Aquatic Polyphaga (Insecta: Coleoptera) from Kampong Speu Province, Cambodia

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Communities & REDD+
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Editorial—REDD+ in Cambodia: how local communities can benefit from forest conservation

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REDD+ is an international initiative for reducing emissions from deforestation and forest degradation, coupled with conservation and sustainable management of forests and enhancement of forest carbon stocks. Key aspects of the initiative include safeguarding local livelihoods, alleviating poverty and improving tenure security for rural people in developing countries (Lawlor et al., 2013). REDD+ also has the potential to deliver enormous benefits for biodiversity conservation because forests in the developing world provide a diverse set of habitats for plants and animals and are widely threatened by ongoing forest clearance and degradation (Gardner et al., 2012).

REDD+ is one of the most developed mechanisms for payments for ecosystem services in Cambodia and can address the urgent national need for sustainable sources of finance for protected area management (Simpson & Souter, 2017). Cambodia has already completed its REDD+ readiness stage, including implementation of several REDD+ demonstration projects. In May 2017, the Royal Government of Cambodia endorsed Cambodia's national REDD+ strategy for 2017–2026. The strategy focuses on improving management and monitoring of forest resources and forest land use, strengthening implementation of sustainable forest management, mainstreaming approaches to reducing deforestation, building capacity and engaging stakeholders.

Cambodian REDD+ demonstration projects have contributed to development of the national REDD+ strategy and will continue to foster improvements to national and project-based approaches. The following projects have been approved by the Royal Government of Cambodia: Oddar Meanchey Community Forestry REDD+ project, Keo Seima Wildlife Sanctuary REDD+ project (previously known as the Seima Protection Forest REDD+ project), Prey Lang REDD+ project, Tumring REDD+ project (in collaboration with the government of the Republic of Korea), and the Southern Cardamoms REDD+ project. All of these operate in the voluntary carbon market, and most are independently accredited using the Verified Carbon Standard (VCS; now known as Verra) and the Climate, Community and Biodiversity Alliance (CCBA) standards. The Prey Lang project is being designed and implemented under the Joint Crediting Mechanism in collaboration with the Japanese government.

One of Cambodia’s first REDD+ projects, in Oddar Meanchey Province, has faced several challenges including land grabbing in community forests by outsiders, which drives deforestation in the project area. In 2008, border conflict with Thailand around the Preah Vihear temple complex resulted in the establishment of several military camps within the project area, which negatively affected project implementation due to security issues. Funding shortfalls and the proposal of a complex mechanism for REDD+ benefit-sharing has delayed the distribution of benefits to communities involved in the project, reducing incentives for cooperation. In addition, due to stakeholders’ limited understanding of how the voluntary carbon market functions, particularly the need to actively pursue sales, the project raised unrealistic expectations of the amount of revenue that the local community would receive from carbon sales. Project partners, particularly technical partners such as Pact Cambodia and Terra Global Capital, and local NGOs, have withdrawn due to the lack of funding from donors and carbon sales available to support project activities. According to the VCS database (www.vcsprojectdatabase.org), 48,000 of the 597,210 carbon credits certified in the first verification (2008–2013) have been sold, although revenue from these sales has yet to be...
distributed to local communities. Much work is needed to restore confidence in this project.

The Cancun safeguards under the United Nations Framework Convention on Climate Change (UNFCCC) require individual countries to make provisions in social, ecological and political spheres to mitigate adverse effects of REDD+ and promote additional benefits. There is particular focus on measures to respect indigenous rights and ensure the “full and effective participation” of communities and equitable benefit sharing (Chapman et al., 2015). The CCBA standard requires local community participation, equitable benefit sharing and improved tenure rights for local and indigenous communities. Often, only the financial benefits of REDD+ are considered. However, a range of non-financial benefits can also accrue from REDD+, including security of land tenure rights, employment, improved wellbeing, promotion of traditional knowledge and cultures and biodiversity conservation (Lawlor et al., 2013). Benefit sharing can help build the legitimacy of and support for REDD+ projects (Lindjem et al., 2011) and is therefore crucial for successful project implementation.

Another early demonstration project, the Keo Seima Wildlife Sanctuary (KSWS) REDD+ project, has the most developed strategy for sharing REDD+ benefits with local communities. This builds upon lessons learned from the Oddar Meanchey project and is adaptive, allowing improvements when additional insights from it or other projects become available. Benefit sharing in KSWS combines strengthened resource rights, alternative livelihood projects and additional bonus incentives for community development projects. The first two are important for long-term project sustainability, whereas additional bonus incentives encourage community engagement and show how local communities can benefit from REDD+ and forest conservation (Duchelle et al., 2017). In the future, some of the additional bonus incentives awarded in KSWS will be performance-based, with the aim of encouraging positive changes in behaviour.

