma* Bleeker, 1846 from Cambodia.

Morphometric characters	Mekong River basin (*n*=4)		Cardamom Mountains (*n*=4)	
	Range	Mean ± SD	Range	Mean ± SD
Total length (mm)	157.0–202.0	178.50 ± 23.30	222.0–270.0	241.50 ± 20.29
Standard length (mm)	135.0–175.0	154.00 ± 21.46	190.0–235.0	209.50 ± 18.73
As % of standard length				
Predorsal length	26.83–28.68	27.56 ± 0.80	26.73–28.40	27.81 ± 0.79
Preanal length	47.75–50.70	48.68 ± 1.36	46.36–50.73	47.78 ± 2.02
Prepelvic length	40.86–43.98	42.46 ± 1.28	39.08–42.99	40.95 ± 2.07
Prepectoral length	18.49–21.02	19.92 ± 1.09	18.40–19.71	19.04 ± 0.58
Length of dorsal fin base	70.05–74.13	72.09 ± 1.68	71.15–75.12	73.08 ± 1.71
Anal fin base length	50.46–52.54	51.23 ± 0.92	51.05–55.29	52.48 ± 1.96
Pelvic fin length	7.10–8.63	8.01 ± 0.65	7.65–9.05	8.40 ± 0.63
Pectoral fin length	12.74–14.75	13.51 ± 0.89	13.69–14.72	14.24 ± 0.45
Pectoral spine length	8.88–11.15	10.00 ± 1.00	9.15–13.33	11.45 ± 2.12
Caudal fin length	15.01–16.24	15.86 ± 0.57	13.36–15.24	14.34 ± 0.90
Distance between occiptial process & dorsal fin	3.67–4.51	4.17 ± 0.41	2.90–3.75	3.36 ± 0.35
Caudal peduncle depth	4.79–6.16	5.61 ± 0.63	5.32–6.14	5.78 ± 0.37
Body depth at anus	16.51–18.96	17.98 ± 1.06	16.19–18.51	17.46 ± 0.96
Head length	23.12–25.27	24.24 ± 0.91	23.49–24.61	24.24 ± 0.52
Head width	17.86–18.35	18.16 ± 0.21	17.56–19.83	18.7 ± 0.94
Head depth	13.26–15.01	14.37 ± 0.77	10.90–12.18	11.66 ± 0.62
As % of head length				
Snout length	26.40–28.58	27.84 ± 0.98	28.16–31.41	29.77 ± 1.34
Interorbital distance	44.15–46.09	45.18 ± 1.05	45.10–47.09	46.31 ± 0.86
Eye diameter	6.20–8.05	7.09 ± 0.83	5.30–6.06	5.59 ± 0.34
Nasal barbel length	83.87–100.78	92.14 ± 6.95	86.09–114.72	99.6 ± 12.42
Maxillary barbel length	127.41–155.70	144.57 ± 13.27	140.33–172.34	158.48 ± 14.89
Inner mandibular barbel length	105.36–117.33	110.92 ± 6.29	74.50–130.81	112.13 ± 26.34
Outer mandibular barbel length	77.71–93.54	86.56 ± 7.53	77.96–95.74	87.46 ± 8.36
Front fontanel length	14.20–23.28	18.92 ± 3.94	12.18–19.56	15.42 ± 3.11
Front fontanel width	7.91–11.13	9.62 ± 1.61	8.20–11.54	9.21 ± 1.57
Occipital fontanelle length	10.52–15.24	12.34 ± 2.14	8.89–12.40	10.63 ± 1.48
Occipital fontanelle width	6.63–14.03	9.47 ± 3.19	5.47–6.73	6.35 ± 0.59
Occipital process length	12.18–14.03	12.87 ± 0.82	10.19–14.67	13.19 ± 2.04
Occipital process width	50.16–58.25	54.21 ± 3.41	50.39–52.01	51.44 ± 0.72
Meristic counts				
Dorsal fin ray number	66–68	66.75 ± 0.96	65–68	66.50 ± 1.29
Pectoral fin ray number	7–8	7.50 ± 0.58	8–9	8.75 ± 0.50
Pelvic fin ray number	6–6	6 ± 0.00	6–6	6.00 ± 0.00
Anal fin ray number	55–56	55.25 ± 0.50	55–60	57.25 ± 2.22
Serrae number	14–17	15.75 ± 1.26	18–18	18.00 ± 0.00

Note: Morphometric data as well as total and standard lengths were measured to the nearest 0.01 mm.

Fig. 2 Dorsal views of *Clarias meladerma* Bleeker, 1846 from Stung Treng Province (RUAFI-F00002, upper panel) and the Cardamom Mountains (IFReDI-P06486, middle panel) and *C. macrocephalus* Günther, 1864 from Siem Reap Province (RUAFI-F00120, lower panel), Cambodia.

Fig. 3 Lateral views of *Clarias meladerma* Bleeker, 1846 from Stung Treng Province (RUAFI-F00002, upper panel) and the Cardamom Mountains (IFReDI-P06486, middle panel) and *C. macrocephalus* Günther, 1864 from Siem Reap Province (RUAFI-F00120, lower panel), Cambodia.

Fig. 4 Lateral view of *Clarias meladerma* Bleeker, 1846 (IFReDI-P06012) from the Cardamom Mountains, Cambodia.

tains (Fig. 3 & 4) and Sumatra (Ng & Kottelat, 2014: Fig. 3), except for the absence of dark blotches on body and head. Despite this difference in colouration, all other characters support our identification of the Mekong specimens as *C. meladerma*. Rainboth (1996) reported *C. meladerma* from the Mekong delta in Vietnam, but this was not a specimen-based record. It might be possible that our specimens represent *C. macrocephalus*, which is a morphologically similar species known from the Mekong River drainage that has a rounded occipital process like *C. meladerma*. However, this is precluded by the presence of a distinctly serrated (vs. smooth in *C. macrocephalus*) anterior edge of the pectoral spine in our specimens (Fig. 5).

Clarias meladerma was described from Indonesia and has been reported widely from mainland Southeast Asia, the Philippines and Phu Quoc Island (Rainboth, 1996; Ferraris, 2007; Vidthayanon, 2008; Vasil *et al*., 2013; Ng & Kottelat, 2014). In previous studies, *C. meladerma* was recorded from the Mekong delta in Vietnam (Rainboth, 1996; Vidthayanon, 2008; Rainboth *et al*., 2012) and from the Mekong River basin in Laos (Kottelat, 2000, 2001), but later studies based on specimen examination confirmed that *C. meladerma* was not found in the Mekong delta (Ng & Kottelat, 2014) and concluded that specimens were required to confirm its occurrence in the Mekong basin in Laos, Thailand, Cambodia and Vietnam (Taki *et al*., 2021). According to Kottelat (2000, 2001), *C. meladerma* from Laos was based on a photographic record from the Mekong River basin in Boeng Kan Province, Thailand.

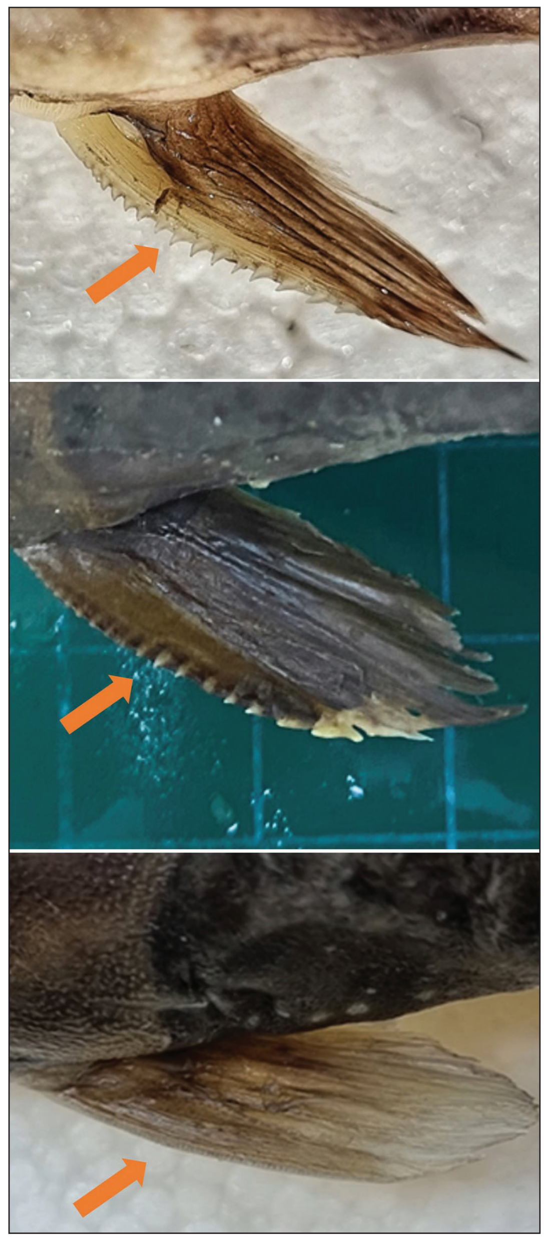

Fig. 5 Left pectoral spines of *Clarias meladerma* Bleeker, 1846 from Stung Treng Province (RUAFI-F00002, upper panel) and the Cardamom Mountains (IFReDI-P06486, middle panel) and *C. macrocephalus* Günther, 1864 from Siem Reap Province (RUAFI-F00120, lower panel), Cambodia.

Ng & Kottelat (2014) examined one photograph of an individual from Boeng Khong Long wetland in Boeng Kan Province and tentatively treated this as *C. meladerma* based on the serrated anterior edge of the pectoral spine, a caudal peduncle depth of about 5% and presence of black blotches on the body. In Cambodia, *C. meladerma* has been collected from the Cardamom Mountains outside of the Mekong basin (So *et al.*, 2018; Taki *et al*., 2021). So *et al*. (2018) reported the occurrence of *C. meladerma* from the Tonle Sap Lake (within the Mekong basin), but did not retain voucher specimens or photographs from the survey. As such, the present study constitutes the first specimen-based confirmation of *C. meladerma* from the Cambodian Mekong drainage. Past records of *C. meladerma* from the drainage may have been misidentifications of *C. macrocephalus* due to the outward similarity of both species. As a result, future studies should incorporate morphological and molecular analyses to confirm the identification of potential specimens of *C. meladerma* from the Mekong River basin.

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Demographics and practices of dog ownership in a rural Cambodian village adjacent to a wildlife sanctuary

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មូលន័យសង្ខេប

សត្វឆ្កែស្រុកមានវត្តមាននៅទូទាំងពិភពលោក ហើយពួកវាបង្កការគំរាមកំហែងដល់សត្វព្រៃក៏ដូចជាសុខភាពមនុស្ស។ ការគ្រប់គ្រងសត្វឆ្កែប្រកបដោយប្រសិទ្ធិភាពមានការប្រឈមជាច្រើននៅក្នុងប្រព័ន្ធអេកូឡូស៊ីសង្គម។ យើងបានធ្វើប្រជាសាស្ត្រសត្វឆ្កែស្រុក និងទំលាប់នៃការចិញ្ចឹមនៅភូមិជនបទមួយដែលជាប់នឹងដែនជម្រកសត្វព្រៃសៀមប៉ាង។ ការសិក្សានេះមានសារៈប្រយោជន៍សម្រាប់ការរៀបចំយុទ្ធសាស្ត្រគ្រប់គ្រងដើម្បីកាត់បន្ថយហានីភ័យចំពោះសត្វព្រៃដែលបង្កឡើងដោយសត្វឆ្កែស្រុក និងការរៀបចំផែនការដ៏មានសក្តានុពលសម្រាប់បង្ការជម្ងឺឆ្លងក្រៅនិងក្នុងស្រុក។ យើងបានចុះសំភាសន៍ប្រជាជននៅភូមិគោលដៅដោយប្រើកម្រងសំណួរគ្រួសារ (*n*=123) ដើម្បីវាយតម្លៃប្រជាសាស្ត្រសត្វឆ្កែ ទំលាប់នៃការចិញ្ចឹមសត្វឆ្កែ ទំនាក់ទំនងរវាងសត្វឆ្កែជាមួយសត្វព្រៃ និងអាកប្បកិរិយាចំពោះការគ្រប់គ្រងសត្វឆ្កែ។ យើងបានរកឃើញអត្រាសត្វឆ្កែចិញ្ចឹមជាមធ្យមចំនួន 2.41 ក្បាលក្នុងមួយគ្រួសារ ហើយសត្វឆ្កែទាំងអស់ត្រូវបានប្រលែងអោយដើរដោយសេរី។ ម្ចាស់ឆ្កែមួយចំនួនតូច (18%) ដឹងថាឆ្កែរបស់ពួកគាត់ឧស្សាហ៍ចេញទៅក្រៅ ខណៈ 40% នៃម្ចាស់ឆ្កែពេលខ្លះបណ្តើរឆ្កែចូលព្រៃជាមួយពួកគាត់។ មានតែ 10% នៃអ្នកឆ្លើយសំណួរបាននិយាយថាប្រទះឃើញសត្វឆ្កែប្រមាញ់ឬយាយីសត្វព្រៃដោយមិនបានដឹងថាជាសត្វឆ្កែរបស់អ្នកណាទេ។ ការយល់ស្របឬបដិសេធទាំងស្រុងទៅលើការគ្រប់គ្រងសត្វឆ្កែនៅមានកម្រិតទាប ដោយប្រជាជនភាគច្រើនបង្ហាញពីភាពនៅកណ្តាល (35–70%) ចំពោះទស្សនៈនៃការគ្រប់គ្រងសត្វឆ្កែចំនួន៦ក្នុងចំណោម៧ទស្សនៈ។ លទ្ធផលនៃការសិក្សារបស់យើងបង្ហាញថា ដើម្បីកាត់បន្ថយផលប៉ះពាល់នៃសត្វឆ្កែមកលើសត្វព្រៃ វិធីសាស្ត្រដ៏មានប្រសិទ្ធភាពក្នុងការគ្រប់គ្រងសត្វឆ្កែគួរតែគ្របដណ្តប់នៅក្នុងសហាគមន៍ទាំងមូល ដោយផ្តោតយ៉ាងសំខាន់ទៅលើសុខភាព អាហារូបត្ថម្ភ និងការចូលមើលជម្រកសត្វព្រៃ។

Abstract

Domestic dogs are abundant worldwide and they pose a threat to wildlife as well as human health. Effective management of dogs is challenged by complex socio-ecological factors. We investigated local dog demographics and cultural practices of dog ownership in a rural Cambodian village adjacent to Siem Pang Wildlife Sanctuary. This knowledge will be useful for preparing a management strategy to mitigate risks posed by dogs to wildlife and potentially contingency planning for exotic and endemic diseases. Household questionnaires (*n*=123) were completed to assess dog demographics, ownership practices, dog-wildlife interactions and attitudes towards dog management. We found a mean ownership rate of 2.41 dogs per household, with all dogs allowed to roam freely. Relatively few owners (18%) were

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aware of their dogs roaming away from their household, while 40% were accompanied by their dogs into the forest at least some of the time. Only 10% of respondents admitted to observing dogs hunting or harassing wildlife without specifying whether they were their own dogs. Strong agreement or disagreement with management statements was low, with respondents commonly indicating that they were neutral (35–70%) on six of seven management statements. Our findings show that a management strategy for dogs will need to target the whole community, and indicate key areas such as health, nutrition and sanctuary visitation, that may be important for reducing the impact of dogs on wildlife.

Keywords Free-roaming dogs, management, protected areas, questionnaires, wildlife conservation.

Introduction

Domestic dogs *Canis familiaris* are abundant worldwide and their management ranges from being owned, fed, sheltered and contained to being feral, where the animal receives no care from humans and they must find their own food (World Health Organization, 1988). Free roaming dogs sit in the middle of this management spectrum, with such animals receiving some care from humans but are largely left to roam freely outside of their homes (Meek, 1999; Hughes & Macdonald, 2013). Such free-roaming dogs are ubiquitous in Southeast Asia and the degree of care they may receive varies depending on location, community and individual ownership.

Domestic dogs can have various impacts on wildlife, from killing (e.g., Kruuk & Snell, 1981; Taborsky, 1988) and harassment (e.g., Banks & Bryant, 2007; Glover *et al*., 2011; Weston & Stankowich, 2015) to competition (e.g., Butler & Du Toit, 2002; Vanak *et al*., 2015) and disease transmission (e.g., Scott, 1988; Butler *et al*., 2004). Dogs have been implicated as having negative impacts, such as predation, disturbance and disease transmission, on a large number of threatened species which can contribute to population declines (Banks *et al*., 2003; Lessa *et al*., 2016; Gatti *et al*., 2018; Augusteyn *et al*., 2021), with the highest number of species known to be impacted occurring in Southeast Asia (Doherty *et al*., 2017). The relative significance of dog impacts on threatened species and how they compare to the other threatening processes impacting these species remains unclear, particularly regarding non-consumptive effects (Hughes & Macdonald, 2013; Doherty *et al*., 2017).

Free-roaming dogs are thought to account for the majority of dogs world-wide (Hughes & Macdonald, 2013) and it is likely that they also hunt and scavenge for food to supplement what is provided to them, particularly in more rural areas where wildlife is typically more abundant. Dogs may target a variety of wildlife species, including large species such as deer, as documented in the United States of America and India (e.g., *Cervus elaphus*, *Odocoileus hemionus*, *Rusa unicolor*) (Bergman *et al*., 2009; Home *et al*., 2018). In addition to killing, the presence of dogs in natural habitats as they hunt and scavenge for food may induce fear-mediated behavioural changes in wildlife, and harassment by dogs may impact survival and fecundity, as found in Ecuador (Zapata-Rios & Branch, 2016).

Dogs are also vectors for diseases affecting wildlife, humans and livestock, with over 40 zoonotic diseases known to be carried and transmitted by dogs (Bergman *et al*., 2009). Disease transmitted by domestic dogs to wildlife have had serious consequences for some species, such as the decimation of black-footed ferret *Mustela nigripes* populations in the USA due to canine distemper virus (Williams *et al*., 1988) and the loss of African wild dog *Lycaon pictus* packs through rabies and canine distemper viruses (Kat *et al*., 1995; Alexander *et al*., 1996). Some of these diseases can also have significant impacts on human health, such as the rabies virus and hydatidosis.

Hence, there is a need to mitigate negative impacts of free-roaming dogs on wildlife, as well as improving human and animal health. Dog populations that have some degree of dependency on humans present particular management challenges that require effective engagement with the local people (Kennedy *et al*., 2018). Before implementing interventions to control dogs, it is necessary to understand the local situation, including local people's perceptions and attitudes, their interactions with the dog population as well as its size and condition, so that the management strategy is tailored to local conditions. The role of people's attitudes and practices towards domestic dogs and how dog interactions with humans are perceived is an important consideration, with concepts such as 'food provision', 'dog walking' and even 'ownership' being variable across cultures (Miller *et al*., 2014). Management decisions need to be developed based on evidence, not assumptions, about community views and practices as this potentially results in ineffective management and may result in conflict (Miller, 2009).

Cambodia supports significant populations of globally threatened vertebrates, including primates (Rawson *et al*. 2009; Phan & Gray, 2010a), large ungulates (Maxwell *et al*., 2007; Phan & Gray, 2010b; O'Kelly & Nut, 2010; Gray *et al*., 2011), carnivores (Gray *et al*., 2010, 2012) and large waterbirds (Seng *et al*., 2003; Wright *et al*., 2012). Dogs have been identified as a threat to species such as dhole *Canis alpinus* (Kamler *et al*., 2015) and Eld's deer *Rucervus eldii* (Gray *et al*., 2015). Domestic dogs are capable of killing juvenile Eld's deer, and likely adult females if the dogs are hunting in packs (Gray *et al*., 2015). The impact of free-roaming dogs on wildlife in Cambodia is largely unknown, with few studies undertaken in the country and these primarily about hunting with people. Coad *et al*. (2019) found that domestic dogs are commonly used to hunt in Cambodia, with just over half of households that reported hunting in the Cardamom Mountains using dogs. This figure was even higher in eastern Cambodia, with 87% of hunters in Keo Seima Wildlife Sanctuary using dogs (Ibbett *et al*., 2020).

In addition to the impact that free-roaming dogs may have on biodiversity in Cambodia, the impact on human health is also a significant concern. Rabies is endemic in Cambodia, with dogs recognised as the main reservoir for the virus in the country (Ly *et al*., 2009). Dog bite incidences are an important proxy for rabies incidence estimation and a survey of four villages in the Cambodian province of Siem Reap recorded a high dog attack incidence rate of five attacks per 100 person-years, with most attacks involving a household dog (Ponsich *et al*., 2016). Parasitic infections are also a concern, with high proportions of Cambodian village dogs found to be infected with hookworm and Echinostomes (Inpankaew *et al*., 2015). In addition, eight helminths and three protozoan parasite species were recorded in a separate village (Schär *et al*., 2014) and taken together with reports of hydatid disease (Garjito *et al*., 2019), dogs pose serious health risks to villagers.

Siem Pang Wildlife Sanctuary (SPWS) in northeast Cambodia is home to a number of threatened species for which dogs have been identified as threats, including Eld's deer (Gray *et al*., 2015). The sanctuary is zoned into a multiple use zone, a conservation zone with restricted access and a strictly protected core zone. Dogs are prohibited across the entire sanctuary, including these areas. However, in practice villagers and dogs generally have open access across the sanctuary. This is due to a lack of regard people have for the rules and a lack of enforcement due to insufficient rangers and resources. There is currently no management strategy relating to domestic dogs in the area. Developing management plans for dogs owned by humans is complex and cannot be undertaken without communication and consultation with the owners and the provisioning of general data on ownership (Murray & Penridge, 1997). As such, we aimed to collect baseline information on the demographics of the dog population, roaming behaviours, dog-wildlife interactions, how dogs are cared for and the attitudes of local people regarding the domestic dog population around SPWS.

Methods

Study site

Siem Pang Wildlife Sanctuary is located in Stung Treng Province, Cambodia, next to the Laos border (Fig. 1). The sanctuary is home to a number of Endangered and Critically Endangered species including Eld's deer and giant ibis *Thaumatibis gigantea*, and is actively managed by Rising Phoenix Co. Ltd., a social enterprise that has absorbed the previous BirdLife International project at the site. Seven villages are located close to or within the wildlife sanctuary and local people from these villages routinely enter the sanctuary to collect non-timber forest products, fish and hunt. These villages are actively engaged with a livelihoods programme designed to benefit local people and encourage sustainable stewardship of the forest (BICP, 2020). We conducted a survey of Khmer families in the village of Khes Svay due to its proximity to the wildlife sanctuary boundary. This village is considered to be largely representative of villages in this area because of its size, industry and use of the forest in the sanctuary, with villagers and dogs regularly observed in the sanctuary. Agriculture is an important economic activity for the villagers and many households have some reliance on non-timber forest products. The village has 233 households, as recorded in 2020 (Siem Pang District Administration Office, unpubl. data).

Ethical considerations

This study received University of Queensland Institutional Human Ethics Research Approval (2019002414). Before speaking to any families, a meeting with the village chief was organised to explain the aims and methods of the research and seek permission to conduct the research. A copy of the survey questions and an explanation of the research, including ethical approval, was provided to him, as well as the commune chief. Prior to any interviews with families, the Khmer interviewers explained the purpose of the survey, risks, benefits and proposed use of the data and verbal consent was sought. All participation was on a voluntary basis, anonymous and no compensation was provided and the survey was undertaken in Khmer by employees of the BirdLife Inter-

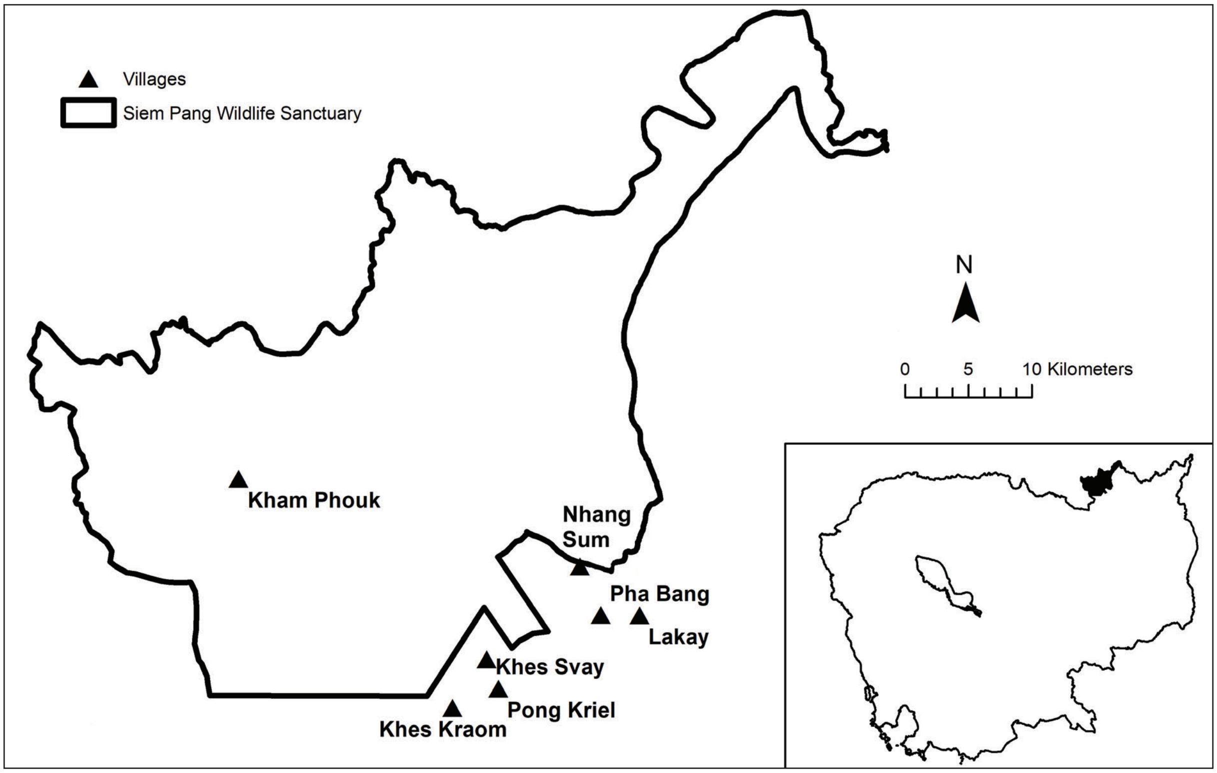

Fig. 1 Location of Siem Pang Wildlife Sanctuary (shaded area of inset map) and surrounding villages in northeast Cambodia. Our survey was conducted in Khes Svay village.

national Cambodia Programme, with nearly all surveys conducted by the same interviewer.

Questionnaire

To ensure that our questions were culturally suitable and relevant, these were developed in consultation with the local BirdLife International team, all of whom are Khmer-speaking Cambodians. The questionnaire included five sections: basic household information, demographics of the dogs owned over the past two years, dog management practices, dog and wildlife interactions, and attitudes on dog management based around the Likert scale. Five points were used on the Likert scale to measure attitudes towards seven statements (Fig. 2), ranging from 'strongly agree' to 'strongly disagree' and included a neutral option (Likert, 1932). There was a total of 36 questions in the survey, eight with additional dependent follow up questions, with each section having between four and 11 questions. The questionnaire was developed in English and then translated into Khmer. The English version is included in Annex 1.

Only one questionnaire was completed per household, with the head of the household being the preferred respondent. However, any adult member of the household was able to participate, and encouraged to do so if the head of the household was unavailable, which was common. We attempted to visit most households within the village, however some households are not occupied year-round, and there were sometimes difficulties finding an available adult willing and able to participate. The survey commenced in January 2020 to avoid the busiest harvesting and planting periods, with staff visiting households sporadically due to resource constraints. However, the survey was postponed in April 2020 due to Covid-19 concerns. The questionnaire recommenced in January 2021 until May 2021. The recorded responses were then translated into English and entered into a database by one member of the team.

Participant responses were tallied and summarised. These included basic demographic details for the villagers, as well as demographic and reproductive details of the dog population including the sex ratio, mean litter size and mean number of surviving puppies produced per adult female dog each year. To examine the potential for health concerns to motivate changes in attitudes and therefore dog management, we grouped respondents according to their awareness of disease

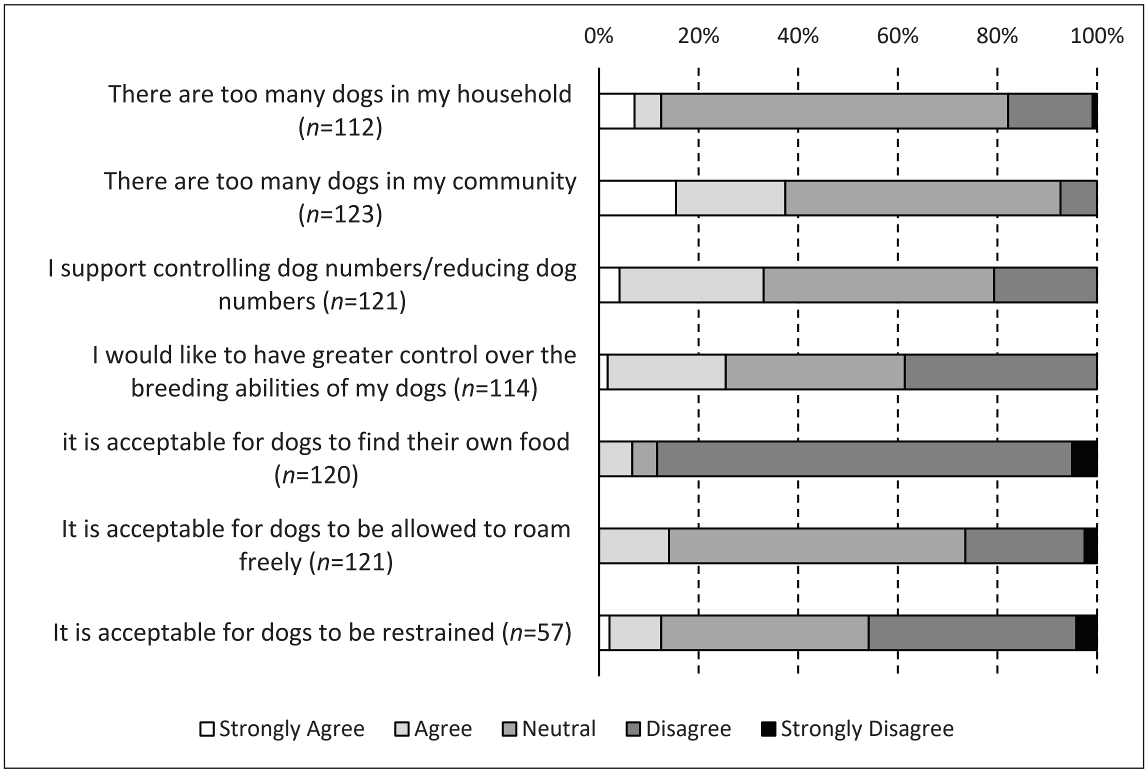

Fig. 2 Proportion of respondents in Khes Svay village who agreed and disagreed with the seven statements regarding dog ownership on a Likert scale.

transmission from dogs to humans (aware or unaware). We used a Mann-Whitney U test in R (R Core Team, 2021) to determine if there were any differences in the attitudes expressed towards the Likert scaled questions and if the mean number of dogs owned differed between these two groups.

Results

We surveyed 123 out of the 233 households (52.8%) in the village (41 of whom were surveyed in 2020). Data from both survey periods was pooled because responses were similar across all categories, except for our question about reasons for owning a dog, with the difference explained below. The response rates for each question were generally high, with fewer than 15 respondents choosing not to answer any given question. Questions that were dependent on preceding questions had smaller sample sizes, however their response rates were similar to independent questions. The exception to the high response rate was in the attitudes section, where 53.7% of respondents did not report their level of agreement regarding the statement about the acceptability of restraining dogs.

A total of 111 households (90.2%) reported owning at least one dog at the time of the survey, with eight households (6.5%) having owned at least one dog in the last two years. One household did not report their dog ownership. The mean number of people per household was 5.3 (range 2–12). The majority of households (82.1%) were involved in raising livestock and growing crops, while some (16.3%) engaged in only one of these activities and two households (1.6%) did not engage in either. Field houses, an additional dwelling of the household located outside of the village at their crop fields, were owned by 89 households (72.4%). When staying at their field houses, 70 households (79%) stated that they took their dogs with them. Nearly all households relied on forest resources to some extent, and mostly on non-timber forest products and fish, but also timber.

Dog demographics

The average rate of dog ownership across the four years discussed in the survey was 2.41 dogs per household (range 1–12), with a dog-to-human ratio of 1:1.8. The average litter size was 3.3 (min=1, max=8), with only one litter produced each year during the dry season or late wet season, most commonly in November or December.

A mean of 1.57 puppies survived per adult female dog in the population per year. The number of dogs taken out of the village, or new dogs brought into the village from elsewhere was not recorded. The sex ratio of the overall dog population was nearly equal with one male to every 1.03 females. However, the adult dog population was skewed towards females, with one male for every 1.5 females. Only five households stated that they attempted to manage their dogs' reproduction, which was achieved by sterilisation in four cases and by euthanasia of puppies in one case.

Dog management

Almost all respondents believed that all dogs in the village were owned, with only one respondent believing that two dogs were unowned. The primary reason for owning dogs in the majority (96.7%) of households was for security. However, only households sampled in 2020 provided multiple reasons for ownership, likely due to misunderstanding about allowing more than one answer to this question when the survey resumed in 2021. It was clear from the respondents in 2020 that multiple reasons for owning a dog is common, with 65.8% of respondents giving two or more reasons. For the 2020 cohort (n=38), pest management (34.2%), culture (28.9%) and hunting (26.3%) were also reported, as well as single mentions of breeding to produce puppies for sale and herding livestock. Nearly all households fed their dogs daily, whereas two households fed their dogs 4–6 times per week and three households only 1–3 times per week. All households fed their dogs rice. Aside from one household, all respondents reported that they fed food waste to their animals. None of households kept their dogs restrained or contained at any time.

In response to questions on roaming behaviour, which was defined as wandering unaccompanied from the household area, 82% of households believed their dogs did not roam, while 17% stated their dogs sometimes roamed and 1.8% believed their dogs often roamed. For households that had roaming dogs, the majority thought their dogs roamed for 1–2 hours or for no more than half a day, with only one household believing their dog roamed for a day, two believing their dogs roaming for 2–3 days and one having a dog that roamed for a week or longer. Most households did not see any particular pattern to their dog's roaming.

Only one household reported that at least one of their dogs had been vaccinated, although they did not specify the vaccine or disease. The type of care provided by households when their dog is sick is indicated in Fig. 3, with the majority (51.2%) providing some form of medicine or traditional remedy, and only two households seeking veterinary care. When asked if they had any awareness of diseases that dogs could transfer to people, 56% of respondents had some level of awareness and 80% of these were concerned with the issue.

Dogs and wildlife

Respondents were asked about their observations of dog-wildlife interactions in the forest. The majority (60%) of respondents reported that they never took their dogs into the forest, whereas 29% stated they sometimes did, 9% said they usually did and 2% always took their dogs with them. Respondents reported seeing other people with dogs in the forest always (5%), usually (16%), sometimes (49%) and never (30%). They also indicated how often the dogs seemed to stay within calling distance of their owners (as estimated by the respondent) while in the forest, with only 15% saying this always seemed to be the case. The number of dogs that respondents took into the forest, or saw other people with, typically ranged from one to five, with two dogs being the most common answer. Only 10% of respondents stated that they had seen dogs hunting or harassing wildlife in the forest, with reptiles and rodents the most common prey, but also birds. Similarly, only 12% of respondents stated that dogs hunted or harassed wildlife around the village and fields and the same prey were specified. Only seven

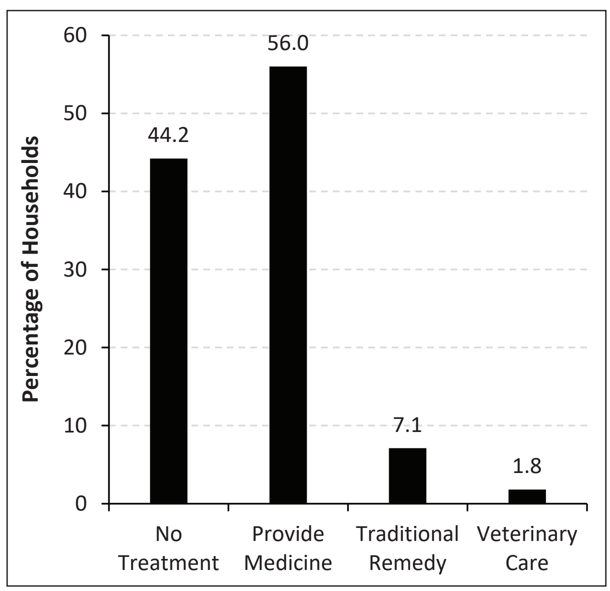

Fig. 3 Percentage of households (n=113) providing different treatments to their dogs when sick. The type of medicine was often unspecified although paracetamol was the most commonly specified, despite its toxicity in dogs. Multiple answers were allowed.

respondents (5.7%) gave incomplete or no answers to our hunting related questions.

Dog ownership and Management options

We found owners had a strong preference for female dogs, with 74.6% of respondents preferring females and only 2.5% having no preference. The main reason (73.9%) given for this preference was that female dogs could be used for breeding, with one respondent specifically mentioning doing this for financial gain. In contrast, of owners that preferred male dogs, just under half (48.1%) preferred them because they did not produce puppies. There was also a belief that one sex was less likely to roam and stay at home. However, 24 respondents believed this was true of female dogs, while seven respondents thought it was true of male dogs. Only 4% of respondents preferred male dogs for reasons relating to improved security, with these indicating they believed male dogs to be bigger or stronger.

The results of attitudinal statements measured on the Likert scale indicated that most respondents did not have strong attitudes about most of the issues we raised (Fig. 2). More respondents indicated that they felt the community had too many dogs (37%) and were in support of controlling dog numbers (33%) than those who disagreed with these statements (7% and 21%, respectively). However, respondents were slightly less keen about managing reproduction among their own dogs (with only 25% wanting greater control of their breeding). Of those that were keen, 76% preferred to euthanise puppies, with the remainder preferring sterilisation. Respondents who felt that they had too many dogs also tended to agree that the community also had too many. In contrast, respondents who did not think they had too many dogs were mixed on whether the community had too many dogs. The majority (88.3%) of respondents did not think it was acceptable for dogs to find their own food. Of the 7% of respondents that thought this was acceptable, three disagreed with allowing dogs to roam, which seems a contradiction in allowing dogs to find their own food. Only 11% of respondents felt it was acceptable to restrain dogs and these respondents also disagreed with or had no opinion about allowing dogs to roam. This disagreement with restraining dogs was confirmed with a follow up question about how often dogs should be restrained, with 84% saying never, and with responses for restraining dogs during the day (7%) only narrowly surpassing restraining dogs only sometimes (5%), or only at night (5%).

Our Mann-Whitney U test did not find any significant differences in opinions between respondents that were aware of disease transmission between humans and dogs (57.5%) and those that were unaware (42.5%), except on whether it was acceptable to restrain a dog (Table 1). In this case, those unaware of disease transmission were more likely to disagree with this statement, while those aware were more likely to be neutral on the matter. The average number of dogs owned by respondents unaware of disease transmission from dogs to humans was higher than those aware of this, although the difference was not statistically significant (p=0.06814).

Table 1 Results of Mann-Whitney U comparisons of the attitudes of respondents aware and unaware of disease transmission between humans and dogs.

Statements	Test Statistic	p
There are too many dogs in my household	1517	0.991
There are too many dogs in my community	1947	0.275
I support controlling dog numbers/ reducing dog numbers	1895	0.230
I would like to have greater control over the breeding abilities of my dogs	1731	0.225
It is acceptable for dogs to roam freely	1974	0.130
It is acceptable for dogs to be restrained	249	0.016

Discussion

Our study provides useful knowledge and insights into the domestic dog population around SPWS and practices and attitudes of Cambodian households towards dog ownership. With a mean ownership of 2.41 dogs per household and virtually all households owning at least one dog at the time of the survey or some point in the prior two years, it is clear that any management interventions for dogs will need to be supported by the entire community through engagement. The dog-to-human ratio (1:1.8) we recorded contrasts with the average ratio recorded in the Kandal and Battambang provinces of Cambodia (1:3.8 and 1:3.3 respectively; Chevalier *et al*., 2021), indicating that rates of dog ownership are lower in our study area. To improve understanding of the dynamics of the dog population, further investigation into the role that in- and out- migration of dogs plays is needed, as well as the causes of and age at death. Given that the rate of puppy survival we recorded per female per year was less than two, emigration likely plays an important role. A longer-term study would be helpful to understand population fluctuations and determine the possible drivers of these.

We found the adult dog population was slightly skewed towards females, although it was unclear whether the local preference for female dogs played a role in this, and the overall sex ratio was approximately equal. Chevalier *et al*. (2021) also found relatively even sex ratios for dog populations in the Battambang and Kandal provinces, as did Morters *et al*. (2014) in South Africa. This contrasts with the usually male-skewed sex ratios reported for village dog populations around the world (Boitani *et al*., 2007; Ortolani *et al*., 2009; Ruiz-Izaguirre *et al*., 2014; World Health Organization, 1988). The preference for female dogs in Khes Svay also contrasts notably with survey responses in Kandal and Battambang, where male dogs were the clear preference (Chevalier *et al*., 2021). The importance many owners place on their dog's ability to breed may pose a challenge to controlling reproduction among domestic dogs. While many respondents felt that there were too many dogs in the community, fewer believed that they owned too many dogs, and the preference for female dogs, largely for their ability to breed, suggests this is important to most owners and may be an economic benefit for some. While only a relatively small number of respondents stated a preference for a reproductive control method, the majority preferred euthanasia of puppies, rather than sterilisation, suggesting that they prefer to maintain their dog's fertility. The use of temporary contraceptives might be more palatable for the community; however, achieving reproductively significant uptake may be a challenge. While the barriers to accessing different reproductive control methods could have influenced the respondent's preferences, this was not examined. Reducing the mortality rate of dogs would likely assist in reproductive management, as owners may feel less need for a ready supply of new puppies. Further investigation into the reasons behind the unwillingness of local people to use fertility control should be undertaken.

It was clear from our study that security was the primary purpose for owning a dog. Unfortunately, only the first cohort of respondents gave secondary reasons, but it is still apparent that culture, hunting and pest management are also important reasons for owning a dog. The importance of security, and hence the requirement of constant presence of the dog at the house to provide this service, may encourage owners to consider changes in the way they manage their dogs to prevent roaming. However, with only 18% of respondents believing or admitting that their dogs roam, and the majority of these for only a few hours, convincing owners to curb roaming behaviour may be challenging. Evidence of the roaming behaviour of dogs from Khes Svay village is available: Ladd *et al.* (2023) determined the activity ranges and patterns of ten dogs from the village and found that half of them entered the wildlife sanctuary on roaming forays. Presenting this information in an appropriate format may be useful for convincing villagers of the problem. The use of dogs for hunting and pest management may conflict with efforts to reduce roaming and the taking of dogs into the forest. While hunting in the wildlife sanctuary is prohibited and ranger patrols attempt to enforce this, it does occur and the areas considered appropriate for pest management by their owners may conflict with wildlife protection. Further investigation is needed to quantify the impact dogs may have on wildlife in Siem Pang Wildlife Sanctuary, as much of the existing evidence has been opportunistically collected by sanctuary rangers when they encounter such events.

Just over half of our respondents had some degree of awareness about diseases that could be transmitted from dogs to humans, and of those that were aware, all were concerned with this issue. However, there was little difference in the attitudes of these two groups towards the management statements and the rate of dog ownership. The depth of knowledge on disease transmission was not investigated, so it is unclear how detailed the knowledge of those aware of the issue was, and whether a deeper understanding might result in changes to attitudes. The integration of human health and animal welfare objectives into dog management programmes has been identified as a useful strategy for increasing community uptake in such programmes, rather than focusing only on conservation (Doherty *et al*., 2017). Health is a more direct concern to the everyday lives of local people and may provide greater motivation for behavioural changes.

While the response rate to all of our questions was generally high, there are indications that respondents may not have been entirely truthful or may have given answers that they thought we wanted to hear in relation to some topics. This issue was discussed during the development of the survey and the role Rising Phoenix (formerly BirdLife International) plays in the enforcement of protected area laws and regulations was seen as a potential hinderance to receiving honest answers, particularly regarding dog-wildlife interactions. This was apparent with many of the respondents who stated one of their reasons for owning dogs was for hunting, yet contrary to this, they also claimed to have never seen their dog or anyone else's dog hunting or harassing wildlife. While there are forested areas outside of the wildlife sanctuary that could be used for hunting, these areas are increasingly degraded by logging and clearance for new rice fields. Such discrepancies in the data seem to be largely limited to the questions regarding dog-wildlife interactions, so these responses need to be considered with some scepticism. To better gauge local experiences of dog-wildlife interactions, an alternative method may be needed in future investigations, such as bringing in an unaffiliated team to undertake research, and/or using an unmatched count technique that allows for differences in responses to sensitive questions to be measured (Hinsley *et al*., 2019; Cerri *et al*., 2021).

The dearth of strong views on options for dog management suggests that the community may be open to education and behavioural change. However, lack of resources poses a major barrier to effective management of dogs. All households feed their dogs with rice and almost all also feed them with food scraps, however it is unclear how often this occurs, and the frequency, quantity and nutritional value is likely highly variable. Given this information on diet, and the relatively poor body condition of many dogs observed in the community, poor nutrition is likely very common. Cambodia is one of the poorest countries in Southeast Asia and households do not always have the resources to consistently provide their dogs with higher quality and quantities of food, or to construct dog proof fencing, actions which may reduce roaming and hunting. Domestic dogs that are underfed are more likely to prey on wildlife compared to dogs receiving adequate nutrition (Silva-Rodriguez & Sieving, 2011) and if an approach to improve nutrition were to be attempted, food subsidies might be needed in addition to education. Additionally, it would be important to create an appropriate experimental design for any trial to improve dog nutrition to determine that the desired result of reduced wildlife predation occurs without unintended consequences such as producing stronger dogs that are more effective hunters, or increasing the dog population. With 40% of respondents indicating they take their dogs with them to the forest at least sometimes, this issue is a potentially more feasible starting point for reducing the number of dogs in the forest, as it does not require owners to invest any resources in curbing roaming behaviour, such as restraining or containing them. However, it will be necessary to investigate the reasons why people take dogs into the forest, so that they can be appropriately addressed when attempting to change this behaviour. There was also a relatively large group that were against restraining dogs, which would challenge efforts to reduce roaming behaviour.

Our survey provides useful baseline data for developing a management strategy for free-ranging dogs. Although most owners stated they never take their dogs into the forest, we found that there was still a large proportion of households (40%) that do, and engaging this group to stop this will be important. Further investigation on why villagers take their dogs with them may be needed to develop a targeted education campaign and enforce rules regarding dogs in the wildlife sanctuary. It will also be critical to convince local people that dogs are a problem. This will require the collation of evidence, such as the roaming behaviour of dogs in Khes Svay village documented by Ladd *et al*. (2023) and ranger reports of dog-wildlife interactions, and presenting this in an appropriate format. The relatively poor nutrition of dogs has potential for improvement to reduce their roaming and hunting, however careful consideration on how this could be made to work is needed given the limited resources locally and potential for unintended consequences. The integration of health into a management strategy for dogs appears to be a promising avenue, with awareness on this topic limited. Improving dog health as part of a more comprehensive strategy would not only have benefits for wildlife, but human, dog and livestock health as well.