Following the sale of the first certified credits from the KSWS REDD+ project, the first round of unconditional bonus incentives has been awarded to local communities. These have supported community development activities including construction of community meeting halls, road culverts, toilets, water pumps, and running water systems; school renovations; and development of community-based ecotourism. Based on preferences expressed by the community and previous research in the project area (Travers et al., 2016), these benefits are shared at the community level rather than as payments to individual households. Challenges encountered during the first round of benefit sharing include a lack of capacity in communities for financial management of their development projects, and the difficulty in optimizing the trade-off between ensuring a high level of transparency and accountability of financial procedures and using processes that are simple enough for community representatives to follow effectively.

Implementation of REDD+ demonstration projects and completion of the REDD+ readiness process have provided invaluable insights to ensuring the long-term success of the initiative in Cambodia. Benefit sharing should play a central role in REDD+ projects, with a focus on delivering financial and non-financial benefits. These can jointly contribute to improving the livelihoods and welfare of local communities that rely on forest resources in project areas. This combined approach to REDD+ benefit sharing can improve community participation in projects and contribute to reducing deforestation (e.g., through participation in community-led patrols and deforestation monitoring). Non-financial benefits provide the bulk of available benefits and their importance to a successful REDD+ project should not be underestimated. Sharing of financial benefits is likely to become possible only at later stages in project implementation, once sales of verified carbon credits have been made.

It is important to ensure that there are adequate sources of funding available to support project costs, especially in the early stages, which are typically cost-intensive and precede the generation of revenue through sale of carbon credits. Securing funding for these early stages is particularly significant as key activities in this period should include creating a benefit sharing system and building community capacity to enable full participation in the project. Priority areas for capacity building include financial management, administration and conceptual understanding of REDD+. Capacity building is essential throughout a project’s lifetime, and dedication of considerable time and effort to raising awareness and building capacity in local communities will further the benefits that can accrue to them as a result of REDD+.

Simple financial procedures and a transparent system for sharing conditional and unconditional incentives from a REDD+ project should be designed with community input. This is an important step in obtaining free, prior, and informed consent and serves to increase the likelihood that communities have realistic expectations of what their REDD+ project can achieve.

Effective implementation of REDD+ benefit sharing depends on specific political and social contexts (Fischer et al., 2016). As such, the lessons learned from REDD+...
projects in Cambodia will be critical for the development of procedures for REDD+ benefit sharing nationally. This is timely because Cambodia is now preparing for REDD+ results-based payments under the UNFCCC. Previously, the impact of pilot REDD+ projects on national decision-making has been limited by a lack of sharing information (Korhonen-Kurki et al., 2012). The national technical working group on REDD+ benefit sharing in Cambodia intends to foster an environment where experiences are readily exchanged to inform the future success of REDD+ in Cambodia. We encourage all those involved in REDD+ projects in Cambodia to continue to share their experiences, both positive and negative, with the working group to maximize the utility of lessons learned and increase the extent to which local communities can benefit from forest conservation.

References


A 10-year national species action plan for the river tern *Sterna aurantia* in Cambodia

The river tern *Sterna aurantia* is a fish-eating bird that nests on sandbars along the Mekong and 3S rivers in the Stung Treng and Kratie provinces during the dry season. During the rainy (non-breeding) season, the species travels to Tonle Sap Lake. Once widespread throughout Southeast Asia, populations of river terns have rapidly declined in recent decades. In Cambodia, they have decreased by over 80% since the late 1990s, from ca. 300 to fewer than 70 adults. This population decline qualifies the species as regionally Critically Endangered.

Threats to the river tern include human harvesting of eggs for food, predation of nest and chicks by wild and domestic animals (especially rats), trampling of nests by domestic water buffaloes, flooding of nests by water releases from upstream hydropower dams, and human and animal disturbance at breeding sites. Other key threats include the destruction of sandbar breeding habitat by human encroachment and agriculture, development of hydropower dams, and gold and sand mining operations. Declines in fish prey and climate change also threaten the species.

To protect river terns in Cambodia, a 10-year national species action plan was developed by the Department of Freshwater Wetlands Conservation of the Cambodian Ministry of Environment in partnership with BirdLife International, NatureLife Cambodia, Worldwide Fund for Nature (Cambodia programme) and the Department of Natural Resource Management and Development of the Royal University of Phnom Penh, with support from the National Geographic Society.

The action plan is based on a consultation workshop held in Phnom Penh on 24 October 2017 that was attended by over 40 stakeholders, including international experts and representatives from government agencies, NGOs and academic institutions. It describes river tern ecology, population trends and threats, and outlines a conservation strategy aimed at species recovery. The full text of the action plan is available at: www.researchgate.net/publication/327136025_Ten-year_species_action_plan_for_the_Cambodian_population_of_River_Tern_Sterna_aurantia_2018-2028

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New Master of Science in Sustainable Ecosystem Management

In developing countries such as Cambodia, sustainable management of natural resources presents challenges for scientists and environmental managers due to strong human dependence on these resources. In particular, the question of how this can be achieved while meeting increasing human needs in a period of rapid global change is a major issue.