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Annex 1 Study Questionnaire

Household Information

1. How many people currently live in your household?
   Adults Children
2. Do you keep livestock and/or grow crops?
   Livestock / Grow crops / Both
   a. What livestock do you keep?
   Chickens / Ducks / Cows / Buffalo / Pigs / Goats / Other (specify)
3. Do you have a field house?
   Yes / No
   a. Do your dogs go with you when you stay there?
   Yes / No
4. How do you dispose of food waste?
   Give to Animals / Burn / Bury / Other
5. Do you use forest resources?
   Non-Timber Forest Products / Fish / Collect Wood / Other (specify)

Dog Demographics

1. During the past 2 years, how many dogs have you owned, how many died or were given away, and how many do you have now?
2. How many of the following do you have now?
   a. Male adult dogs
   b. Female adult dogs
   c. Male juvenile dogs
   d. Female juvenile dogs
   e. Male puppies
   f. Female puppies
3. How many litters have your female dogs had during the past 12 months?
   Dog 1: Dog 2: Dog 3:
   a. How many puppies in each litter?
   Litter 1: Litter 2: Litter 3:
   Litter 4: Litter 5:
   b. How many puppies died, how many lived?
   Litter 1: Died Lived
   Litter 2: Died Lived
   Litter 3: Died Lived
   Litter 4: Died Lived
   Litter 5: Died Lived
   c. What times of year were the litters of puppies born?
   OR
   Wet season / Dry season
   d. Are your female dogs currently pregnant or have puppies?
   Dog 1: No / Pregnant / Puppies
   Dog 2: No / Pregnant / Puppies
   Dog 3: No / Pregnant / Puppies
4. Do you try to control your dogs' reproductive output?
   Yes / No
   a. How, and for which dogs?
   Spay or Neuter / Prevent Mating / Euthanise Puppies / Other (specify)

Dog Management

1. Why do you own dogs? (indicate order of importance if multiple reasons)
   Security / Pest Management / Hunting / Herding Livestock / Breeding / Cultural / Other (specify)
2. In the last week, how often did you feed your dogs (not leftovers/food waste)?
   Daily / 4-6 times / 1-3 times / Never
3. In the last week, what did you feed your dogs?
4. In the last week, on how many occasions was it restrained or contained (i.e. kept from roaming)?
   Never / 1–3 days / 4–6 days / Always
   a. On these occasions, for how long was it restrained?
   All Day & Night / Night Only / Day Only / More than Half the Day / Less than Half the Day
5. How do you restrain or contain your dog?
   Tethered / Penned / Dog-Proof Fenced Yard / Other (specify)
6. How often does your dog roam outside of your yard/roam outside of calling distance?
   Never / Sometimes / Often / Always
   a. When roaming, in general, how long is the dog away before coming back?
   1–2 hours / Half a Day / One Day / 2–3 Days / A Week or More
7. What time does your dog start roaming (i.e. not being around the house)?

Morning / Midday / Afternoon / Evening / Night / Anytime

8. When does your dog usually return?
   Morning / Midday / Afternoon / Evening / Night / Anytime
9. Has your dog ever been vaccinated?
   Yes / No
10. If your dog is sick or injured, what do you do?
    Do Nothing / Treat with Traditional Remedies / Seek Veterinary Care / Other (specify)
11. Are you aware of the diseases that dogs can pass on to humans, as well as other dogs, such as rabies and toxocariasis?
    Yes / No
    a. Are you concerned about this?
    Very concerned / concerned / neutral / unconcerned / very unconcerned

Dogs and Wildlife

1. Do your dogs go with you into the forest?
   Never / Sometimes / Usually / Always
2. Do you see other people taking dogs into the forest?
   Never / Sometimes / Usually / Always
3. How many dogs usually go with you or do you see with others?
4. Do the dogs stay within calling distance during the trip?
   Never / Sometimes / Usually / Always
5. In the last 12 months, have you observed your own or other dogs hunting or harassing wildlife in the forest?
   Yes / No
   Ungulate / Rodent / Bird / Reptile / Other (specify)
6. In the last 12 months, have you observed your own dogs or other dogs hunting or harassing wildlife around the village, or fields maintained by your village?
   Yes / No
   Ungulate / Rodent / Bird / Reptile / Other (specify)

Opinions

1. How many unowned dogs do you think live in the village?
2. Do you have a preference for male or female dogs?
   Males / Females. Why? Reason:

What is your opinion on the following statements?

1. There are too many dogs in my household.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
2. There are too many dogs in this community.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
3. I support controlling dog numbers/reducing dog numbers.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
4. I would like to have greater control over the breeding abilities of my dogs.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
   a. What are your preferred methods, in order of preference?
   Spay and Neuter / Euthanise Adults / Euthanise Pups / Other (specify)
5. It is acceptable for dogs to find their own food.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
6. It is acceptable for dogs to be allowed to roam freely.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree
7. It is acceptable for dogs to be restrained.
   Strongly Agree / Agree / Neutral / Disagree / Strongly Disagree

A first survey of the dragonflies (Odonata) of Siem Pang Wildlife Sanctuary, northeast Cambodia

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មូលន័យសង្ខេប

ដែនជម្រកសត្វព្រៃសៀមប៉ាង គឺជាតំបន់ជីវចម្រុះសំខាន់ជាសកលដែលស្ថិតនៅភាគឦសាននៃប្រទេសកម្ពុជា។ លក្ខណៈពិសេសនៃតំបន់នេះគឺមានប្រភពទឹកជាង 200 កន្លែងដែលគេស្គាល់ថាជាត្រពាំង និងមានលក្ខណៈឆាប់រីងស្ងួត។ ការសិក្សានេះបង្ហាញពីទិន្នន័យដំបូងស្តីពីសត្វកន្ទុំរុយ (លំដាប់ Odonata) នៅដែនជម្រកសត្វព្រៃសៀមប៉ាងដែលមិនធ្លាប់មានការសិក្សាពីមុនមក។ សត្វកន្ទុំរុយចំនួន ៥៧ ប្រភេទ ត្រូវបានកត់ត្រា។ ទាំងនេះរួមមានកំណត់ត្រាលើកទីពីរនៃ *Copera chantaburii* (Asahina, 1984) និងកំណត់ត្រាប្រទេសដំបូងនៃ *Aciagrion paludense* (Fraser, 1922) សម្រាប់ប្រទេសកម្ពុជា ដែលពីមុន *A. paludense* ត្រូវបានគេចាត់ទុកថាជាប្រភេទតែមួយ (synonym species) ទៅនឹង *A. occidentale* (Laidlaw, 1924) ។ ភស្តុតាងសម្រាប់ចំណែកថ្នាក់ *A. paludense* ជាប្រភេទមួយផ្សេងដាច់ដោយឡែកត្រូវបានបង្ហាញផងដែរក្នុងការសិក្សានេះ។

Abstract

Located in northeast Cambodia, Siem Pang Wildlife Sanctuary is an internationally important site for biodiversity. A significant feature of the wildlife sanctuary is the presence of at least 200 water bodies known as trapeangs, which are often ephemeral. This paper presents the first data on the Odonata (dragonfly) fauna of the sanctuary which had not previously been studied. Fifty-seven species were recorded. These include the second record of *Copera chantaburii* (Asahina, 1984) for Cambodia and the first country record for *Aciagrion paludense* (Fraser, 1922), which was formerly considered a junior synonym of *A. occidentale* (Laidlaw 1924). A justification for why *A. paludense* should be considered a distinct species is also provided.

Keywords Dragonflies, lowland deciduous dipterocarp forest, lowland semi-evergreen forest, Odonata, Siem Pang Wildlife Sanctuary.

Introduction

Dragonflies and damselflies belong to the insect order known as Odonata. The term 'dragonflies' is often used to refer to both dragonflies (suborder Anisoptera) and damselflies (suborder Zygoptera) and is used in this paper unless differentiation is required. Dragonflies and damselflies have similar structure in possessing a head, thorax and a ten-segment abdomen, but differ in the following ways: dragonflies are bigger with broader bodies, whereas damselflies are smaller and more delicate insects. Both have two pairs of wings although dragonflies have unequal sized forewings and rear wings and generally rest with their wings open, whereas damsel-

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flies generally close their wings together when resting, although there are exceptions to this rule. Both have large compound eyes, although unlike dragonflies, the eyes of damselflies are widely spaced. Dragonflies and damselflies are predatory insects which emerge from aquatic larvae. They are voracious predators at all stages of their life cycles. Worldwide, there are currently 6,410 recognized species of dragonfly (Paulson *et al*., 2023).

Including Thailand, Vietnam, Laos and Cambodia, Indochina supports over 500 species of dragonflies (Hämäläinen, 2004). Hämäläinen (2004) also stated "Thailand has the most diverse and best known odonate fauna, but knowledge of the Laotian and Vietnamese fauna has increased rapidly over the last 10 years". However, the Odonata of Cambodia remains the most poorly studied within the region (Kosterin, 2016). The exact number of species recorded in Cambodia has not been published but based on information from Oleg Kosterin is currently 203.

The primary aim of this study was to increase knowledge of the odonate fauna of Siem Pang Wildlife Sanctuary in northeast Cambodia (Fig. 1) and the importance of trapeangs (natural waterholes) for these. A secondary aim was to increase knowledge of Cambodian and Indochinese Odonata. Dragonflies are aquatic in their larval stage and are considered key indicators of pollution, habitat quality and landscape disturbance (Šigutová *et al*., 2022). Because monitoring within the wildlife sanctuary relies on such species information to assess environmental changes, the data provided by this study will enhance understanding of the sanctuary. The dragonflies of Siem Pang had not been previously studied and the nearest lowland site where these have been surveyed in Cambodia is the Prey Long Wildlife Sanctuary (Kosterin, 2020), which extends into southwest Stueng Treng Province.

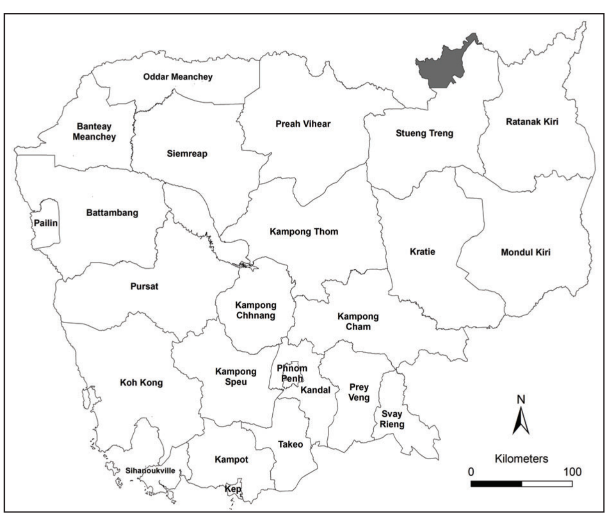

Fig. 1 Location of Siem Pang Wildlife Sanctuary (grey shading) in northeast Cambodia (© Rising Phoenix).

Methods

Siem Pang Wildlife Sanctuary was visited at the start of the dry season in Cambodia from 10–26 November 2022. The timing was partly chosen to improve access but primarily to maximize time in the field, although it is acknowledged that species diversity may be different or greater during the wet season. At the time of survey, the landscape was in transition from the wet to the dry season, with much standing water remaining. However, most days were dry and sunny which aided observation and daily excursions of varying length were made to various study locations.

Adult dragonflies were photographed and videoed using a Panasonic Lumix GH5S camera with a Panasonic 100–400mm lens. Site coordinates were recorded using a Garmin Fenix 6S Pro watch with the Garmin Explore Application. Photographs of the lectotype and paralectotype of *Aciagion paludense* (Fraser, 1922) from the Natural History Museum, London, were kindly provided by Dan Hall and Benjamin Price and used to review its taxonomic status.

Siem Pang Wildlife Sanctuary covers a total of 132,321 ha in Stung Treng Province. The sanctuary is contiguous to the northwest with Xe Pian National Park in Laos and to the east and southeast in Cambodia with Virachey National Park and Veun Sai-Siem Pang National Park. Virachey National Park borders Chu Mom Ray National Park in Vietnam (Loveridge *et al*., 2018). This 11,207 km^2 corridor of protected areas forms one of the largest nominally protected areas in the Mekong Basin (Eang *et al*., 2021). The elevation of the sanctuary ranges from 60 m a.s.l. at the Sekong River to 400 m a.s.l. on the northwestern boundary with Laos (United States Army Map Service, 1967, as cited by Loveridge *et al*., 2018).

The wildlife sanctuary has been managed since 2016 by Rising Phoenix, a social enterprise company, in partnership with the Ministry of Environment. The sanctuary consists of two main habitat zones. To the north of the O'Khampa River and the east of the Sekong River, the sanctuary supports lowland semi-evergreen forests with smaller areas of evergreen and deciduous dipterocarp forest. Lowland open deciduous dipterocarp forests occur south of the O'Khampa River and west of the Sekong River (Eames & Costello, 2012) (Fig. 2). The latter area was previously designated as Siem Pang Khang Lech Wildlife Sanctuary until it was combined with Siem Pang Wildlife Sanctuary in 2019 (Eang *et al*., 2021).

Within the area of deciduous dipterocarp forest there are at least 200 often ephemeral water bodies known in Khmer as trapeangs (Eames, 2014), a phrase which translates as a natural waterhole or pond. Trapeangs range significantly in size from 100 m^2 to 10,000 m^2 and vary in depth and vegetation cover. Domestic buffaloes now use the trapeangs as wallows which serves to maintain these flooded depressions. Historically they would have also been used by wild water buffalo *Bubalus arnee* (now extirpated) and Asian elephants *Elephas maximus*, although the last documented record of the latter was in 2016 (Loveridge *et al*., 2018).

The monsoonal climate of northern Cambodia gives rise to significant dry and wet seasons and many trapeangs dry out whereas others retain small amounts of water. Six trapeangs are now managed with the provision of solar-powered water pumps which maintain water levels during the dry season. Some trapeangs have a rich growth of sedges and emergent vegetation whereas others are largely free of plant life and have barren muddy banks such as Trapeang Thamatkon (Fig. 3C) which was not surveyed. Surrounding habitat also varies, as some trapeangs are in open savannah type landscape and others within more shaded forest (Fig. 3).

Trapeangs are an important part of the ecosystem in Siem Pang Wildlife Sanctuary and provide essential feeding and breeding habitats for a variety of animal species. As such, this study provided the opportunity to assess their importance for dragonflies. In addition, the study also looked at two larger waterbodies, Beoung Khampa, which was deepened in early 2022, and Beoung Nava, the site of a Siamese crocodile *Crocodylus siamensis* reintroduction. Both are situated in semi-evergreen forest. The word "Beoung" translates as lake.

Riverine habitats were also visited. These included a small seasonal stream the O'Anchan in deciduous dipterocarp forest. As the visit occurred in the dry season, the stream was not flowing and reduced to pools in the stream bed, although some were ten metres long. Flowing water was found on the Sekong River which bisects Siem Pang Wildlife Sanctuary, and a tributary known as the O'Khampa. Three days were also spent in the northern part of the protected area in semi-evergreen forest close to the Laos border. This included the O'Chongheang River (Fig. 4) which had a mix of narrow fast-flowing water and wider slow-flowing areas. Accessing riverine habitats was often quite difficult due to steep-sided river banks, deep water and muddy riverbeds.

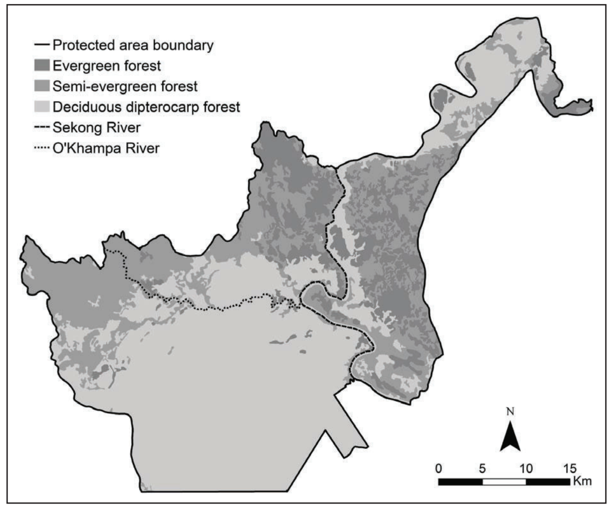

Fig. 2 Major rivers and habitat types in Siem Pang Wildlife Sanctuary (© Rising Phoenix).

Eighteen locations were surveyed in Siem Pang Wildlife Sanctuary which broadly fell into two areas: i) trapeangs and a single stream in deciduous dipterocarp forests south of the O'Khampa River (sites 1–9; Fig. 5) and, ii) areas within semi-evergreen forests north of the O'Khampa River (sites 10–18; Fig. 6). With the exception O'Chongheang River, all locations were visited during daytime excursions from a tented camp (site 9; Fig. 5). The locations surveyed were as follows:

Site 1. Trapeang Lumtea (14.154634°N, 106.277208°E) (Fig. 3A). A medium-sized trapeang covering 2,500 m^2. This was the most vegetated trapeang visited, with extensive areas of sedge and emergent vegetation surrounding a small area of open water. The area was surrounded by open deciduous dipterocarp forest which did not shade the trapeang. There was evidence of recent domestic buffalo wallows although this was relatively small compared to other trapeangs. Water levels in the trapeang are maintained by a solar-powered water pump. The site was visited more than any other during the study, with four visits in total. All visits were made in the morning, no earlier than 08:00 hrs and not after 12:30 hrs. The dates of visits were 10, 11, 14 and 26 November 2022. Seventeen species were recorded at the location.

Site 2. Trapeang Thymea (14.179192° N, 106.289633° E). This trapeang covers 2,500 m^2, similar to Trapeang Lumtea, but it was difficult to access sedges and emergent due to high buffalo wallowing activity during the visit. Only one species, *Diplacodes trivialis*, was commonly seen although the trapeang was judged to have far more potential given the extent of vegetation and open water. This site was visited for just 15 minutes on 10 November

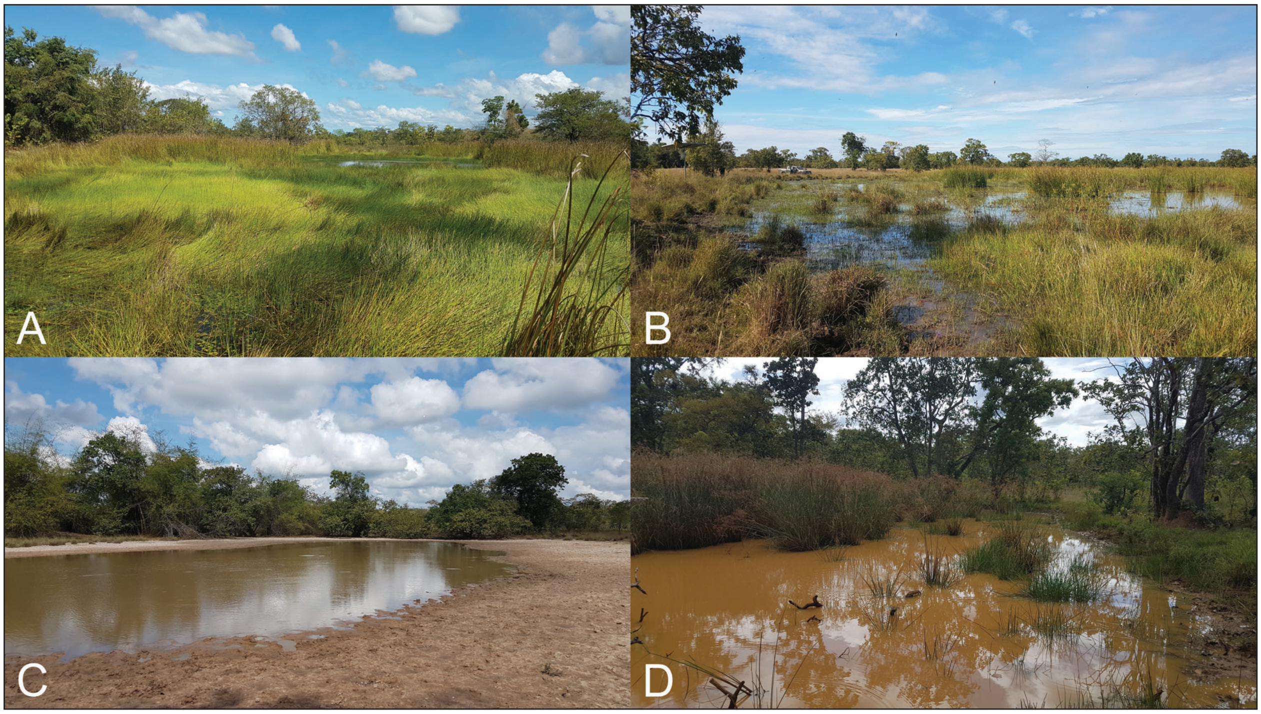

Fig. 3 Examples of trapeangs in Siem Pang Wildlife Sanctuary: A) Trapeang Lumtea, B) Trapeang K'Dung, C) Trapeang Tamatkon, D) Trapeang Buon Ch'rung.

2022 but was often briefly visited as an unrecorded stop while returning to camp from other sites and the heat of the day may have also reduced dragonfly activity.

Site 3. Trapeang Lumpon (14.186599° N, 106.261940° E). A trapeang measuring 2,500 m^2 with muddy edges and a small amount of surface vegetation. The site was only visited for 15 minutes on the morning of 11 November 2022, with just five species of dragonflies noted.

Site 4. Trapeang K'mun (14.170951° N, 106.232289° E). A muddy-edged trapeang similar in size to Trapeang Lumpon. This was visited briefly for 15 minutes on the morning of 11 November 2022 and no dragonflies were recorded.

Site 5. O'Anchan stream (14.177317° N, 106.254600° E). The stream was largely reduced to a series of stagnant pools along a high-sided streambed surrounded by areas of thick bamboo and deciduous dipterocarp forest. Much of the stream was heavily shaded and offered potential for interesting dragonflies not found in the trapeangs. The site was visited four occasions, although three comprised brief stops at the bridge on the way to and from other sites. One full morning on 15 November 2022 was spent exploring the area and ten species were recorded.

Fig. 4 O'Chongheang River, Siem Pang Wildlife Sanctuary.

Site 6. Trapeang K'Dung (14.181020° N, 106.219584° E) (Fig. 3B). A fairly large trapeang within an open savannah grassland known as Veal Kreel. The site had extensive sedge borders and quite a lot of open water with small amounts of emergent vegetation. Water levels are maintained with a pump and there was some evidence of buffalo wallows. This site was visited on 11 and 13 November 2022. Each visit lasted approximately one hour and the first visit was in mid-afternoon and the second was in mid-morning. Surprisingly few dragon-

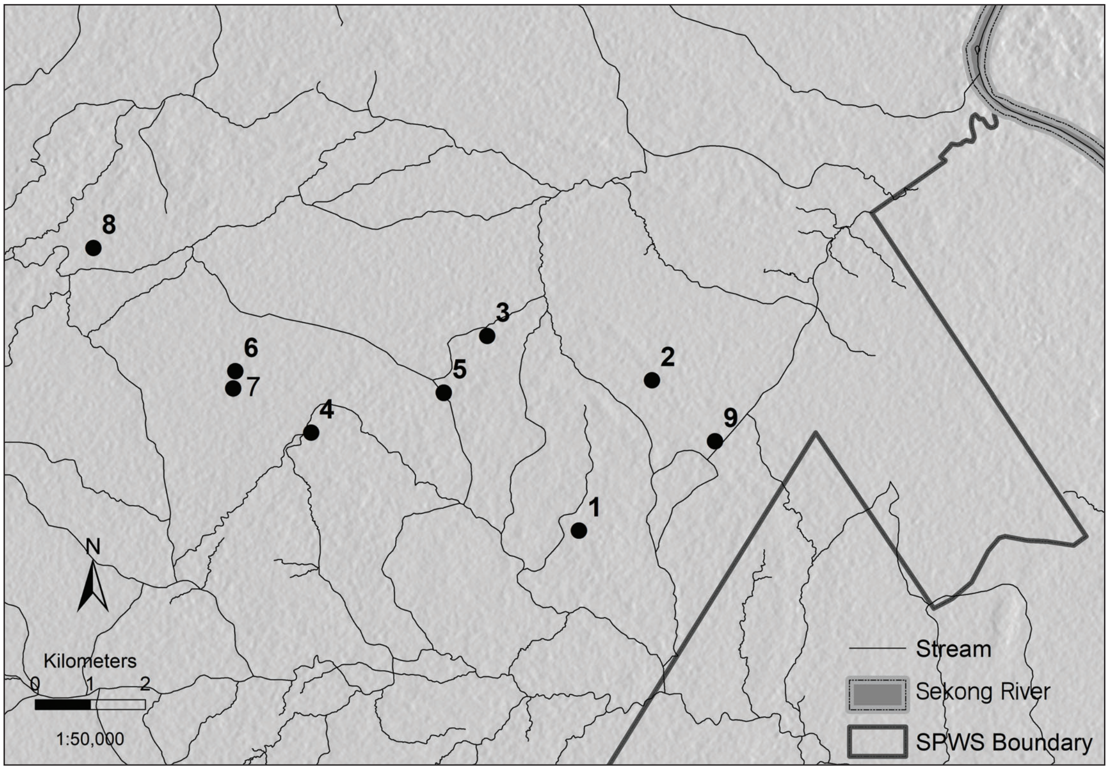

Fig. 5 Areas visited in deciduous dipterocarp forest south of the O'Khampa River (© Rising Phoenix).

Fig. 6 Areas visited in semi-evergreen forest north of the O'Khampa River (© Rising Phoenix).

flies were present, although plenty of fish were present and it is unknown if these may have reduced the diversity of dragonflies. Seven species were recorded at this location.

Site 7. Trapeang Trau (14.178204° N, 106.219224° E). A small trapeang close to Trapeang K'Dung and on the edge of Veal Kreel and so surrounded by large deciduous dipterocarp trees and large tracts of bamboo. Very little emergent vegetation. There were significant areas of buffalo wallows which made access difficult. The site was visited once on 13 November 2022 from 08:10 to 09:00 hrs. Five species were recorded at this location.

Site 8. Trapeang Buon Ch'rung (14.201366° N, 106.195833° E) (Fig. 3D). Quite different from other trapeangs in being quite small and shaded by tall deciduous dipterocarp forest. The water was stagnant and muddy from wallowing buffaloes. Extensive areas of sedge but little other emergent vegetation was present. Four species were recorded at this location which was visited once for 45 minutes around noon on 13 November 2022.

Site 9. Tented camp (14.169157° N, 106.300168° E). The camp was situated in deciduous dipterocarp forest adjacent to a small strip of semi-evergreen woodland. A small trapeang occurs behind the camp which is fenced and has little emergent vegetation. This is surrounded by channels which were dry at the time of the survey but contain water during the wet season. Six species were recorded at the camp.

Site 10. Beoung Nava (14.303005° N, 106.179607° E). A sedge-filled lake in the northwest of the sanctuary. The lake had some open water and large areas of boggy margins. At the western end, there was a boggy area which received a lot of shade from small trees. The area is situated in semi-evergreen forest. The lake was visited on 12 and 17 November 2022. The first visit was from 14:20 to 16:00 hrs and the second was from 11:00 to 12:30 hrs. Eighteen species were recorded at the location, although it may support more dragonfly species.

Site 11. Stream near Beoung Nava (14.307832° N, 106.189416° E). Surveyed on 17 November 2022 from 13:00 to 13:45 hrs. It was quite well shaded within semi-evergreen forest and thick bamboo. Five species were recorded at this location.

Site 12. Beoung Khampa (14.302597° N, 106.217422° E). A large lake that was deepened in early 2022. There was open water with areas of sedge in the middle. The site is close to the O'Khampa River and surrounded by a mixture of semi-evergreen and deciduous dipterocarp forest. Considering significant digging had occurred as part of the deepening work less than ten months before my visit, the site was regenerating very well and numerous dragonflies were present during my first visit. The site was visited from 11:20 to 13:30 hrs on 12 November and from 14:30 to 16:00 hrs on 17 November 2022. The first visit was far more productive and 23 species were recorded at the location.

Site 13. Unnamed trapeang on approach track to Beoung Khampa (14.314928° N, 106.272947° E). An overgrown trapeang which was visited for 50 minutes at 10:00 hrs on 12 November 2022. The actual trapeang was not accessed but a small area of tall grassland adjacent to it was searched. Two species were recorded.

Site 14. Koh Dat Tum, Sekong River Island (14.312215° N, 106.340867° E). This river island was visited from 08:30 to 10:00 hrs on 16 November 2022. The habitat was really just a large sand bank with little vegetation in the middle of the Sekong River. Six species were recorded at this location.

Site 15. Section of O'Khampa River approximately 2 km upstream from the confluence with the Sekong River (14.297723° N, 106.287076° E). A deep-sided, slow-flowing river approximately 20 m wide and bordered by thick semi-evergreen dipterocarp gallery forest. All observations were made from a boat. Attempts were made to walk on the riverside but this proved difficult and probably influenced the number of species observed. The location was visited from 10:30 to 14:00 hrs on 16 November 2022 and 13 species were recorded.

Site 16. O'Khampa River, on the Sekong River side of old bridge (14.214326° N, 106.319378° E). A narrower section of the O'Khampa River. Entered below the bridge and easier to access on foot than site 15, although wading chest deep in the river was required to survey the area. The river was faster flowing than downstream areas and the banks were a little more open with smaller shrubs rather than trees. The location was visited from 09:00 to 13:00 hrs on 19 November 2022 and seven species were recorded.

Site 17. O'Khampa River, west of the new bridge (14.296045° N, 106.281176° E). Significant areas of damage were evident due to construction of the new bridge but a small area of rocky rapids warranted investigation. The location was visited from 13:30 to 14:30 hrs on 16 November 2022 and 11 species were recorded.

Site 18. O'Chongheang River (14.350838° N, 106.254641° E to beyond 14.352010° N, 106.253729° E) (Fig. 4). Located in semi-evergreen forest in the northern part of Siem Pang Wildlife Sanctuary, three days were spent at the river from 21 to 23 November 2022. Each day we walked upstream through some narrow, faster-flowing areas and some wider, slower-flowing areas, all of which were in semi-evergreen gallery forest. Water depths varied from shallow to chest deep. The substrate was generally stony in the fast-flowing sections and silty in slower-flowing sections. Looking at the area after our visit, I felt we did not explore far enough upstream as an area of rocky river bed, visible on Google Earth, would have warranted exploration. Walking downstream could also have been beneficial. However, the areas we did visit were interesting, with 27 species recorded, making it the most productive of all sites visited.

Results

A total of 57 dragonfly species were recorded during the survey, comprising 50 species recorded north of and including the O'Khampa River and 32 species south of this (Table 1). Survey results are presented in the order of species provided in the *World Odonata List* (September 2023 revision) by Paulson *et al*. (2023) which is based on Bybee *et al*. (2021) in turn.

Euphaeidae

1. *Euphaea inouei* Asahina, 1977 (Fig. 7)

Males were seen daily along faster flowing stretches of the O'Chongheang River. The highest count was of 12 individuals on 23 November.

Philosinidae

2. *Rhinagrion hainanense* Wilson & Reels 2001 (Fig. 8)

Only recorded on the O'Chongheang River. All individuals were found along a steep-sided section of riverbank which was heavily shaded by bankside vegetation. Six individuals were observed over two days, including three males and two females on 22 November.

Calopterygidae

3. *Neurobasis chinensis* (Linnaeus, 1758) (Fig. 9)

Only recorded in two locations in semi-evergreen forest north of the O'Khampa River. A single female was recorded at a stream near Beoung Nava on 17 November and the species was recorded daily as common (>20 individuals) on the O'Chongheang River.

4. *Vestalis gracilis* (Rambur, 1842)

Commonly encountered in three locations where streams were surrounded by lush vegetation and shaded by the tree canopy. Often encountered in surrounding forest. Maximum daily counts of ten individuals were recorded on the streams near Beoung Nava and the O'Chongheang River. The species was also recorded on each visit to dry dipterocarp forests alongside the O'Anchan Stream.

Chlorocyphidae

5. *Heliocypha biforata* (Selys, 1859)

Recorded in low numbers in two locations. A single immature male was collected on the O'Anchan Stream on 13 November and the species was recorded daily on the O'Chongheang River with a maximum count of three.

6. *Libellago lineata* (Burmeister, 1839) (Fig. 10)

Recorded in three locations. One record of a single female at the tented camp on 17 November was unusual, and the only record in dry dipterocarp forest. However, the species was abundant along the O'Khampa and O'Chongheang rivers, with daily numbers exceeding hundreds.

Platycnemididae

7. *Copera chantaburii* Asahina, 1984

Only recorded on the O'Anchan Stream. Initially overlooked among the more common *Copera marginipes*. Examination of photographs proved at least three individuals were present in the shaded streambed on 15 November. Photographs (Figs. 11 & 12) and video were taken of an individual male and a copulating pair. More individuals were likely present and overlooked. Identification of the male was based on the pale abdomen tip being reduced to abdominal segment 10 and not extending into segments 8 and 9 (as it does in the case of *C. marginipes*) and most importantly, short paraprocts (Asahina, 1984). *Copera chantaburri* occurs in neighbouring Thailand and Vietnam but is largely unobserved in Cambodia. The first record for Cambodia was in June 2018 in Prey Long Wildlife Sanctuary (Kosterin, 2020).

8. *Copera marginipes* (Rambur, 1842)

The species was recorded at four locations where it was generally common. Common (≥20 individuals) at the O'Anchan Stream on 15 November, where it far outnumbered the similar *C. chantaburri*. Only two individuals were noted at Beoung Khampa on 17 November, although the species was common on all three days spent at the O'Chongheang River.

9. *Onychargia atrocyana* Selys, 1865

A single female was observed on 12 November at an unnamed and overgrown trapeang on the track leading to Beoung Khampa.

Table 1 Locations of dragonflies recorded during the study in Siem Pang Wildlife Sanctuary. Numbers indicate survey sites described in the methods section.

#	Species	Dry dipterocarp forest south of the O'Khampa River	Semi-evergreen forest north of the O'Khampa River
1	*Euphaea inouei*		18
2	*Rhinagrion hainanense*		18
3	*Neurobasis chinensis*		11, 18
4	*Vestalis gracilis*	5	11, 18
5	*Heliocypha biforata*	5	11, 18
6	*Libellago lineata*	9	15, 16, 18
7	*Copera chantaburii*	5	
8	*Copera marginipes*	5	12, 15, 18
9	*Onychargia atrocyana*		13
10	*Prodasineura autumnalis*		11, 15, 16, 18
11	*Prodasineura coerulescens*		15, 16
12	*Pseudocopera ciliata*		10
13	*Aciagrion borneense*	1	10, 12
14	*Aciagrion pallidum*	5	10, 18
15	*Aciagrion paludense*	1	
16	*Agriocnemis minima*	1, 6, 7	10, 12
17	*Agriocnemis nana*	7	13, 18
18	*Ceriagrion cerinorubellum*		10
19	*Ceriagrion indochinense*	1	
20	*Ceriagrion malaysei*	1	10
21	*Ceriagrion olivaceum*	1	
22	*Ischnura senegalensis*	3	12
23	*Paracercion calamorum*		12
24	*Pseudagrion australasiae*	1	10, 12
25	*Pseudagrion microcephalum*		18
26	*Pseudagrion pruinosum*		18
27	*Pseudagrion rubriceps*		12, 15–18
28	*Pseudagrion williamsoni*		12, 18
29	*Anax aurantiacus*		15
30	*Ictinogomphus decoratus*		15
31	*Acisoma panorpoides*	1	10, 12
32	*Brachydiplax farinosa*	1	12
33	*Brachydiplax sobrina*	1	
34	*Brachythemis contaminata*	9	10, 12, 15–17
35	*Crocothemis servilia*	6	10, 12, 17
36	*Diplacodes nebulosa*	1, 6, 8	10, 12
37	*Diplacodes trivialis*	1–3, 5, 6, 8, 9	10, 12, 17, 18
38	*Hydrobasileus croceus*		12
39	*Indothemis limbata*	1, 7	10, 12
40	*Lathrecista asiatica*		18
41	*Neurothemis fulvia*	5, 7–9	12, 18
42	*Neurothemis intermedia*		10, 12
43	*Neurothemis tullia*	1	10, 12, 18
44	*Orthetrum chrysis*	5	15, 17, 18
45	*Orthetrum pruinosum*		11, 17, 18
46	*Orthetrum sabina*	1, 3, 5–7, 9	10, 12, 14–18
47	*Pantala flavescens*		12, 14, 17, 18
48	*Potamarcha congener*	3, 8	
49	*Pseudothemis jorina*		15
50	*Rhodothemis rufa*		16
51	*Rhyothemis triangularis*	1	
52	*Rhyothemis variegata*	1	12
53	*Tholymis tillarga*		11, 18
54	*Trithemis aurora*	3, 5, 9	10, 12, 14–18
55	*Trithemis festiva*		17, 18
56	*Trithemis pallidinervis*	6	10, 12, 14, 15, 17
57	*Zyxomma petiolatum*		18

Fig. 7 Male *Euphaea inouei*, O'Chongheang River, 21 November 2022.

Fig. 8 Male *Rhinagrion hainanense*, O'Chongheang River, 23 November 2022.

Fig. 9 Female *Neurobasis chinensis*, stream near Beoung Nava, 17 November 2022.

Fig. 10 Male *Libellago lineata*, O'Khampa River, 19 November 2022.

Fig. 11 Male *Copera chantaburri*, O'Anchan stream, 15 November 2022.

Fig. 12 Male and female tandem pair of *Copera chantaburri*, O'Anchan stream, 15 November 2022.

10. *Prodasineura autumnalis* (Fraser, 1922) (Fig. 13)

The species was recorded in four locations. A single male was recorded on a stream near Beoung Nava on 17 November and the species was otherwise found to be common (≥20 individuals) on every visit to the O'Khampa and O'Chongheang Rivers.

11. *Prodasineura coerulescens* (Fraser, 1932) (Figs. 13 & 14)

Recorded at three locations and common (≥20 individuals) during all three visits to the O'Chongheang River. Also common at the O'Khampa river on the Sekong River side of the road bridge. Unusually, only one female was observed at the confluence of the O'Khampa River and the Sekong River on 16 November, although the species was probably overlooked due to difficulty accessing the site away from the boat.

12. *Pseudocopera ciliata* (Selys, 1863) (Fig. 15)

Four males and two females were seen and photographed at Beoung Nava on 17 November. These were found in a shaded marshy area at the western end of the site. This was the only location where this species was found.

Coenagrionidae

13. *Aciagrion borneense* Ris, 1911 (Fig. 16)

Recorded in three locations. Numbers at Trapeang Lumtea ranged from common (≥20 individuals) on the second visit and less (ten individuals) on the last visit. Usually seen in more open channels of water with intermittent vegetation. Males were mainly seen although some pairs in copulation were also noted. Up to three males were recorded at Beoung Nava and Beoung Khampa.

14. *Aciagrion pallidum* Selys, 1891

Recorded in three locations. A single male was photographed at the O'Anchan Stream on 15 November. Another male was recorded at Beoung Nava on 17 November and up to six males were seen in forest near the O'Chongheang River on 23 November. The latter observations were in the same habitat as *Vestalis gracalis*.

15. *Aciagrion paludense* Fraser, 1922 (Figs. 17–19)

Only recorded at Trapeang Lumtea. A single male with the black basal-pointing "chess pawn" variant marking on segment 8 was photographed on 12 November and single male was photographed on 26 November. The latter was initially identified as *A. occidentale* based on Laidlaw (1924), as it had the diagnostic black triangle on segment 8 with the apex pointing towards segment 7. The former was initially overlooked among photographs of the similar *A. borneense*. However, following review and consultations with O. Kosterin, the initial identifications were revised (a justification for this revision is provided in the Discussion). These are the first records of this species in Cambodia.

16. *Agriocnemis minima* Selys, 1877

Recorded at five sites and the most commonly recorded species in the genus. Common (≥20 individuals) on all four visits to Trapeang Lumtea, especially around its muddy margins. Individuals seen were mainly males with smaller numbers of females and orange teneral females. The species was also common at Trapeang Trau, Beoung Nava and Beoung Khampa, but was not recorded in riverine habitats.

17. *Agriocnemis nana* (Laidlaw, 1914)

Individuals were recorded at three locations. One female was photographed at the unnamed and overgrown trapeang on the track leading to Beoung Khampa on 12 November. Single males were also recorded at Trapeang Trau and the O'Chongheang River.

18. *Ceriagrion cerinorubellum* (Brauer, 1865)

Only recorded at Beoung Nava, where two males were recorded on 12 and 17 November.

19. *Ceriagrion indochinese* Asahina, 1967 (Fig. 20)

Only recorded at Trapeang Lumtea, but noted on all four visits. The maximum count was of six males and a single female was noted on the 26 November.

20. *Ceriagrion malaisei* Schmidt, 1964 (Fig. 21)

Recorded in two locations and seen on all four visits to Trapeang Lumtea. A single male was recorded at Beoung Nava on 12 November.

21. *Ceriagrion olivaceum* Laidlaw, 1914 (Fig. 22)

A single male was photographed at Trapeang Lumtea on 10 November. This was the only record during the study.

22. *Ischnura senegalensis* (Rambur, 1842)

This otherwise widespread and ubiquitous species was only recorded in two locations. These comprised a single male at Trapeang Lumpon on 10 November and two males at Beoung Khampa on 12 November.

23. *Paracercion calamorum* (Ris, 1916) (Fig. 23)

The only record during the survey was of a single male photographed at Beoung Khampa on 12 November.

24. *Pseudagrion australasiae* Selys, 1876

Recorded in three locations and on all four visits to Trapeang Lumtea with a maximum count of ten males. The species was common on both visits at Beoung Nava and Beoung Khampa. A maximum count of 20 was made at Beoung Nava on 12 November.

Fig. 13 *Prodasineura autumnalis*, *P. coerulescens* and *Psudagrion rubriceps* ovipositing in tandem pairs, O'Khampa River, 19 November 2022.

Fig. 14 Male *Prodasineura coerulescens*, O'Khampa River, 19 November 2022.

Fig. 15 Male *Pseudocopera ciliata*, Beoung Nava, 17 November 2022.

Fig. 16 Male *Aciagrion borneense*, Trapeang Lumtea, 26 November 2022. Note the broad black bar on segment 8 and thin black line on segment 9.

Fig. 17 *Aciagrion paludense*, Trapeang Lumtea, 26 November 2022. Note the black basal-pointing triangle on segment 8 which differs from *A. boneense* and *A. occidentale*.

Fig. 18 *Aciagrion paludense*, Trapeang Lumtea, 26 November 2022.

Fig. 19 *Aciagrion paludense*, Trapeang Lumtea, 12 November 2022. Note black basal-pointing "chess pawn" marking on segment 8.

Fig. 20 Male *Ceriagrion indochinese*, Trapeang Lumtea, 26 November 2022.

Fig. 21 Male *Ceriagrion malaisei*, Trapeang Lumtea, 11 November 2022.

Fig. 22 Male *Ceriagrion olivaceum*, Trapeang Lumtea, 10 November 2022.

Fig. 23 Male *Paracercion calamorum*, Beoung Khampa, 12 November 2022.

Fig. 24 Male *Pseudagrion williamsoni*, Beoung Khampa, 12 November 2022.

25. *Pseudagrion microcephalum* (Rambur,1842)

Only recorded at the O'Chongheang River where it completely replaced the similar *P. australasiae* and was common (≥20 individuals) on all three days.

26. *Pseudagrion pruinosum* (Burmeister, 1839)

Only recorded at the O'Chongheang River, where one male was observed on 22 November and eight were observed on 23 November.

27. *Pseudagrion rubriceps* Selys, 1876

Recorded in five locations. Observed and photographed in small numbers at all three locations on the O'Khampa River. A tandem pair were photographed ovipositing on the same floating stick as *Prodasineura autumnalis* and *P. coerulescens* (Fig. 13). A single male was recorded at Beoung Khampa on 19 November and the species was common (≥20 individuals) on all three days spent at the O'Chongheang River.

28. *Pseudagrion williamsoni* Fraser 1922 (Fig. 24)

Only recorded in two locations in semi-evergreen forest north of the O'Khampa River. Two and one individuals were recorded at Beoung Khampa on 12 and 17 November. Recorded on two out of three days at the O'Chongheang River with a maximum count of ten individuals.

Aeshnidae

29. *Anax aurantiacus* Makbun, Wongkamhaeng & Saetung Keetapithchayakul, 2022

Four males were seen patrolling at separate locations on the O'Khampa River at the confluence with the Sekong River on 16 November 2022. These were initially identified as *Anax immaculifrons* Rambur, 1842. The primarily orange-ground colour of the male abdomen is quite different to the colouration of individuals in the western part of its range, although this was previously considered a different colour form of the same species. However, Makbun *et al*. (2022) described individuals with orange abdomens in Cambodia, Laos, China, Hong Kong, Thailand and Vietnam as a new species (*A. aurantiacus*), based on colouration as well as morphological and molecular differences.

Gomphidae

30. *Ictinogomphus decoratus* (Selys, 1854)

Five males were seen (but not photographed) on the O'Khampa River at the confluence with the Sekong River on 16 November.

Libellulidae

31. *Acisoma panorpoides* Rambur, 1842

Recorded in three locations. Seen on three of the four visits to Trapeang Lumtea, with a maximum count of five individuals. Also common (≥20 individuals) on both visits to Beoung Nava and Beoung Khampa.

32. *Brachydiplax farinosa* Krüger, 1902 (Fig. 25)

Recorded in two locations. Seen on all four visits to Trapeang Lumtea with a maximum count of 12 males. Only one female was seen. Five males were recorded at Beoung Khampa on 12 November.

33. *Brachydiplax sobrina* (Rambur, 1842) (Fig. 26)

Only recorded at Trapeang Lumtea, where single males were seen on 14 and 26 November. This species is very similar to *Brachydiplax farinosa*, but has seven antenodal cross veins, whereas the latter has eight.

34. *Brachythemis contaminata* (Fabricius, 1793) (Fig. 27)

Recorded in eight locations. As many as ten individuals were recorded on Koh Dat Tum Island within the Sekong River. Recorded at all locations visited on the O'Khampa and O'Chongheang Rivers and also in the dry dipterocarp forest areas of Trapeang K'Dung and around the tented camp.

35. *Crocothemis servilia* (Drury, 1773)

Somewhat unusually, this widespread Asian species was only recorded in low numbers in four locations. Maximum counts of two males were registered on each visit to Beoung Nava, single males were observed at Beoung Khampa and the new bridge side of the O'Khampa River, and an immature male was observed at Trapeang K'Dung.

36. *Diplacodes nebulosa* (Fabricius, 1793)

Recorded in five locations. Common (≥20 individuals) on each visit to Trapeang Lumtea, Trapeang K'Dung, Beoung Nava and Beoung Khampa. Two individuals were recorded at Trapeang Buon Ch'rung on 13 November.

37. *Diplacodes trivialis* (Rambur, 1842)

Recorded in eleven locations. Commonly encountered but not formally recorded at numerous locations within dry dipterocarp forest. Common (≥20 individuals) at Trapeang Thymea were it was the only species recorded. Common on all four visits to Trapeang Lumtea, as well as Trapeang K'Dung and Trapeang Trau. Also common at the tented camp, Beoung Nava, Beoung Khampa and Koh Dat Tum. Smaller numbers were recorded on the

O'Anchan Stream, O'Chongheang River and at the new bridge side of the O'Khampa River.

38. *Hydrobasileus croceus* (Brauer, 1867)

The only record during the survey was of a single male which was observed hawking over the water at Beoung Khampa on 12 November.

39. *Indothemis limbata* (Selys, 1891)

Recorded in four locations and males mainly seen on all four visits to Trapeang Lumtea. A maximum count of ≈12 males was made on 11 November. The only female recorded was in copulation with a male and seen at the same location on 26 November. Common (≥20 individuals) on both visits to Beoung Khampa and common at Beoung Nava on 17 November, although only two were recorded there on 12 November. Two males were also observed at Trapeang Trau on 13 November.

40. *Lathrecista asiatica* (Fabricius, 1798)

The only records were of two immature males which were recorded on the O'Chongheang River on the 22 November.

41. *Neurothemis fulvia* (Drury, 1773)

Recorded in six locations and seen on all three visits to the O'Chongheang River, with a maximum count of ten individuals on 22 November. As many as six males were recorded at Beoung Khampa on 17 November. Three males were recorded at Trapeang Buon Ch'rung on 13 November and one female was observed at Trapeang Trau on the same day. Single males were also observed at the tented camp and O'Anchan Stream.

42. *Neurothemis intermedia* (Rambur, 1842) (Fig. 28)

Recorded in two locations. Six males were seen at Beoung Khampa on 12 November. An immature male was seen at Beoung Nava on 17 November.

43. *Neurothemis tullia* (Drury 1773)

Recorded in four locations and common (≥20 individuals) on all visits to Beoung Nava and Beoung Khampa. Two males were seen at Trapeang Lumtea on 14 and 26 November, whereas single males were recorded on the O'Chongheang River on 21 and 22 November.

44. *Orthetrum chrysis* (Selys, 1891)

Recorded in three locations. Recorded on each visit to the O'Chongheang River with a maximum count of four males on 22 November. A single male was seen on the O'Anchan Stream on 15 November. One male was recorded on the confluence of the O'Khampa and Sekong rivers and a female was noted further upstream on the new bridge side of the river.

45. *Orthetrum pruinosum* (Burmeister, 1839)

Recorded in three locations. As many as two were recorded daily at the O'Chongheang River from 21 to 23 November. One male was photographed at the stream near Beoung Nava on 17 November and two males were observed to the west of the new bridge on the O'Khampa River on 19 November.

46. *Orthetrum sabina* (Drury 1773)

The most widely recorded species during the survey, being registered at 13 of the 18 survey locations. Seen on three of four visits to Trapeang Lumtea, with a maximum count of approximately ten individuals on 14 November. Recorded as common (≥20 individuals) at Beoung Nava, Beoung Khampa, all three locations on the O'Khampa River, Koh Dat Tum and the O'Chongheang River. Smaller numbers were also recorded at Trapeang Lumpon, Trapeang Trau and Trapeang K'Dung, the tented camp and the O'Anchan Stream.

47. *Pantala flavescens* (Fabricius, 1798)

This highly nomadic and common species was surprisingly only recorded in four locations. As many as ten were observed on Koh Dat Tum island on 16 November. Two were recorded west of the new bridge on the O'Khampa River and singles were observed at Beoung Khampa and the O'Chongheang River.

48. *Potamarcha congener* (Rambur, 1842)

Recorded in two locations. Two males were recorded at Trapeang Buon Ch'rung on 13 November and a single male was recorded at Trapeang Lumpon on 10 November.

49. *Pseudothemis jorina* Förster, 1904

Only recorded at the confluence of the O'Khampa and Sekong rivers, where at least 20 males were observed.

50. *Rhodothemis rufa* (Rambur, 1842)

Only recorded on the Sekong River side of the old bridge on the O'Khampa River. A single immature male was seen but not photographed.

51. *Rhyothemis triangularis* Kirby, 1889

Only recorded at Trapeang Lumtea, where five males were registered on 11 November and two were registered on 26 November.

52. *Rhyothemis variegata* (Linnaeus, 1763)

Recorded in two locations. As many as ten individuals were noted at Beoung Nava on 12 November and a further eight were registered on 17 November. Single

Fig. 25 Male *Brachydiplax farinosa* with eight antenodal crossveins, 26 November 2022.

Fig. 26 Male *Brachydiplax sobrina* with seven antenodal crossveins, 26 November 2022.

Fig. 27 Male *Brachythemis contaminata*, Koh Dat Tum, 16 November 2022.

Fig. 28 Male *Neurothemis intermedia*, Beoung Khampa, 12 November 2022.

Fig. 29 *Trithemis festiva*, O'Chongheang River, 21 November 2022.

females were recorded at Trapeang Lumtea on 11 and 26 November.

53. *Tholymis tillarga* (Fabricius, 1798)

This crepuscular species was recorded at two locations. One male was seen briefly hanging on bamboo at the stream near Beoung Khampa on 17 November. Six and ten individuals were recorded on the O'Chongheang River on 21 and 22 November. The latter sightings were made while camping in the forest whereas the single male was flushed from a perch in the middle of the day. This species may be found to be widespread if suitable areas are searched at dusk or roosting sites are checked.

54. *Trithemis aurora* (Burmeister, 1839)

Recorded in ten locations. This widespread Asian species was common (≥20 individuals) on Koh Dat Tum Island

and all locations along the O'khampa River. It was common during all visits to Beoung Nava and Beoung Khampa, as well as on each visit to the O'Chongheang River where *Trithemis festiva* also occurred. So much so, that close inspection of females and subsequent identification were not undertaken as this reduced the time available for searching for other species. As many as four individuals were recorded at the tented camp on 10 November, and singles were recorded at Trapeang Lumpon and the O'Anchan Stream.

55. *Trithemis festiva* (Rambur, 1842) (Fig. 29)

Recorded in two locations. Common (≥20 individuals) on all three visits to the O'Chonheang River. Similar to *Trithemis aurora*, close inspection of females and subsequent identification was not undertaken as this reduced the time available for searching for other species. Four males were recorded west of the new bridge on the O'Khampa River on 19 November.

56. *Trithemis pallidinervis* (Kirby, 1889)

Recorded in six locations. Common (≥20 individuals) at Beoung Nava and Beoung Khampa on 12 and 17 November, and at the confluence of the O'Chongheang and Sekong rivers on 16 November. Additional counts include six at Koh Dat Tum Island on 16 November, two westwards of the new bridge on the O'Khampa River on 19 November and one male at Trapeang K'Dung on 13 November.

57. *Zyxomma petiolatum* Rambur, 1842

This crepuscular/nocturnal species was only recorded at the O'Chongheang River, with eight individuals registered at dusk on 21 November and two on 22 November. As with *Tholymis tillarga*, these observations were made as we camped in the forest. This species will likely be found to occur more widely if suitable habitats are searched at dusk.

Discussion

Fifty-seven dragonfly species were recorded during the survey. This figure is comparable to the Prey Long Wildlife Sanctuary (Kosterin, 2020) where 60 species were recorded over two visits (June 2018 & December 2019), although Siem Pang Wildlife Sanctuary was only visited in November. An additional visit in May/June during the wet season would likely increase the number of species recorded. It is also notable that the only gomphid species recorded was *Ictinogomphus decoratus*, which is predominantly a lentic species. As most gomphids are lotic species inhabiting fast flowing water, they are more prevalent during the wet season.

It is not possible to draw any conclusions about the importance of trapeangs within the wildlife sanctuary for dragonflies other than they generally held common wetland species in low numbers during the survey period. With 27 species registered, the O'Chongheang River supported the most species, whereas among the trapeangs, the most visited site (Trapeang Lumtea) supported the highest number of species with 17 taxa. Beoung Khampa and Beoung Nava both supported more species than Trapeang Lumtea, with 23 and 18 species respectively, and were visited only twice (as opposed to the latter which was visited four times). I suspect more visits to Beoung Nava and Beoung Khampa would have yielded more species. I also received unpublished information from Jeremy Holden that he observed *Rhyothemis plutonia* (Selys, 1883) (photographic evidence) and *R. phylis* (Sulzer, 1776) earlier in 2022 at Beoung Nava. The report of *R. plutonia* suggested there were up to 12 individuals flying low above the reeds at 10:00 hrs but they had all gone by midday. Given that my earliest arrival at Beoung Nava was at 11:20 hrs, the species was either missed or not present in November.

Over the course of the survey, 50 dragonfly species were recorded north of and including the O'Khampa River whereas 32 species were registered in the area south of the O'Khampa River. The former was likely due to the wider diversity of habitats in the north of the sanctuary. Most of the species recorded were expected dragonflies which are ubiquitous in open wetland habitats in the region. However, there were notable exceptions including the second country record for *Copera chantaburri* Asahina, 1984 (Figs. 11 & 12). The most significant records were arguably those for *Aciagrion paludense* (Fraser, 1922) (Figs. 17–19) at Trapeang Lumtea on 12 and 26 November which was initially identified as *A. occidentale* (Laidlaw, 1924). This incorrect identification and subsequent re-identification is considered below due to the complexity of taxonomic issues regarding this species and indeed the wider *Aciagrion* genus. Other interesting records include *Euphaea inouei* Asahina, 1997 (Fig. 7) which was observed on the O'Chongheang River from 21 to 23 November, a species whose taxonomic status also varies in the literature. Its taxonomy is therefore also considered below.

Aciagrion occidentale (Laidlaw, 1924) & *Aciagrion paludense* (Fraser, 1922).

Aciagrion occidentale was originally described as *Aciagrion hisopa* (Selys)? race *occidentalis* from three specimens (two males & one female) collected in India in October 1916 (Laidlaw 1919). In his description Laidlaw stated "The two males that I have seen are, however, characterized by having a black triangle on the dorsum of the eighth abdominal segment, with its apex directed towards the hinder end of the segment". However, referring to specimens from Cochin (now Kochi) and Ceylon (now Sri Lanka) in his later review of the genus, he stated "My description states that the black mark on segment eight of the abdomen has its apex directed toward the hinder end of segment. This should read "directed towards the base of the segment". This was supported by a drawing showing the apex pointing towards the base (Laidlaw, 1924). Laidlaw's correction appeared to close issues related to segment 8, but further complications later arose when an individual matching his original description was found.

Joshi & Kunte (2014) stated "In Intanki NP on 24 May 2013 SJ recorded an *Aciagrion* male with S7 having a black triangle directed to the end of the segment (Image 5), as in the original description of *occidentale* by Laidlaw (1919). This fact made us suppose that this description was correct and the later correction by Ladilaw [sic] (1924) was based on other specimens, with the opposite direction of the triangle.". A photo of this individual is shown on p. 6468 of Joshi & Kunte (2014) which matches the original species described by Laidlaw (1919). This should now be considered the true description of *A. occidentale*.

It should be noted that *A. occidentale* was reported from Cambodia by Asahina (1967). However, Kosterin (2010) stated "There is a problematic species reported by Asahina (1967), *A. occidentale*, which is rare in Indochina, while the very similar *Aciagrion borneense* Ris, 1911 is common in dry season. In those early years, Asahina could still confuse them. Noteworthy that in his later paper (Asahina, 1982) devoted to Coenagrionidae of Thailand, most specimens of *A. borneense* were collected from November to January, the scarce *A. occidentale* in July and August. The specimens reported in Asahina (1967) were collected, at Phnom Penh and Bokor, on 15 XI and 2 XII, respectively. (Meanwhile, the conspecifity of Indochinese specimens referred to as *A. occidentale* and the true Indian A. occidentale was doubted by Hamalainen, 2001). Therefore, among the Cambodian Odonata we should list *A. occidentale* with caution.". However, Oleg Kosterin has since shared photographs taken by Stephane De Greef in Siem Reap Province (Fig. 30) of an individual *Aciagrion* which shows the segment 8 pattern of the "true Indian *A. occidentale*" of Laidlaw's (1919) original description. This proves that genuine *A. occidentale* does occur in Cambodia.

In discussing Laidlaw's (1924) revised description of *A. occidentale*, Joshi & Kunte (2014) suggested that "The pattern of S7 and S10 corresponds to *Aciagrion paludense* Fraser, 1922, considered by a junior synonym of *A. occidentale* (Laidlaw, 1924; Fraser, 1933).". This required further exploration as Fraser (1922) actually stated "Segment 8 all blue save with a black mark shaped like a chess pawn with its apex pointing basal.". The black mark on segment 8 of the individual in Fig. 17 and Fig. 18 appears to be triangular and not "chess pawn" shaped, but closer examination shows the edges do not form a neat triangle. However, the individual in Fig. 19 has a marking on segment 8 which is intermediate between the triangle of the individual in Fig. 17 and Fig. 18 and the classic *Aciagrion borneense* in Fig. 16. In fact, the marking on segment 8 of the *A. paludense* in Fig. 19 is more like the "Chess pawn with apex pointing basal" originally described by Fraser (1922) as *A. paludensis* (*paludense*).

Fraser (1933) later changed his description of segment 8 of *A. occidentale* (which he assumed was a senior synonym of *paludense*) to "segment 8 with a narrow dorsal triangle of black, the base of the triangle on apical border of the segment, its point extending variably nearly quite up to base of segment.". This description better matches the segment 8 pattern of the *A. paludense* in Fig. 17 and Fig. 18.

Photographs of the original lectotype and paralectotype of Fraser's (1922) description of male *A. paludense* (Figs. 31–34) were obtained from the Natural History Museum, London. The lectotype (Fig. 31) clearly shows the basal-pointing "chess pawn" marking on segment 8 which correlates with the *A. paludense* photographed by the present study on 12 November 2022 (Fig. 19). The photograph of the paralectotype (Fig. 34) is inconclusive, as this is not so well preserved. The two individual males of *A. paludense* photographed in this study show variation of the marking on segment 8 with the individual recorded on the 26 November 2022 (Figs. 17 & 18) having a more triangular black marking. From this point of view, both of Fraser's descriptions (1922, 1933) are correct and the pattern on segment 8 is clearly variable. The photographs of the head and thorax of the lectotype (Fig. 32) appear identical to the individuals from Siem Pang Wildlife Sanctuary.

It should be noted that Joshi & Kunte (2014) referred to Fraser's original name *A. paludense*, correcting both his 1922 and 1933 publications where he referred to it as *A. paludensis*. Although not stated by Joshi & Kunte (2014), they fulfilled the change of spelling needed to correspond with the neutral gender of the generic name, as required by article 34.2 of the International Commission of Zoological Nomenclature.

Fig. 30 *Aciagrion occidentale* (Laidlaw, 1919), Siem Reap, 2022 (© S. De Greef).

Fig. 31 Dorsal and lateral views of the abdomen of *Aciagrion paludense*, lectotype (male).

Fig. 32 Dorso-lateral views of the head and thorax of *Aciagrion paludense*, lectotype (male).

Given these issues, I conclude that two species exist within the *A. occidentale* conundrum proposed by Joshi & Kunte (2014), namely the genuine *A. occidentale* (Laidlaw, 1919) (as shown in Fig. 30) and *A. paludense* (Fraser, 1922 & 1933) (as shown in Figs. 17–19). As a consequence, I propose that *A. paludense* be recognised as a species in its own right, rather than as a junior synonym of *A. occidentale*. An individual identical to the specimen in Fig. 17 & 18 has been photographed in Vietnam (Kompier, 2022). While this was identified by the author as *A. occidentale*, it should be regarded as *A. paludense*. The two records of *A. paludense* at Trapeang Lumtea on 12 and 26 November (Figs. 17–19) constitute the first records for Cambodia.

Given the foregoing, it is not surprising that Hämäläinen (2001) opined "It is hoped that someone will undertake a review of the whole genus, which is undoubtedly one of the most difficult and poorest known among the oriental damselflies."

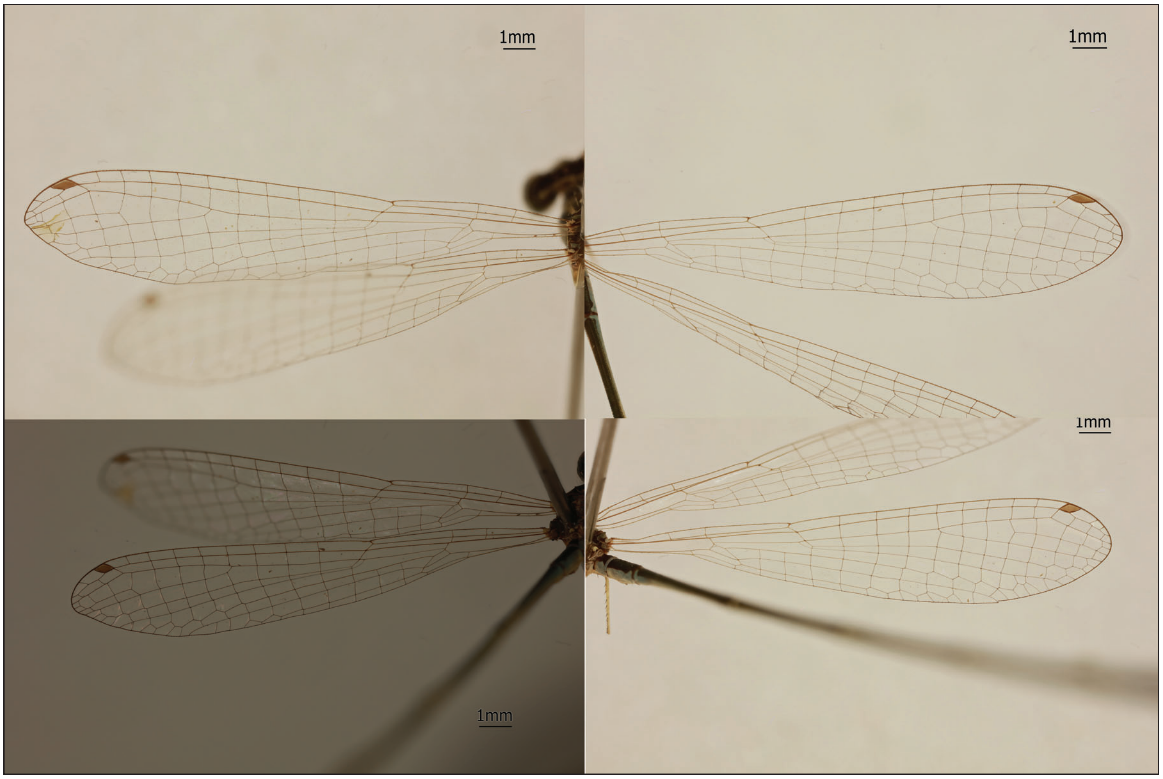

Fig. 33 Wings of *Aciagrion paludense*, lectotype (male).

Fig. 34 Dorsal view of *Aciagrion paludense*, paralectotype (male).

Euphaea inouei Asahina,1997

The record of *Euphaea inouei* (Fig. 7) along the O'Chongheang River requires further discussion. Species within the genus Euphaea are challenging to identify due to the number of similar species. The individuals observed during the survey were initially identified as *E. masoni*, but following discussion with Oleg Kosterin, reassigned to *E. inouei* on the basis of having largely dark wings with coppery iridescence when opened. The specific status of *E. inouei* is not without controversy. Kosterin (2016) treated *inouei* as a subspecies of *E. masoni* and detailed how *masoni* and *inouei* were considered subspecies of *E. guerini* by Rambur (1842) and Asahina (1977), although Kosterin also stated "this did not hold much water (van Tol & Rosendaal, 1995; Hämäläinen & Karube, 2001).". Conversely, Phan *et al*. (2018) discussed how some early authorities, notably Selys Longchamps (1879) and Martin (1904), treated *E. guerini* and *E. masoni* as distinct species. Further, Asahina (1977) and others failed to identify structural differences between *E. guerini* and *E. masoni.* These include the lack of a ventral tuft of bristles on segment 9, with the upperside of the hindwing of *E. guerini* also possessing a green lustre (Phan *et al*., 2018).

Phan *et al*. (2018) did not attempt to separate specimens from Vietnam into the taxa *masoni* and *inouei*, but listed all of them as *masoni*. They did state "However, the wing upperside of males from Vietnam (and eastern Cambodia) show a strong iridescent coppery-red flash in sunshine and the HW underside, except for its distal part, shows a slight deep-blue flash. At the same time males from Thailand and south-western Cambodia of the typical *Euphaea masoni*, described from the border between Myanmar and Thailand, show only a very slight purple shine on the wing upperside and no flash on the wing underside (Kosterin, 2014, 2016). These differences indicate the possibility that the Vietnamese populations concern a different taxon from *Euphaea masoni* sensu stricto". Phan *et al*. (2018) also state "The genital ligula and anal appendages of *inouei* are identical to those of *masoni*. It should be noted here that recently Hämäläinen (2016: 25, Note 39) listed *Euphaea inouei* as a good species based on morphological and unpublished molecular evidence.". Additionally, Paulson *et al*. (2023) also listed *E. inouei* as a full species. As a consequence of these points, the individuals recorded along the O'Chongheang River are attributed to *E. inouei*.

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The use of bear parts as traditional Khmer medicine in Cambodia

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មូលន័យសង្ខេប

សត្វខ្លាឃ្មុំធំ *Ursus thibetanus* និងសត្វខ្លាឃ្មុំតូច *Helarctos malayanus* មានដើមកំណើតនៅព្រះរាជាណាចក្រកម្ពុជា និងនៅតំបន់អាស៊ីអាគ្នេយ៍។ ការជួញដូរខុសច្បាប់ និងការប្រើប្រាស់បំណែកសរីរាង្គសត្វខ្លាឃ្មុំធ្វើឱសថបុរាណជាបញ្ហាប្រឈមដ៏ចម្បងមួយនៅក្នុងការអភិរក្សប្រភេទសត្វខ្លាឃ្មុំទាំងនោះ ដោយបង្កឱ្យមានកំណើននៃការបរបាញ់សត្វខ្លាឃ្មុំព្រៃកាន់តែខ្លាំង និងជំរុញឱ្យមានការបង្កើតកសិដ្ឋានចិញ្ចឹមសត្វខ្លាឃ្មុំនៅប្រទេសមួយចំនួនដើម្បីផ្គត់ផ្គង់តម្រូវការឱសថបុរាណ។ ការយល់ដឹងអំពីប្រភេទអ្នកប្រើប្រាស់ និងការប្រើប្រាស់បំណែកសរីរាង្គសត្វខ្លាឃ្មុំធ្វើឱសថបុរាណ មានសារៈសំខាន់សម្រាប់ការរៀបចំយុទ្ធសាស្ត្រអភិរក្សដ៏មានប្រសិទ្ធភាពដោយផ្តោតទៅលើកត្តាដែលជំរុញឱ្យមានការប្រើប្រាស់បំណែកសរីរាង្គសត្វខ្លាឃ្មុំធ្វើឱសថ។ បំណែកសរីរាង្គរបស់សត្វខ្លាឃ្មុំត្រូវបានប្រើប្រាស់ក្នុងវិស័យវេជ្ជសាស្ត្របុរាណខ្មែរសម្រាប់ធ្វើជាឱសថព្យាបាលជំងឺ និងជំនឿអរូបិយ។ យើងបានធ្វើការដកស្រង់ផ្នែកខ្លះនៃទិន្នន័យពីការសិក្សារបស់ Lim *et al.* (2022) ដើម្បីស្វែងយល់ថាតើការប្រើប្រាស់បំណែកសរីរាង្គសត្វខ្លាឃ្មុំកំពុងកើនឡើងនៅក្នុងវិស័យវេជ្ជសាស្ត្របុរាណខ្មែរដែរឬទេ ហើយតើវាជាទំនោរនៃការប្រើប្រាស់ដែលរងឥទ្ធិពលពីខាងក្រៅក្នុងអំលុងពេលប៉ុន្មានឆ្នាំចុងក្រោយនេះ ឬមាននៅក្នុងប្រវត្តិសាស្ត្ររបស់វេជ្ជសាស្ត្របុរាណខ្មែរយូរលង់មកហើយ។ យើងបានធ្វើការសម្ភាសន៍អ្នកប្រកបរបរវេជ្ជសាស្ត្របុរាណខ្មែរចំនួន៣៣នាក់ នៅទីជនបទក្នុងខេត្តស្ទឹងត្រែង និងខេត្តមណ្ឌលគិរី ព្រមទាំងនៅមជ្ឈមណ្ឌលជាតិស្រាវជ្រាវវេជ្ជសាស្ត្របុរាណ និងសមាគមន៍គ្រូឱសថបុរាណខ្មែរនៅទីក្រុងភ្នំពេញដោយប្រើកម្រងសំនួរ semi-structured interviews។ លទ្ធផលនៃការសិក្សានេះបង្ហាញថា ប្រមាត់ខ្លាឃ្មុំត្រូវបានគេយល់ថាជាឱសថដ៏មានតម្លៃ និងគុណភាពខ្ពស់ ហើយអ្នកប្រកបរបរវេជ្ជសាស្ត្របុរាណខ្មែរបាននិយាយថាមនុស្សនៅតែត្រូវការវា។ ទោះជាយ៉ាងណាក៏ដោយ ការឱ្យប្រើបំណែកសរីរាង្គសត្វខ្លាឃ្មុំសម្រាប់ព្យាបាលជំងឺមានការថយចុះអំលុងពេលប៉ុន្មានឆ្នាំចុងក្រោយនេះ ហើយអ្នកប្រកបរបរវេជ្ជសាស្ត្របុរាណខ្មែរគិតថាការថយចុះចំនួនសត្វខ្លាឃ្មុំព្រៃនៅកម្ពុជាដោយសារការបាត់បង់ទីជម្រក។ អ្នកប្រកបរបរវេជ្ជសាស្ត្របុរាណខ្មែរមិនសូវឱ្យបំណែកសរីរាង្គសត្វខ្លាឃ្មុំទៅអ្នកជំងឺទេ ប៉ុន្តែពួកគាត់ណែនាំឱ្យអ្នកត្រូវការប្រើប្រាស់រកបំណែកសរីរាង្គសត្វខ្លាឃ្មុំដោយខ្លួនឯង។ អង្គការអភិរក្សដែលធ្វើការលើការកាត់បន្ថយតម្រូវការនៃការប្រើប្រាស់ទឹកប្រមាត់ខ្លាឃ្មុំគួរសហការជាមួយមជ្ឈមណ្ឌលជាតិស្រាវជ្រាវវេជ្ជសាស្ត្របុរាណ សមាគមន៍គ្រូឱសថបុរាណខ្មែរ និងអ្នកប្រកបរបរឱសថបុរាណខ្មែរ ដើម្បីលើកកម្ពស់ឱ្យមានជម្រើសប្រើប្រាស់ឱសថផ្សេងទៀតប្រកបដោយនិរន្តរភាពដូចជាឱសថរុក្ខជាតិ។ នេះគឺជាយុទ្ធសាស្ត្រដ៏សំខាន់សម្រាប់អនុវត្តនៅតំបន់ជនបទដែលការប្រើប្រាស់ឱសថបុរាណខ្មែរនៅមានអត្រាខ្ពស់នៅឡើយ។

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Abstract

Asiatic black bears *Ursus thibetanus* and sun bears *Helarctos malayanus* are native to Cambodia and Southeast Asia. Illegal trade and use of their body parts for traditional medicine are among the main conservation challenges facing these species, because they increase hunting pressure on wild populations and motivate bear farming in certain countries to supply demand for traditional medicine. Understanding the types of consumers and use of bear parts as traditional medicine is crucial to design effective conservation strategies that target the motivations of users of bear part medicine. Bear parts have been used in traditional Khmer medicine (TKM), both as medicinal remedies and in spiritual practices. We explored a subset of data from Lim *et al*. (2022) to understand whether the use of bear parts is growing in TKM and if it has been influenced by external factors in recent years or whether it has occurred throughout the history of TKM. We conducted semi-structured interviews with 33 TKM practitioners in rural areas in the Stung Treng and Mondulkiri provinces, at the National Center for Traditional Medicine and in TKM associations in Phnom Penh, Cambodia. Our results suggest bear gallbladders are perceived as valuable and high-quality medicine and practitioners stated that people still demand it. However, prescriptions have decreased in recent years and practitioners attribute this to declines in bear populations in Cambodia due to habitat loss. Practitioners are less likely to provide bear parts to patients, but may influence demand by advising them to seek these out. Conservation organisations seeking to reduce demand for bear bile should engage with the National Center for Traditional Medicine and other TKM associations and practitioners to promote sustainable alternatives such as plant-based medicines. This is a particularly important strategy to apply in rural areas, where use of TKM appears to be higher.

Keywords *Helarctos malayanus*, practitioners, semi-structured interview, sustainable use, *Ursus thibetanus,* wildlife trade.

Introduction

Throughout human history, people have used traditional medicine derived from natural resources such as plants, animals and minerals (Alves & Rosa, 2005). The World Health Organization reports that 80% of people in developing countries still rely on animal- and plant-based medicine for primary health care, especially where natural resources are part of their livelihoods (WHO & MOH, 2012). The use of wild animal parts and derived products (e.g., skins, bones, tusks, bile & feathers) for traditional medicine is one of the major threats driving the loss of biodiversity worldwide (Alves & Rosa, 2013; Ogada *et al*., 2016; Alves *et al*., 2018). For instance, traditional Chinese medicine (TCM) uses over 1,500 animal species and is traded worldwide, whereas 15–20% of Ayurvedic medicine (India) is based on animal-derived substances (Alves & Rosa, 2005). Alves & Rosa (2013) found that body parts of 108 species of carnivores have been exploited for use in traditional medicine worldwide and nearly half of species listed on the *IUCN Red List* as Near Threatened to Critically Endangered are used as medicine or food.

Bears (Ursidae) are one family of wildlife that are threatened by illegal trade and use for medicine. All bear species are listed in Appendix I or II of the *Convention on International Trade in Endangered Species of Wild Fauna and Flora*, which regulates trade in live bears and bear parts and products (MAFF, 2002; CITES, 2021). Bear parts are in high demand for traditional medicine worldwide but particularly in Asia (e.g., Davis *et al*., 2019a, b, 2020a–c; Davis & Glikman, 2020; Hinsley *et al*., 2021; Davis *et al*., 2022). Demand for bears is a serious threat to the conservation of Asian bear species because it increases hunting pressure on wild populations and motivates farming of bear bile to supply the demand (Alves & Rosa, 2013; Livingstone & Shepherd, 2016; WWF, 2016; Crudge *et al*., 2019).

In this study, we focus on two bear species native to Southeast Asia which are threatened by widespread trade in their parts for traditional medicine: Asiatic black bears *Ursus thibetanus* and sun bears *Helarctos malayanus* (Burgess *et al*., 2014). Populations of both species have declined by ≈30% over the last 30 years (Scotson *et al*., 2017; Garshelis & Steinmetz, 2020). Cambodia may hold regionally significant populations of bears (Gray *et al*., 2017a), but these are threatened by high levels of trade and known consumption of their parts (Burgess *et al*., 2014; Davis *et al*., 2019a, b, 2020a–c). Although national legislation exists to protect wildlife in Cambodia, these laws are poorly enforced in most areas and illegal hunting—particularly the use of snares—remains a significant threat to all large fauna (Gray *et al*., 2017b, 2021; Marx *et al*., 2020). In the Cardamom Rainforest Landscape of southwestern Cambodia, the largest mammalian predators, tigers *Panthera tigris* and leopards *P. pardus*, are likely to be now extinct, with dhole *Cuon alpinus* and bears the largest carnivores remaining (Gray *et al*., 2017a). Since 1997, over 200 Asiatic black bears and sun bears have been rescued from illegal wildlife trade in Cambodia (Free the Bears, unpubl. data), indicating that the hunting and trade of bears is a considerable threat. Marx *et al*. (2020) reported that two of three released sun bears in a protected area were killed by snaring. Bear parts, in particular the gallbladder, are highly sought-after for use in traditional medicine (TM) in Cambodia and throughout the region (Burgess *et al*., 2014).

Bear gallbladder has been used in TCM for over 2,000 years to treat a variety of illnesses (Feng *et al*., 2009). Recent research in Cambodia has documented widespread use of bear bile or gallbladder as medicine to treat ailments ranging from bruises to menstrual bleeding (Davis *et al*., 2019a, 2020b, 2020c). These findings are concerning from a conservation standpoint, as the practice poses a threat to wild bear populations. However, little is known about traditional practices of using bear bile in Cambodia and specifically, whether these are an inherent component of traditional Khmer medicine (TKM), or a more recent addition due to the influence of TCM and/or other medicinal systems, such as traditional Vietnamese medicine, which also prescribe bear bile (Feng *et al*., 2009; Davis *et al*., 2019b).

In Cambodia, TKM is still widely used, possibly due to long-standing acceptance in Khmer culture and poor access to western medicines (Richman *et al*., 2010). Ros *et al*. (2018) estimated that 40–50% of rural populations in Cambodia still depend on TM for primary health care, although this use may be more "informal" than in previous decades e.g., individuals are less likely to actively consult a TM practitioner (Meessen *et al*., 2011; Lim *et al*., 2022). Like some other TMs, TKM ingredients are derived from plants, animals and also minerals (Hieng *et al*., 2011). Wildlife species cited in the Khmer medicine pharmacopeia by Hieng *et al*. (2011) include mammals, reptiles, birds, aquatic animals and insects, along with specific organs used e.g., rhino horn, tiger bone, bear gallbladder, porcupine stomach or pangolin scale.

Lim *et al*. (2022) showed that animal products are frequently used in contemporary practices of TKM. They also found that loris species *Nycticebus* sp. were most commonly reported in TKM (49% of respondents), followed by serow *Capricornis sumatraensis* (39%) and porcupine (Hystricidae spp., 27%). Bear parts were the fifth most commonly reported wildlife parts used by respondents (15.2%) in the five years prior to the research, yet were also reported as the most commonly prescribed parts (36.4%) prior to this (i.e. more than five years previously) (Lim *et al*., 2022).

The aim of our study was to explore the use of bear parts in TKM using a subset of data collected by Lim *et al*. (2022) that focuses on this topic. We also aimed to investigate the history of bear part use in TKM. This research is particularly relevant for behavioural interventions that aim to reduce the use of bear bile and bear gallbladders in rural areas of Cambodia as well as possible alternatives that might be acceptable to communities in these areas.

Methods

Semi-structured interviews

We conducted semi-structured interviews with TKM practitioners in rural areas of Cambodia where we assumed based on the number of health centres that people have limited access to western medicine and are more reliant on TM. We also interviewed teachers of TKM at the National Center of Traditional Medicine and TKM associations. The interview was divided into subsections intended to investigate: TM practice in general (e.g., what type of schooling practitioners may have, where their knowledge came from, etc.); use of wildlife (animal and plant) products in TKM; specific applications of bear-derived medicinal products (e.g., which ailments are treated, how the products are used); and how TKM has changed over the course of the practitioners' lives. The interview can be found as an appendix to Lim *et al*. (2022). The study was carried out between June 2018 and January 2019. NVivo (vers. 12) was used to analyse the data in terms of visualizing, classifying, sorting and arranging the data into themes.

Study areas

Our study was undertaken in Stung Treng Province (13°31′ N, 105°57′ E) and Mondulkiri Province (12°27′ N, 107°14′ E) in northeast and east Cambodia, respectively. With a total area of 11,092 km^2, Stung Treng is the least densely populated province in Cambodia and abuts the border with Laos (NIS, 2013a). Following Khmer, the most prevalent ethnicity in the province is the Lao people (Try & Chambers, 2006). Mondulkiri, which borders Vietnam to the east, has a total area 14,288 km^2 and is one of the largest provinces in Cambodia. The most common indigenous group are the Bunong people (NIS, 2013b). These provinces were selected for the study because they have large areas of forest with rich biodiversity and are less developed with fewer medical clinics where people can access western medicine (Open Development Cambodia, 2016).

Sampling strategy

We interviewed TKM practitioners who comprised traditional Khmer healers (*Kru Khmer*), traditional birth attendants / grandmother midwives (*Chhmob boran* or *Yeiy mop*) and Buddhist monks—who practice TKM in addition to performing exorcism ceremonies and spiritual healing based on Buddhist principles (Ovesen & Trankell, 2010). We also interviewed key informants from the National Center for Traditional Medicine, the Cambodian Traditional Healer Association and the Association of Traditional Cambodian Medicine in Phnom Penh. As we had no prior information about the TKM practitioners in our study areas, convenience and snowball sampling were employed (Etikan *et al*., 2016; Parker *et al*., 2020). As such, the first village in each study area was randomly selected and on arrival there, we asked village chiefs for the location of TKM practitioners. After we interviewed a few TKM practitioners in each village, we used snowball sampling to find other practitioners, although we found that TKM practitioners did not like to introduce other practitioners because they were direct competitors for clients.

Ethical approval

This study was approved by the relevant agencies in study areas, as well as the Cambodian Ministry of Environment. Ethical approval was granted by the Miami University of Ohio's Internal Review Board (Protocol ID: 02106e). All respondents were informed of their confidentiality and told they could stop the interview at any time. All interviews were conducted by the lead author and an assistant note taker who directly transcribed the interview as it occurred in Khmer (national language of Cambodia). Once transcribed, all respondents were assigned a random ID and to ensure their confidentiality, the physical interview documents were kept in a sealed cabinet in a locked office in Phnom Penh, which only the study authors had access to. For additional details of the study methods, please refer to Lim *et al*. (2022).

Results

Demographic information

We interviewed a total of 35 people who comprised TKM practitioners in Stung Treng (*n*=20) and Mondulkiri (*n*=10) and key informants (*n*=5) in Phnom Penh. Of the five key informants, two were institutional management staff and three were TM teachers and researchers who actively practice TKM. As such, data provided by the latter were pooled with other TKM practitioners in the relevant analyses. Overall, we interviewed 25 men and 10 women, with an average age of 62 years. All of the participants still practiced TM and had 25 years of experience on average, with only 12% (n=4/33) of interviewees having practiced for less than five years. On average, our respondents met 31 people seeking treatment or purchasing TM each month. The ethnicities of our respondents were: Khmer (*n*=20/33), Khmer-Lao (*n*=8/33), Lao (*n*=3/33), Bunong (*n*=1/33) and Khmer-Chinese (*n*=1/33). Further details are provided by Lim *et al*. (2022).

Use of bear parts in TKM

Nearly all respondents including TKM teachers at the National Center for Traditional Medicine and Association of Traditional Cambodian Medicine (*n*=32/33) reported that bear parts were historically used in TKM, although the uses varied depending on the parts employed. When asked which bear parts can be used in TKM and what ailments these may be used to treat, gallbladder was most commonly identified by respondents (*n*=31/33), followed by liquid bile (*n*=29/33). Both were reported as valuable medicine and identified as the most sought-after bear parts, with four respondents comparing the price to gold.

"*Oh, not easy to find bears, but if we can get one bear we will become the rich person, will be "tilt the village" [idiom] because the bear's gall bladder is expensive like the gold, they use the gold scale to weight it and sell...*" [Female, 41, Khmer-Lao, TKM healer, Stung Treng]

"*Yes, bear bile and gallbladder are TKM too, it is the high-quality medicine because it is as expensive as gold. My brother who lives at Koh Kong province, used to be the middleman who find to buy bear from hunters since 1990s...*" [Key informant #3, Phnom Penh]

Bear gallbladder was reported to have the same medicinal effect as liquid bile and as used much more than the latter. Although well known to practitioners, liquid bear bile was reported as being infrequently used in practice. Gallbladders used for medicine were generally reported to be sun-dried or roasted. The dry bear gallbladder was said to be chopped into small slices, soaked in local wine, or rubbed with water (*Thnam dos*). Bear gallbladder and/or bile were frequently prescribed for aliments including body pain (*n*=6), as a tonic (*n*=5), internal/external bruising (*n*=4), diabetes (*n*=3), cool medicine & fever (*n*=2), improving vision (*n*=2) and post-partum treatment (*n*=2). Bear gallbladder was also reported by at least one respondent as used to treat various other illnesses including chicken pox, HIV, kidney disease, heart disease, gallbladder disease, cancer, lung and liver disease, headache, haemorrhoids and insomnia (Table 1).

In descending order, other bear parts valued for TKM included the claws (*n*=30/33), teeth (*n*=26/33), bone (*n*=26/33), blood (*n*=23/33), paw (*n*=22/33) and skin (*n*=16/33) (Table 1). Respondents reported that bear claws are frequently used in spiritual treatments as a necklace pendant (*n*=11). The purpose was for a child or newborn baby to wear these to protect them from spirits called "Old mother" (*Preay Madai Derm*) who wake babies up and make them cry at night. Bear claw pendants were also used by older people to bring luck and power, and as "rubbed medicine" for fever (*n*=5) and body pain (*n*=2). Teeth were also used in a similar manner as claws in being worn as a necklace for protection against bad luck and danger (*n*=10), and employed as rubbed medicine for cooling and treatment of fevers (*n*=2). Bear bone was soaked in rice wine or ground on stone and drunk in rice wine or water to treat body pain (*n*=4), or applied on the skin as a healing agent for wounds and dermatoses (*n*=4), a cooling medicine for fever (*n*=3) and for internal/external bruising (*n*=2). People also drank bear blood mixed with rice wine to treat body pain (*n*=8) and internal/external bruising (*n*=4), as a general tonic (*n*=4) and for women to get fair skin (*n*=2). Paws were roasted or soaked in rice wine or made into a soup used for body pain (*n*=7) and as a tonic (*n*=5). They were also reported by at least one respondent as being used to improve sexual ability, to treat myelodysplastic syndromes and fever, or used as an antibiotic or healing agent. Skin was roasted and powdered to use for healing agent for dermatoses (*n*=5) and when soaked in wine, used for respiratory illnesses such as asthma and tuberculosis (*n*=2).

Sourcing bear parts

Similar to other wildlife used in TKM (Lim *et al*., 2022), our results suggest that use of bear parts has declined in recent years: five of 33 practitioners reported using bear parts in the five years prior to the survey, whereas 12 reported using these in the time before that. Bear parts were typically obtained from the wild, where the bears were killed and the valuable parts harvested. All of the 12 respondents that historically prescribed bear parts reported that they had purchased these directly from hunters, with some adding that they had requested hunters to target bears. In the five years before the survey, respondents tried methods such as purchasing from hunters (*n*=3/5), seeking to buy from markets (*n*=1/5), TCM shop (*n*=1/5) and from another country (*n*=1/5), where the practitioner reported getting bear parts from Myanmar through Laos to Cambodia. Hesitancy to speak openly about use of high-profile wildlife and the need for discretion when buying parts (Lim *et al*., 2022) indicates that the practitioners we interviewed understood that hunting and purchase of bear parts is against the national law. They also understood that bears are increasingly rare in the wild, and that this is why these are now more difficult and expensive to obtain. However, even respondents that said they had never used bear parts or had not used them in the previous five years mentioned that clients still request these. Some also reported that they were still asked by rich and high-status customers to obtain bear parts.

"*In past five years, I use only alternative plant medicines to bear parts because I could not find them anymore. But there still have people who come to ask me but I could not find for them. Now is no more forest, so it is hard to find bears. In past 40 years, there were a lot of bears; it was easy for me to contact to hunters for buying. I used to pet them as well, I got two bears, one of them I just got it by exchanging with 1 rice sack which 50kg rice to hunter. The most of people who asked me to find bear parts were high status person, Oukhna status*." [Female, 68, Khmer-Lao, TKM practitioner & traditional birth attendant, Stung Treng]

"*It is very difficult to get them during these past five years because there is no more forest in Cambodia; it is substituted by cashew nut and rubber fields. If we want, we can buy from Vientiane in Laos, but it is so expensive...; Nowadays, it's imported from Burma to Laos, then to Cambodia. In past 40 years, it was easy to find. Since after Pol Pot regime, other countries bought those parts from us*." [Male, 54, Khmer-Lao, TKM practitioner & Khmer healer, Stung Treng]

When asked how they were more likely to act, most practitioners (*n*=17/33) declined to answer, whereas some (*n*=9/33) said that they would advise bear products to people suffering from an ailment that they knew could be treated with these and others (*n*=7/33) reported that they would supply bear products if requested to.

Gallbladder and bile from different bear species

When asked which bear species provides the most medicinally effective gallbladder and/or bile, most respondents (*n*=22/33) stated that they did not know, whereas some (*n*=7/33) believed that Asiatic black bear and sun bear have the same medicinal properties. Three regarded Asiatic black bears as more desirable, although all respondents suggested that this was not related to medical efficacy but to the larger size of the species which has a bigger gallbladder and produces more bile liquid. Only one practitioner regarded the gallbladder of sun bears as being more effective due to the belief that its diet mainly comprises honeybees, which results in the gallbladder containing high concentrations of medicinal ingredients.

Table 1 Bear parts used in traditional Khmer medicine and their associated ailments, according to TKM practitioners.

Bear parts	Confirmed as TKM		Used for (no. of practitioners)
	n	%	
Gallbladder	31	94	Body pain (6), tonic (5), internal/external bruising (4), diabetes (3), cool medicine and fever (2), improving vision (2), post-partum treatment and women-related ailments (1). Other (*n*=1): shattered head, chickenpox, HIV, kidney disease, heart disease, gallbladder disease, cancer, lung disease, liver disease, sleeping pill, headache, haemorrhoids.
Bile	29	88	Body pain (7), tonic (6), internal/external bruising (4), diabetes (4), cool medicine and fever (3), post-partum treatment and women-related ailments (2). Other (*n*=1): shattered head, chickenpox, HIV, kidney disease, heart disease, gallbladder disease, cancer, lung disease, liver disease, sleeping pill.
Claws	30	91	Worn to protect from bad spirits (11), cool medicine and fever (rubbed medicine) (5), body pain (2). Other (*n*=1): nner organ disease, post-partum treatment, healing agent, headache, malaria, internal bruising.
Teeth	26	79	Personal protection (spiritual belief) (10), cool medicine and fever (2). Other (*n*=1): healing agent, tonic, post-partum treatment, bruising.
Bone	26	79	Body pain (4), healing agent for dermatosis (4), cool medicine and fever (3), internal/external bruising (2). Other (*n*=1): myelodysplastic syndromes, inner organ disease, female urethritis, personal protection.
Blood	23	70	Body pain (8), tonic (4), internal/external bruising (4), getting fair skin (2). Other (*n*=1): myelodysplastic syndromes, sexually transmitted diseases, post-partum treatment, diabetes, lung disease.
Paws	22	67	Ailments related to body pain (7), tonic (5). Other (*n*=1): improving sexual ability, myelodysplastic syndromes, antibiotic, healing agent, fever.
Skin	16	48	Healing agent for dermatosis (5), asthma (2), tuberculosis (2). Other (*n*=1): post-partum treatment, cancer, scrofula, internal/external bruising.

Alternatives to bear parts

According to almost half of our respondents (*n*=15/33), aliments that can be treated with bear gallbladder and bile can alternatively be treated with plant-based medicine. The specific alternative depends on the type of illness and the experience of the practitioner. However, the identity of these plant-based alternatives was not clearly reported and we are not aware of any official documents that support this suggestion. Some respondents (*n*=7/33) refused to identify the alternative plants and ingredients because they were hesitant to share their knowledge, whereas others (*n*=8/33) did not know of alternatives. One respondent stated that he could not share the information because according to rules of practice provided by his teacher, he could provide medicines but not information on their ingredients. Another possible reason why practitioners might not have wanted to share certain information is that they may have felt their knowledge was not supported by the official texts on TKM, such as the *Traditional Medical Treatments—Translated from Ancient Khmer Palm-Leaf Manuscript* used by the National Center for Traditional Medicine. The TKM practitioners we interviewed commonly reported that they had learnt their skills from kin who were practitioners that had not been formally trained. They also noted that they remembered the remedies without having written records or reference books for these.

Although plant-based alternatives were reported as substitutes by some respondents, bear gallbladder and bile were still perceived as the most effective medicine in TKM. For instance, respondents stated that bear gallbladder can help the speedy recovery from illnesses faster than alternatives. However, respondents did cite plant-based medicines which could treat illnesses also treated with bear bile such as body pain, fever and post-partum disorders, which are common ailments. One respondent who noted that bear gallbladder is used to treat illnesses such as HIV, cancer, lung disease and liver disease also stated that there is no alternative, while another noted that bear gallbladder can be substituted with elephant ivory for treating serious illnesses. These reported uses are not supported by TKM texts employed by the National Center for Traditional Medicine.

Discussion

Our study indicates that several bear parts including dry gallbladder, bone, blood and paws were historically used in TKM. Among these parts however, only bear gallbladder is explicitly mentioned in the Khmer medicine pharmacopeia—*Materia Medica* (Hieng *et al*., 2011). This text is used in training courses for TM practitioners and health subject students at the National Center for Traditional Medicine in Phnom Penh. However, the illnesses for which bear gallbladder are prescribed by our respondents are not mentioned in this. According to a key informant at the centre, the use of wildlife parts is only mentioned in Hieng *et al*. (2011) as an introduction to animal-based medicine and the centre does not encourage use of wildlife in medicine. Our respondents also reported that liquid bear bile was part of TKM and has the same healing properties as gallbladder. However, the practitioners we interviewed stated they mainly used bear gallbladder in dried form and rarely used liquid bile. Additionally, liquid bile is not mentioned in Hieng *et al*. (2011) and according to informants at the National Center for Traditional Medicine and Cambodian Traditional Healer Association, it does not form a part of TKM practice. As such, knowledge about bear bile could be a recent introduction into TKM practice in Cambodia and an influence of TM practices in China and Vietnam where bear bile extraction occurs (Foley *et al*., 2011). In response to separate questions, all of our key informants claimed that TKM has been influenced by TM systems in neighbouring countries in recent decades e.g., China, Vietnam, Thailand and Laos. It was further suggested that this influence was because TM products from these countries are imported to Cambodia and because foreign nationals in Cambodia seek such products from their home countries.

We found that bear gallbladders were prescribed to treat common illnesses and improve health (Table 1). Bear bone, blood and paws were also used to treat these ailments. However, claws and teeth were prescribed for spiritual matters such as warding off bad spirits. These results are similar to Davis *et al*. (2020b, 2020c) who found that individuals across Cambodia believe that bear bile and gallbladder are effective for treating certain illnesses. These studies found that bear parts were used for common ailments such bruising, which is consistent with our study. However, our respondents identified further ailments that may be treated with gallbladder and bile. This is to be expected as the TKM practitioners we interviewed would have greater medicinal knowledge than the average member of the general public.

Asiatic black bears produce especially high levels of tauroursodeoxycholic acid, a conjugated form of ursodeoxycholic acid, which is a medicinally active ingredient of bear bile (Hagey *et al*., 1993; Foley *et al*., 2011). Historically, the bile of Asiatic black bear has been more sought-after than that of other bear species (Feng *et al*., 2009; Foley *et al*., 2011). Although sun bears produce relatively low concentrations of tauroursodeoxycholic acid, their gallbladders are used in TM despite there being no studies confirming their effectiveness (Crudge *et al*., 2019). Medicinal knowledge is documented in ancient and modern medicine in countries such as South Korea, China and Vietnam, where bear farming has been established to supply TM practices (Foley *et al*., 2011). This farming led to a decline in wild bear populations due to the need for continual supply of bears to the farms (Feng *et al*., 2009). While bear farms have yet to be created in Cambodia, the kingdom held the record for the highest number of bear-related seizures between 2000–2011 and is a major source country for the supply of live bears to farms in other countries, as well as the illegal market (Burgess *et al*., 2014). Our study also supports previous studies that found a relatively high level of domestic demand exists for bear gallbladder and other products in Cambodia (Davis *et al*., 2020b, 2020c). However, prescription of bear products was apparently higher in historical times, having reportedly decreased in recent years for bears and other wildlife (Lim *et al*., 2022). The rarity of certain animals and difficulty in finding wildlife products including bear parts was noted by our respondents as having resulted in their replacement with plant-based alternatives. However, they also stated that products such as bear gallbladder are still perceived as high-quality medicine in TKM and that this motivates some practitioners to continue seeking these. Our study highlights that while the supply of bear parts to TKM practitioners may have lessened in recent years, their advice to patients may continue to promote demand for these in TKM. Notwithstanding this, the rural TKM practitioners (such as *Kru Khmer* or *Kru Thnam Khmer*) in our study mostly hailed from relatively poor households. As a result, purchase of valuable bear parts might require more money than these typically possess and so they may be less likely to supply such medicines to customers. As such, they may advise customers on which animal parts they should find by themselves from sources such as hunters, middlemen, other TM shops and markets. Rural communities are often closer to protected areas where bears are still extant and so can more easily access wild bears if they have the right hunting tools (Davis *et al*., unpubl. data).

Conservation implications

Bears persist in Cambodia despite high levels of habitat loss and illegal wildlife trade and consumption (Burgess *et al*., 2014, Davis *et al*., 2019a). Understanding of traditional knowledge and cultural practices regarding natural resources is important to promote effective communication between conservation communities, TM communities and rural communities, who often live at critical geographic junctions for conservation (Alves & Rosa, 2005; Alves *et al*., 2020). TKM practice is still important for local primary healthcare in rural areas and we found that consumers of TKM trust the knowledge of TKM practitioners and follow their advice. Lim *et al*. (2022) highlighted that TKM practitioners also have a role in guiding and advising about medicine to patients. Additionally, when it comes to bear parts in TKM practice, we found that practitioners may be less likely to provide bear parts to patients, but may advise people to seek these out. This may contribute to the continued use and trade of bear parts in Cambodia. This behaviour is similar to how TCM practitioners in China informally recommend bear bile to their customers, rather than providing a formal prescription (Hinsley *et al*., 2021).

Behaviour change interventions should be implemented in rural areas of Cambodia that continue to be more isolated from biomedical facilities. These should encourage the use of plants over animal species in TKM treatments. Because use of bear parts has deep roots in Khmer tradition and is perceived as highly effective, this behaviour may be ingrained and difficult to change. One possible leverage opportunity could be to strengthen pride in natural heritage, similar to the framework of successful RARE Pride campaigns (Butler *et al*., 2013). Such campaigns could recruit TKM practitioners as spokespeople, drawing on their reliance on the forest and the wildlife within to encourage a conservation ethos. However, such campaigns would need to be carefully researched, designed and tested to ensure that they do not cause unsustainable exploitation of plant species, and/or engender greater interest in animal-based TKM use. This could be accomplished with research that builds on our study, such as focus group discussions with TKM practitioners and local community groups, to identify common plants that could be grown in local communities as a more accessible alternative to bear bile, gallbladder and other animal products.

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First description of the ranging behaviour of Asian woollynecks *Ciconia episcopus* using GPS tracking in Cambodia

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មូលន័យសង្ខេប

យើងបង្ហាញការពិពណ៌នាលើកដំបូងអំពីអាកប្បកិរិយានៃការកំណត់តំបន់ក្រោយប្រលែងសត្វកុកពាក់អំបោះ *Ciconia episcopus* ចំនួនបួនក្បាលដែលបានប្រលែងក្នុងព្រៃរបោះជម្រុះស្លឹក នៅដែនជម្រកសត្វព្រៃសៀមប៉ាងនៃព្រះរាជាណាចក្រកម្ពុជានាឆ្នាំ២០១៩។ សត្វកុកពាក់អំបោះចំនួនបីក្បាលត្រូវបានជួយសង្គ្រោះពីការជួញដូរខុសច្បាប់ រីឯសត្វកុកមួយក្បាលទៀតត្រូវបានបង្កាត់ និងថែរទាំក្នុងទីបង្កាត់រយៈពេលប្រាំឆ្នាំមុននឹងធ្វើការប្រលែង។ សត្វកុកពាក់អំបោះទាំងបួនក្បាលត្រូវបានភ្ជាប់ជាមួយឧបករណ៍ GPS ដើម្បីអង្កេតពីអេកូឡូស៊ី និងទំនោរទីជម្រករបស់ពួកវាក្រោយធ្វើការប្រលែង។ យើងបានគណនាព្រំខណ្ឌតំបន់រស់នៅរបស់ឯកត្តៈនីមួយៗដោយប្រើវិធីសាស្ត្របីផ្សេងគ្នា (minimum convex polygon, kernel-density estimation, និង Brownian bridge) ហើយបានសិក្សាពីទំនាក់ទំនងរវាងឥរិយាបទដែលបានកត់ត្រា និងវត្តមាននៃអូរ វាលស្រែ និងថ្លុកទឹក។ ដោយរួមបញ្ចូលវិធីសាស្ត្រទាំងអស់ យើងបានរកឃើញតំបន់ប្រើប្រាស់ស្នូលដូចដែលបានកំណត់ដោយរបាយបម្រើបម្រាស់ (UD=50%) ដែលមានរបាយពី ២.៣ គម² ទៅ ៥០.៦ គម² ($\bar{x}$=15.66, SD=13.76) ចំណែកឯព្រំខណ្ឌតំបន់រស់នៅត្រូវបានកំណត់ដោយ UD=95% លាតសន្ធឹងពី ៨.២ គម² ទៅ ២៥៤.៥ គម²($\bar{x}$=89.22, SD=66.00)។ តំបន់រស់នៅមានទំនាក់ទំនងវិជ្ជមានជាមួយវត្តមានព្រៃឈើ ហើយមានទំនាក់ទំនងតិចតួចជាមួយវត្តមានវាលស្រែ និងថ្លុកទឹក។ ភាពរស់រានមានជីវិតរបស់សត្វកុកពាក់អំបោះពីរក្បាលអស់រយៈពេលបួនឆ្នាំ និងការបង្កាត់ពូជដោយជោគជ័យក្នុងព្រៃបង្ហាញពីការស្តារឡើងវិញនូវសត្វកុកពាក់អំបោះដែលបានរឹបអូសមកមានតម្លៃអភិរក្ស ហើយដែនជម្រកសត្វព្រៃសៀមប៉ាង និងតំបន់ការពារផ្សេងទៀតនៅព្រះរាជាណាចក្រកម្ពុជាជាកន្លែងសមស្របសម្រាប់នាំត្រឡប់មកវិញនូវប្រភេទសត្វកុកពាក់អំបោះ។

Abstract

We present the first description of the post-release ranging behaviour of four Asian woollyneck storks *Ciconia episcopus* released in 2019 in the deciduous dipterocarp forests of Siem Pang Wildlife Sanctuary, Cambodia. Three of the birds were rescued from illegal trade as chicks and the fourth was captive bred and rehabilitated in captivity for five years before release. The birds were fitted with GPS trackers to investigate the ecology and habitat preferences of the species after release. We calculated home ranges for each individual using three different methods (minimum convex polygon, kernel-density estimation & Brownian bridge) and studied correlations between recorded positions and the presence of streams, rice fields and waterholes. Combining all methods, we found core-use areas as defined by utilization distribu-

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tion (UD=50%) ranged from 2.3 to 50.6 km^2 ($\bar{x}$=15.66, SD=13.76), whereas home range as defined by UD=95% extended from 8.2 to 254.5 km^2 ($\bar{x}$=89.22, SD=66.00). There was a positive correlation with the presence of forest streams and a weaker correlation with the presence of rice fields and waterholes. Survival of two birds for four years and successful breeding in the wild demonstrates that rehabilitation of confiscated Asian woollynecks has conservation value and that Siem Pang Wildlife Sanctuary and equivalent protected areas in Cambodia are suitable reintroduction sites.

Keywords Asian woollyneck, Cambodia, *Ciconia episcopus*, home range, kernel density, woolly-necked stork.

Introduction

The Asian woolly-necked stork or Asian woollyneck *Ciconia episcopus* [Boddaert, 1783] (Fig. 1) occurs as *C. e. episcopus* in South and Southeast Asia and *C. e. neglecta* in Indonesia. Depending on the source, the species is usually split from its counterpart, the African woolly-neck *C. microscelis*, based on geographical separation and remains one of the least studied waterbird species in the world (Sundar, 2020). Most studies concerning Asian woollyneck have been conducted in South Asia (India, Nepal & Bangladesh) (Sundar, 2006; Hasan & Ghimire, 2020; Roshnath & Greeshma, 2020; Ghimire *et al*., 2022), where the species inhabits a variety of habitats over a wide range, thrives in anthropogenic landscapes and the population is considered at least stable (Hasan & Ghimire, 2020; Roshnath & Greeshma, 2020; Win *et al*., 2020). As a consequence, the IUCN threat category for the species has recently been downgraded from Vulnerable to Near Threatened to reflect these findings (BirdLife International, 2023). However, in Southeast Asia, the population is thought to have undergone a considerable decline and the species' range is presumed to have contracted due to multiple factors including habitat loss, hunting and nest disturbance (IUCN, 2020). Contrary to what is observed in South Asia, it has also been proposed that habitat destruction and degradation, and conversion to agriculture has been the leading driver of the decline in Southeast Asia (IUCN, 2020). Habitat use and occupation of human modified landscapes has not yet been studied for populations in Southeast Asia and it is unclear what role they really play in the species ecology. The reality is that the species is poorly studied and as such, the lack of information on its behaviour, ecology and breeding success in Southeast Asia hinders its conservation and requires specific scientific and conservation attention (Ghimire *et al*., 2021a). It is also likely that the best conservation outcomes will be achieved by specifically addressing local particular threats.

At a national level in Cambodia, Asian woollyneck has been listed as Near Threatened (Goes, 2013). As elsewhere in Southeast Asia, conservationists lack reliable data to assess the population or range, as a census has yet to be undertaken. Occurrence data from the Global Biodiversity Information Facility (GBIF, 2023) suggests a decreasing population since 2017 (Fig. 2), but it is not possible to determine the Cambodian population trend from this data alone as it is biased by the number of observers or locations surveyed. In Siem Pang Wildlife Sanctuary (SPWS), the location of every sighting of the species has been recorded since November 2021. Histori-

Fig. 1 Asian woollyneck fitted with a GPS-GSM tracker on a nest in Siem Pang Wildlife Sanctuary, 2023.

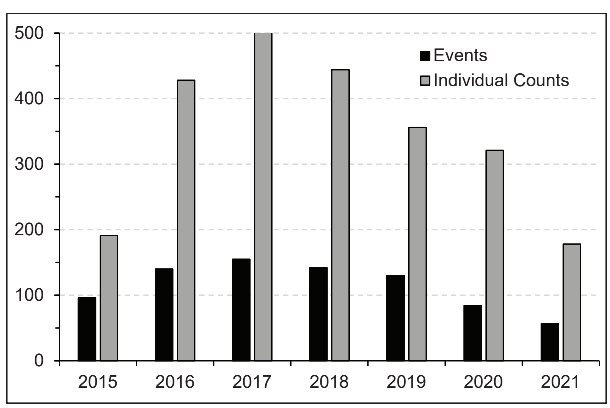

Fig. 2 Frequency of records in GBIF for Asian woollyneck in Cambodia, 2015–2021.

cally the species has been recorded breeding in Pong Kreel village (Eames, 2014) with one nest found along the O'Khampa river in 2021. Yet large numbers were recorded in the area in the past, with as many as 44 seen in a single flock over the sanctuary on 16 June 2010 (Eames, 2014) and a flock of 15 in July and 22 in October 2020. In 2022, 92 sightings were recorded, suggesting a total of 212 birds and a maximum of nine at the same time (Rising Phoenix, unpubl. data). Asian woollyneck has been identified as a species population in SPWS that could benefit from reinforcement from a captive source (Gray *et al*., 2019). Informed conservation of the species in SPWS requires at least a basic understanding of its ecology and habitat preferences and it is with this in mind that four Asian woollynecks were fitted with GSM-GPS trackers and released in SPWS in 2019. Habitats in SPWS were deemed suitable for the species given the numerous previous records and it was expected that the birds released would settle a short distance from the release site due to their captive origin. The aims of our study were to evaluate the survival of captive birds released in a new environment, to improve understanding of their ranging behaviour and habitat preferences and to compare soft-released and hard-released animals. Obvious limitations included a small sample and that birds of a captive origin may behave differently from wild birds. Initial planning included augmenting our sample size by annually releasing additional storks under the same conditions in subsequent years.

The concept of animal home ranges has experienced a new research boom in the last two decades, as demonstrated by a significant number of studies concerning the ranging behaviours of more and more species (Mertzanis *et al*., 2011; Silva-Opps & Opps, 2011; Ram *et al*., 2022). Home range was originally defined as the "area traversed by the individual in its normal activities of food gathering, mating and caring for young" (Burt, 1943). It has now given way to a more statistical approach, most commonly defined as "densities of use", that reflect estimates of the locations of an animal across a landscape (Powell, 2000; Laver & Kelly, 2008; Powell & Mitchell, 2012). The development of GPS technologies and their miniaturization have greatly improved our ability to track animal movements over large temporal and spatial scales (Bridge *et al*., 2011; Udyawer *et al*., 2018), allowing researchers to refine and improve knowledge of habitat species preferences. These have especially been an asset in the study of elusive or migratory species.

Several methods have been developed to calculate home range, although the two most common approaches are traditionally the minimum convex polygon and the kernel-density estimation (Worton, 1987; Laver & Kelly, 2008). Minimum convex polygon continues to be used for comparative purposes, although the technique has numerous biases (Börger *et al*., 2006; Nilsen *et al*., 2008). For example, minimum convex polygon may overestimate the actual home range by incorporating unused areas between peripheral locations or underestimate it if sampling duration is too short (Burgman & Fox, 2003; Getz *et al*., 2007). This is particularly a problem for migratory species or species that travel between feeding and nesting grounds and explains why kernel-density estimation is often preferred for estimating a species home range (Fieberg & Börger, 2012). However, the main issue with the kernel-density estimation model lies in the selection of a proper bandwidth and the assumption that locations are not correlated, which is not respected for data recorded from animals equipped with GPS devices (Nelson, 2011; Walter *et al*., 2011). The Brownian bridge movement model, an evolution of the fixed kernel-density estimation method, appears to solve the problem of autocorrelation as it also considers the time between successive locations in estimation of the utilization distribution (Horne *et al*., 2007) and is of particular interest for migratory species. However, for species that occupy a relatively small home range, area differences calculated by the different models are likely to be small (Fieberg & Börger, 2012).

In this study, we present the first description of post-release ranging behaviour of four Asian woollynecks in Cambodia using GPS tracking. In doing so, we provide results of home range calculations for this species using different methods to allow for greater comparison with other studies, as well as spatial correlation analysis between recorded positions and the presence of streams, rice fields and waterholes.

Methods

Study area

Siem Pang Wildlife Sanctuary is a 130,000 ha (1,300 km²) protected area in Stung Treng Province, Cambodia. It has been identified as a Key Biodiversity Area (Tordoff *et al*., 2012) and was designated as a wildlife sanctuary in 2016. The sanctuary is bordered to the north and west by the Xe Pian National Park (2,400 km²) in Laos and to the east by Virachey National Park (3,380 km²) in Cambodia. Habitats in SPWS include a mosaic of deciduous dipterocarp forest which account for 50% of the sanctuary and semi-evergreen forests which comprise 40%. The remaining area comprises degraded forests or grassland (8%) and riverine habitat (2%) (BirdLife International Cambodia Programme, 2012). The sanctuary's connectivity with neighbouring protected areas is a conservation asset, in allowing animals to move between the different geographies (Brennan *et al*., 2022).

Equipment and satellite tracking

Four Asian woollynecks (ACCB local ID numbers 0160009, 0160010, 0160012 and 0160015, hereafter WNS 09, WNS 10, WNS 12 and WNS 15, respectively) were released in October 2019 in SPWS following a protocol designed by the Angkor Centre for Conservation of Biodiversity (ACCB), a conservation centre of the Allwetterzoo Münster, Germany. All individuals were sourced from ACCB where they were either captive bred (WNS 12) or had been rescued from the illegal wildlife trade as chicks (WNS 09 & WNS 10) or a sub-adult (WNS 15) and rehabilitated between 2014 and 2015. Two of these were hard-released directly into the wild on 29 October, without previous acclimatization or supplementation (WNS 12 a female and WNS 10 a male) and two were soft-released (WNS 09 a male and WNS 15 a female). Hard release usually excludes any training, but in our protocol, we ensured that our hard-released birds were able to catch live prey and did not exhibit imprinted behaviour before release. Conversely, soft-release normally includes an acclimatization period, pre-release animal training and post-release food supplementation (Resende *et al*., 2021), whereas in our protocol, our soft-released birds were kept in an aviary in the sanctuary and fed with live prey for one month until their release on 6 November. Supplementary food was also offered for one week after release and then discontinued as the storks did not return to the aviary. Birds were sexed by DNA testing on feather samples collected at ACCB and analysed in Germany. Before their release, all four birds were fitted a solar-powered OrniTrack-25 GSM-GPS tracker (Ornitela, Vilnius, Lithuania) which was attached with a teflon ribbon. The total weight of the system was 33g, representing 1.65% of the body weight of each bird (2 kg), less than the 3% usually deemed acceptable for this kind of tracking (Murray & Fuller, 2000; Kenward, 2001; Barron *et al*., 2010). Following the same protocol, a fifth bird (a male Asian woollyneck, WNS 0160002) was hard-released on 4 December 2020. Rescued from the illegal wildlife trade, this bird arrived at ACCB in December 2009 and hence was at least 11 years old at the time of release. Release sites in the sanctuary were chosen in deciduous dipterocarp forest, not far from cultivated areas located on the edge of SPWS, as it was anticipated that the birds might favour a mix of both habitats.

We programmed our transmitters to record a GPS position every three to six hours depending on the battery load and to attempt to transmit data by GPS every three hours. Because of these restrictions, we did not extract a daily time-budget from our data and all datapoints were pooled in analysis, irrespective of their time of day (morning, afternoon or evening).

Home range calculations

Data were processed in R 4.2.2 (R Core Team, 2022) using the adehabitatHR package (Calenge, 2006) and ArcGIS Pro for graphical presentation. Prior to calculations, coordinates in our datasets were converted in R from WGS 1984 to UTM 48N. A total of 67,944 raw positions were recorded from four storks (WNS 09=27,729 logs, WNS 10=4,126 logs, WNS 12=7,605 logs, WNS 15=28,484 logs). These data were cleaned by removing logs that lacked latitude or longitude coordinates, were triangulated by less than three satellites, where recorded speed was higher than 90 km/h and logs with inconsistent figures for altitude (i.e., under -200m or over 1,000m). Data points obtained before release were also removed (before 29 October 2019 for WNS 12 and WNS 10 and before 6 November 2019 for WNS 09 and WNS 15), as well as outliers. The tracker of the fifth bird (WNS 0160002) released in December 2020 was not fully functional and recorded only 57 datapoints between 5 December 2020 and 26 January 2021 and was not included in the study. This stork was found dead of unknown causes in February 2021 at which point the tracker was recovered.

We calculated home range using three methods, the minimum convex polygon (MCP) method, the kernel-density estimation (KDE) method and the kernel-density Brownian bridge method (BBM). Home ranges were calculated for the dry season (November to April) and the wet season (May to October) using the three methods, as it was hypothesized that habitat use would differ seasonally, consistent with variation in habitat use observed for the species in India and Nepal (Kittur & Sundar, 2020; Roshnath & Greeshma, 2020; Tiwary, 2020). It should be noted that each method has its own biases, which is why they are often used in combination for comparative purposes. For instance, MCP often overestimates home ranges as it includes outermost locations and so includes areas that an animal never uses (edge effect). The KDE method is heavily influenced by the bandwidth parameter and is also subject to edge-effect, whereas BBM requires a high frequency of location data to accurately estimate home range and assumes that the movement of an animal follows a Brownian motion model, which may not always be accurate. These biases are usually balanced by confining the analysis to a smaller utilization distribution. In our study, core-use area was defined as the 50% isopleth area of the utilisation distribution and home range as the 95% isopleth area. Those thresholds are standard in ecological research and are commonly used to facilitate comparisons between studies and species as they balance precision and accuracy and may be of behavioural relevance in often corresponding to areas of highest use (Laver & Kelly, 2008; Silva-Opps & Opps, 2011; Abril-Colón *et al*., 2022; Lee *et al*., 2022).

In employing the KDE method, we set the smoothing parameter *h* to *href*. For BBM, we calculated the smoothing parameter *sig1* related to the speed of the animal using the function *liker* to estimate *sig1* using the maximum likelihood approach (Calenge, 2006; Horne *et al*., 2007). The *liker* function determines the most likely path an animal took between observed locations, which in turn helps estimates the better-suited smoothing parameter *sig1*. We then used a Mann-Whitney test to compare home ranges provided by the different methods and to compare these between seasons and sexes.

Spatial regression analysis

Spatial regression analysis was conducted in ArcGIS Pro v3.0.3 (Esri Inc., California, USA). A rectangular grid of 139,435 cells each measuring one hectare was created, covering all positions recorded of storks within and outside of SPWS. Shapefiles of known rivers, trapeangs (seasonal waterholes) and rice fields were obtained. These features were selected due to higher detection of storks around streams and trapeangs inside the sanctuary, and because of the hypothesized importance of rice fields, as demonstrated in South Asia (Kittur & Sundar, 2020; Ghimire *et al*., 2021b). Forest cover and land cover were not employed as variables, as deciduous dipterocarp forest is the dominant landscape in the area. The analysis was undertaken by fitting a Poisson model using the number of GPS logs in each cell as the dependent variable, and the presence of a stream, a waterhole, or a rice field as the explanatory variables. The same protocol was used to explore spatial correlation of 107 positions of stork sightings recorded by Rising Phoenix staff between November 2021 and July 2023.

Ethics

Every precaution was taken during the reseach to minimize stress and disturbance to the study birds. The weight of the transmitters and the fitting method were chosen in accordance with standard protocols and recommendations. Handling was minimal and undertaken by professionals. Release protocols were designed to maximize the chances of each individual returning successfully to the wild according to the IUCN/SSC (2013) and were approved by the Forestry Administration of the Cambodian Ministry of Agriculture, Forestry and Fisheries and the Cambodian Ministry of Environment.

Results

Following cleaning of datasets, a total of 65,468 data points remained inside the boundaries of 13.99–14.32°N and 106.12–106.42°E for analysis (WNS 09=26,527 logs over 1,364 days, WNS 10=3,963 logs over 196 days, WNS 12=7,243 logs over 463 days & WNS 15=25,454 logs over 1,364 days). The last recorded movement of WNS 10 occurred on 11 May 2020, 196 days after hard release, after which the bird was found dead and the tracker recovered on 2 June 2020. The last recorded position of WNS 12 occurred on 3 February 2021, 464 days after hard release (battery was 94% charged) and the last data transmission was on 13 February 2021, indicating a low battery. The latter tracker was not recovered and the fate of the stork is unknown. The trackers of WNS 09 and WNS 15 trackers remain active at the time of writing, over three years and nine months after their soft release.

We considered that the birds would be settled into their new habitats by the end of December 2019, more than a month after release. At that time (mean position from 25–31 December 2019), three of the birds had moved an average of two kilometres from their release site (WNS 09=1.4 km, WNS 10=2 km, WNS 12=2.6 km), whereas WNS 15 had flown 12 km to the west. At the start of September 2023, both of the remaining tracked birds had paired with wild individuals within the sanctuary, with two chicks successfully fledging from one of the two nests.

Home ranges

Combining all methods, core-use areas (defined as 50% of the utilization distribution) ranged from 2.3 to 50.6 km^2 ($\bar{x}$=15.66, SD= 13.76), whereas home ranges (defined as 95% of the utilization distribution) extended from 8.2 to 254.5 km^2 ($\bar{x}$=89.22, SD=66.00) (Table 1, Fig. 3).

WNS 10 and WNS 15 had the smallest and largest home ranges respectively, regardless of season (Fig. 4). Home ranges in the wet season (May to October) were not significantly smaller than the dry season (November to April) (Mann-Whitney test, p>0.05). Ranges calculated using MCP for 50%, 90% and 95% of the utilization distribution were always larger than those provided by KDE and BBM, although not significantly and the differences in ranges provided by KDE and BBM were not significant. Additionally, no significant differences in range size were found between soft and hard released birds or between males and females. These results should be

Table 1 Home range areas for different percentages of the utilization distribution for four study birds. N/A indicates failure to successfully compute a test.

Home Range (km²)	WNS 09 (Male, hard release)			WNS 10 (Male, soft release)			WNS 12 (Female, hard release)			WNS 15 (Female, soft release)		
	50%	90%	95%	50%	90%	95%	50%	90%	95%	50%	90%	95%
Minimum convex polygon												
Dry & wet season	11.0	57.5	88.2	9.0	20.5	24.3	50.6	102.7	111.0	31.7	199.1	248.5
Dry season	14.2	72.1	136.2	8.6	19.8	22.9	28.0	95.4	101.6	49.6	179.5	254.5
Wet season	8.7	27.0	50.5	3.1	8.0	8.2	36.7	55.5	61.2	14.4	101.5	162.4
Kernel-density estimation												
Dry & wet season	2.3	30.5	42.2	6.0	19.8	24.3	17.8	61.1	81.1	16.8	81.3	106.5
Dry season	11.4	57.1	82.3	5.7	19.5	23.8	15.6	58.9	76.7	23.2	100.3	133.8
Wet season	0.0	21.1	30.5	5.7	20.6	27.0	14.3	51.8	69.7	9.7	52.2	67.7
Brownian bridge movement												
Dry & wet season	8.4	52.4	78.7	3.7	18.1	23.9	9.5	52.2	72.2	21.1	109.6	169.2
Dry season	20.4	7.4	N/A	3.8	18.1	23.7	19.9	820.2	N/A	40.4	1753.7	N/A
Wet season	11.5	N/A	N/A	7.6	N/A	N/A	9.2	N/A	N/A	26.3	N/A	N/A

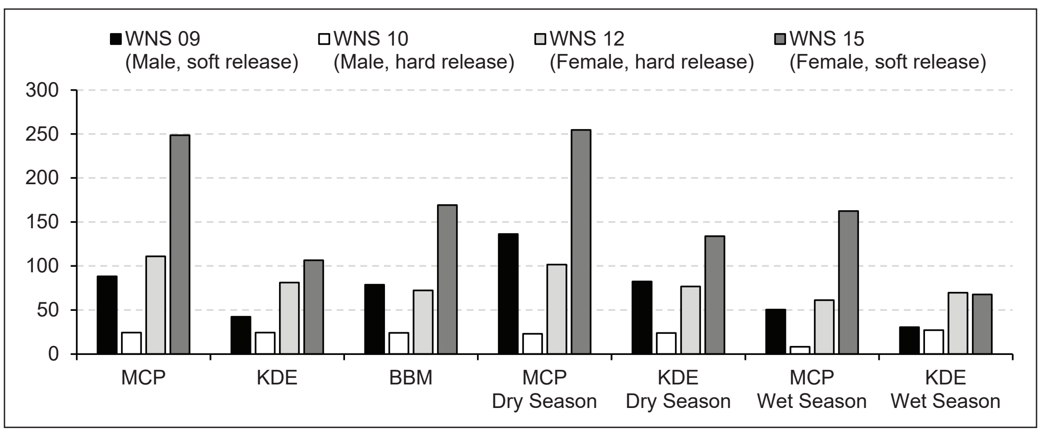

Fig. 3 Home ranges in km² (95% of utilization area) of study birds by season using different estimates.

regarded with caution however, considering our sample comprised only four individuals.

Spatial regression analysis

A generalized linear regression undertaken using the number of GPS logs in each cell as the dependent variable and the presence of a rice field, a waterhole or a river as explanatory variables returned respective log coefficients of 0.502, 0.808 and 1.079 (Table 2), all of which were statistically significant at $p<0.01$. These results indicate that all else being equal, we observed a positive correlation between the presence of storks and the three explanatory variables, with the presence of a river being the most strongly associated (Fig. 5). Moran's I index value was 0.5 and z-score was 265.4, indicating spatial auto-correlation of the data.

Performed on our set of 107 sightings, the same analysis returned log coefficients of 0.247 for rice fields, -11.342 for waterholes and 0.520 for streams (Table 3). As such, the occurrence of storks and presence of rice fields and streams was positively correlated, with no or very little correlation with the presence of waterholes (expo-

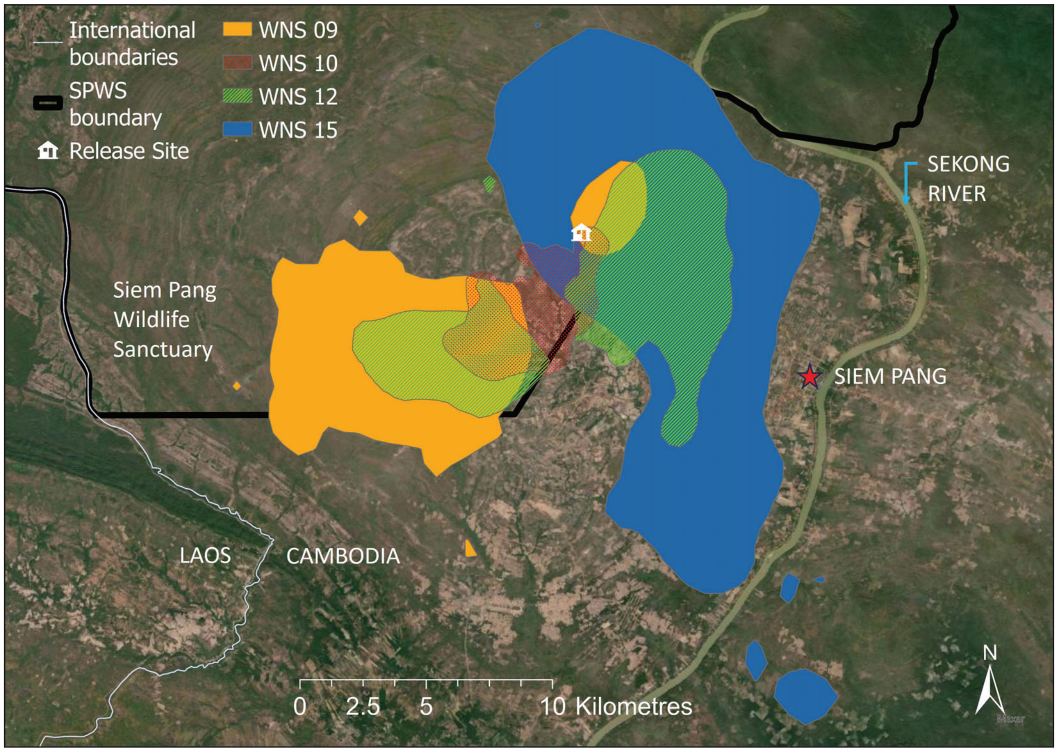

Fig. 4 Home ranges of four study birds based on 95% of utilization area and Brownian bridge method.

Table 2 Results of model of habitats in locations recorded for four study birds.

Variable	Coefficient	Standard Error	z	*p*
Intercept	-0.986	0.004	-218.024	<0.01
Rice fields	0.502	0.005	99.584	<0.01
Waterholes	0.808	0.045	17.574	<0.01
Streams	1.079	0.008	134.861	<0.01

Table 3 Results of model for habitats of Asian woollyneck sightings from November 2021 to July 2023.

Variable	Coefficient	Standard Error	z	*p*
Intercept	-7.040	0.095	-73.997	<0.01
Rice fields	0.247	0.156	1.581	0.114
Waterholes	-11.342	482.122	-0.024	0.981
Streams	0.520	0.240	2.167	0.030

nentiated coefficient of 0.000012). However, only the correlation with waterways was statistically significant at $p<0.05$. Further, the standard error for the presence of waterholes was very large, indicating that this estimate is very imprecise.

Discussion

We tracked four Asian woollynecks over a period of three and half years, from their release in October 2019 to July 2023. Based on the data collected, these individuals were sedentary and spent significant time in SPWS. One month after release, three of the four individuals had settled less than 3 km from the release site, which could reflect the captive habituation of the birds but also indicates the availability of suitable habitat for the species. The two storks that were soft released remain alive after 3.5 years with active trackers and bred in 2023. This proves that captive-reared woollynecks can survive to breed in the wild. Of the two storks that were hard released, one was found dead almost 200 days afterwards and contact with the transmitter of the other bird was lost after more than 15 months. It is difficult to draw significant conclusions as our sample is very small, but this suggests that soft release may be beneficial for the long-term survival of

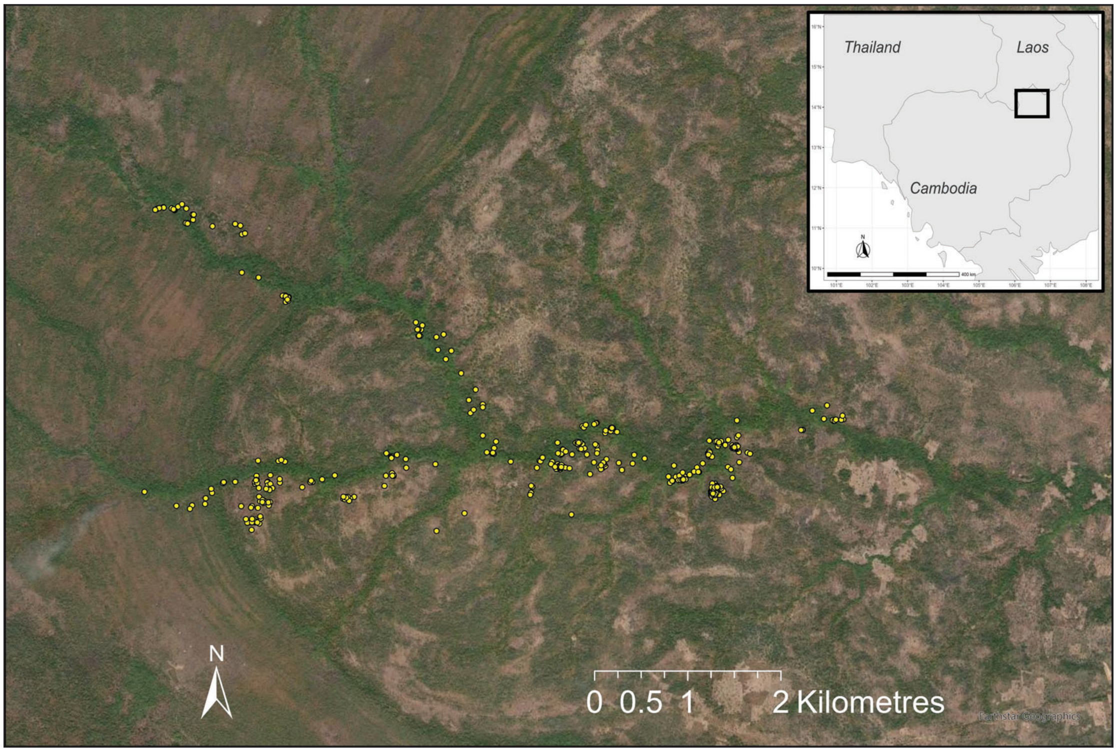

Fig. 5 Data points for WNS 09 in April 2020 (dry season), highlighting the occurence of the bird in the immediate vicinity of watercourses, which are dry at this time of year.

released birds. Our study also demonstrates that SPWS provides suitable habitat for rehabilitated and released Asian woollynecks, as we also documented breeding of released birds with wild partners. Release of additional individuals of this and other stork species should be considered to strengthen local populations.

To our knowledge, we provide the first estimate of the core-use and home range areas for Asian woollynecks. As our results are based on captive released birds, it would be interesting to compare our results with metrics obtained from wild birds. We found MCP 50% was 25.6 ± 19.6 km^2, MCP 90% was 95 ± 77.1 km^2, MCP 95% was 117.75 ± 94.4 km^2 and KDE 95% was 63.5 ± 37.1 km^2 (Table 2). Of the eight species in the *Ciconia* genus, white storks *C. ciconia* have been the most studied. Zurell *et al*. (2018) found the home range of the species was variable with an overall mean of 78.3 ± 219.9 km^2 for MCP 95%. No significant differences in home range were found between sexes, locations or years, whereas breeding status significantly reduced home ranges, consistent with findings for other bird species (Tanferna *et al*., 2013; Stenhouse & Moseby, 2023). Mean MCP 95% was 21.4 ± 29 km^2 for breeding individuals, compared to 205.8 ± 80.5 km^2 for non-breeding storks. This likely reflects pressure to gather food to feed young under time and movement constraints, whereas non-breeding birds are relieved from such constraints (Johst *et al*., 2001; Zurell *et al*., 2018). As the breeding status of our study birds was uncertain, we did not investigate home range in this context.

Xu *et al*. (2021) studied four juvenile Oriental white storks *C. boyciana* which were tagged in the nest in the wetlands of Xingkai Lake National Reserve in China and followed by telemetry in the wild before they migrated. They found that the core home range (50% MCP) of fledglings ranged from 0.08 to 6.15 km^2, whereas the 95% MCP ranged from 6.10 to 14.24 km^2. The birds were tracked for 31 to 44 days in summer and provided 3,253 locations in marsh meadow habitats. In studying the reintroduction of *C. boyciana* in Japan, Ezaki & Sagara (2014) calculated a home range (90% MCP) of less than 12.5 km^2 for seven breeding pairs, which was centred around the nest. This contrasts with Jiguet & Villarubias (2004) who found a mean core range of 538.5 ± 278.58 km^2 (core range defined as 75–95% of the utilization distribution, based on fixed kernel density estimation) for 12 black storks *C. nigra* of varying status (breeding, non-breeding, young and adults). They also noted that range sizes between breeding and non-breeding adults were similar.

Our results suggest that *C. episcopus* may occupy a smaller home range compared to *C. nigra* and *C. ciconia*, but a larger one compared to *C. boyciana*. Asian woollynecks occur in the tropics where food availability per unit of area may be greater than temperate latitudes where the other species occur. A smaller home range could be expected for this tropical species, but this was not the case compared to *C. boyciana*. This could be due to the small sample size of both studies, alongside other limitations inherent to these. For instance, Jiguet & Villarubias (2004) studied free-living black storks, whereas Xu *et al*. (2021) studied fledglings of oriental white storks in the first days of flight and our study concerns captive-released adult birds. This is a major caveat which may bias our understanding of ranging behaviour. Indeed, birds released from captivity often tend to range around release sites (Wilson *et al*., 1992; Van Heezik *et al*., 2009), although this is not always observed and may be species dependent and influenced by variables such as the presence and density of conspecifics around the release site (Lockwood *et al*., 2005). It should also be noted that species have differing breeding habits that may influence their home range. For example, *C. ciconia* breeds in colonies and may have to share resources with other breeding pairs, increasing its foraging area. *Ciconia nigra* and *C. episcopus* are solitary breeders but do not depend on the same habitats as *C. nigra* uses temperate forests, which could also account for the higher home range. Consequently, it must be kept in mind that our results should be regarded as preliminary and that studies of entirely wild birds will be required to determine if these behave in similar ways. Furthermore, studies of wild birds are needed to understand the impact of captive released birds on wild populations and if such these releases could lead to populations with restricted or biased behaviours as a result of the released birds having been in captivity for a long period of time.

We found a positive correlation between the occurrence of storks and from the least to the strongest association, rice paddies, waterholes and streams. The correlation with streams seems obvious when visually displayed on a satellite basemap (e.g., Fig. 5). The positive Moran's index value of 0.5 indicates a moderate to strong spatial autocorrelation in the data, which is easily explained by autocorrelation of recorded GPS logs. This is unavoidable and warrants caution in interpreting the results of spatial regression. When comparing these results with analysis of visual sightings of storks in and around SPWS, we found the same positive association with streams and rice fields and no correlation with trapeangs. However, our sample only comprised 107 locations which is a relatively small data set. Both datasets indicate that the presence of waterways is the most useful indicator for the occurrence of Asian woollynecks and this should be considered in planning for future releases.

It is hard to draw definitive conclusions in relation to our release protocol or the sex of the birds due to our small sample size and a larger sample will be needed to improve understanding of how these factors may influence ranging behaviour. We hypothesized that the home range of the storks would vary seasonally in relation to food availability, water levels in waterholes or streams and/or breeding status. However, no significant differences were found, which is not consistent with observations in South Asia (Kittur & Sundar, 2020). Likewise, the home range of some of our birds overlapped at times during the first study year but it is unknown whether the birds actually interacted or not.

White storks frequently use artificial nests or artificial structures when nesting (Vaitkuvienė & Dagys, 2015; Bialas *et al*., 2020) and habitat quality and food availability are regarded as deciding factors for breeding success and positive population trends (Nowakowski, 2003). Numerous studies have shown *Ciconia* species (or at least *C. ciconia*, *C. boyciana* and *C. episcopus*) favour nesting close to human settlements and crops and are positively associated with traditional agriculture but negatively associated with pesticide use (Ezaki & Sagara, 2014; Kittur & Sundar, 2021; Xu *et al*., 2021). Our study suggests that Asian woollynecks in Cambodia may have habits similar to the species in Nepal, Myanmar and India where positive associations with agricultural landscapes have been reported (Sundar, 2006; Win *et al*., 2020; Ghimire *et al*., 2021b).

In Japan, friendly farming methods play an important role in conserving reintroduced oriental white storks by preserving biological diversity and sufficient prey to support their feeding in paddy fields (Naito *et al*., 2014). These approaches could provide a solution for Asian woollyneck conservation in Cambodia. Wildlife-friendly agricultural practices that do not use pesticides are already in use in Siem Pang District as a result of the IBIS Rice scheme (Rising Phoenix, unpubl. data). Further development of farming methods to reduce or eliminate pesticide use could also be encouraged for conserve other waterbird species in Cambodia.

Reliable and increasingly smaller GPS trackers can provide very precise and frequent data on bird movements, allowing for better understanding of habitat preferences and ranging behaviour. Our successful use of data loggers on Asian woollynecks in SPWS demonstrates that the sanctuary provides suitable habitats, with sufficient foraging opportunities for the birds to settle at short distances from release sites and occupy a mean home range of <65 km^2 (KDE 95%). No significant differences were found home ranges between seasons or sex and the influence of breeding status could not be assessed. Further studies with a large sample size are needed to facilitate more robust estimates of the home range and habitat preferences of Asian woollynecks in Cambodia.

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Rawson, B. (2010) The status of Cambodia's primates. In *Conservation of Primates in Indochina* (eds T. Nadler, B. Rawson & Van N.T.), pp. 17–25. Frankfurt Zoological Society, Frankfurt, Germany, and Conservation International, Hanoi, Vietnam.

Reports:

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