The Master of Science in Sustainable Ecosystem Management has been created by the University of Battambang with support from the European Erasmus+ programme and a consortium of university partners in Europe and Southeast Asia. The MSc addresses the urgent need to strengthen capacity for biodiversity conservation and ecosystem management in Cambodia and Mekong River Basin. The degree curriculum is designed to provide students with advanced statistical, analytical and problem-solving skills in relation to trade-offs between biodiversity conservation and socio-economic development in the region. It also combines natural and social science subjects and integrative modelling techniques which emphasize the development of:

- Scientific knowledge and concepts for sustainable development
- Practical skills and leadership
- Independent academic and research skills
- Ability to convey new knowledge and research into decision support tools
- Global perspectives on ethics and sustainability under rapid global change
- Publications in international peer-reviewed journals

The degree is a two-year part-time (weekend) study programme which is taught in English and Khmer and includes foundation, core, elective and research courses and thesis studies. Students also have the possibility to undertake internships with associate partners in Cambodia, Thailand, China and Europe. Further details are available at www.conseaedu.eu/study-programs/ubbmaster/

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Workshop discusses vision for management of migratory fish in Cambodia

Despite the importance of migratory fish to Cambodian food security and livelihoods, no clear vision currently exists to manage and conserve these species across their diverse habitats throughout their life cycles. To help address this gap, the Wonders of the Mekong project held a workshop entitled “Needs, techniques, and risk assessment: toward a vision for migratory fish in Cambodia” in Phnom Penh on 17–18 October 2018. The workshop was organized by FISHBIO, hosted by the Inland Fisheries Research and Development Institute, Cambodia and the University of Nevada, Reno, and funded by USAID.

The workshop brought together over 65 participants from Cambodia and the Mekong Basin, as well as international speakers. The goal of the event was to identify high-priority data needs regarding fish migration in the Mekong Basin, develop a shared vision for management and conservation of migratory fish in Cambodia, and to discuss tools that can be used to achieve this vision. Presentations and group discussions identified key data needs such as identification of important habitats and spawning areas, migration routes and timing, and species’ life cycles, among others.

Speakers at the event presented a variety of tools and techniques for studying migratory fish which ranged from telemetry and sonar cameras to otolith microchemistry and genetic work, as well as local ecological knowledge of fishers. Themes that emerged during discussions on developing a vision for migratory fish included maintenance of abundant fish populations that can sustain themselves, migrate freely and support local food security. Good governance and enforcement were viewed as necessary to address threats and illegal fishing, as were conservation measures to protect fish habitats and raise awareness about sustainable fishing practices. The Wonders of the Mekong project will continue to develop this vision for management and conservation of migratory fish. A workshop report and policy brief are available at www.mekongfishnetwork.org/vision-migratory-fish-cambodia/

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International workshop for Eld’s deer conservation in Phnom Penh, 2018

Eld’s deer Rucervus eldii is a globally Endangered species in need of immediate conservation action across its range to prevent extinction of its geographically distinct subspecies. Conservationists from Cambodia, China, India, Laos, Myanmar and Thailand met in November for a workshop hosted by Kadoorie Farm and Botanic Garden and BirdLife International Cambodia Programme in Phnom Penh. The objectives of the event were to share expertise and lessons learned, to develop regional conservation strategies for the species, and to promote the maintenance of genetic diversity of populations through translocation or exchange. The meeting also hoped to raise momentum and support for Eld’s deer conservation.

The meeting comprised two days of indoor sessions and a half-day visit to Phnom Tamao Zoo and Wildlife Rescue Centre. The agenda included status reviews for each of the range countries and case studies regarding in-situ conservation, captive management, genetic studies and ecology. Roundtable panel discussions with leading practitioners and group and plenary discussions on specific conservation strategies were also held.

Eld’s deer was once widely distributed across lowland dry forests in tropical Southeast Asia. Of the three subspecies currently recognized, R. e. eldii has been reduced to a single site in India. Numbers of R. e. thamin in Myanmar and Thailand comprise perhaps 3,000 individuals centred on two protected areas in Myanmar. Thailand has reintroduced this subspecies. Small, scattered populations of R. e. siamensis occur in protected areas in Cambodia and Laos which are vulnerable to hunting and habitat loss. Re-introduction of the siamensis subspecies is underway in Thailand. Recent research indicates that the semi-wild population in Hainan (China) is genetically distinct and warrants appropriate conservation efforts.

Delegates at the meeting collectively expressed a desire to form an Eld’s deer working group under the IUCN Deer Specialist Group and agreed to identify national country coordinators to facilitate effective communication and international collaborations.

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Short Communication

Attracting white-shouldered ibises to safe roosting sites in Siem Pang Kang Lech Wildlife Sanctuary, Cambodia

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We assessed the roosting preferences of the Critically Endangered white-shouldered ibis Pseudibis davisoni and used decoys and played calls to attract the species to novel roost trees in Siem Pang Kang Lech Wildlife Sanctuary, north-eastern Cambodia. In roost assessments, the ibis showed a preference for particular tree species, notably Dipterocarpus intricatus. Roost trees were taller, in a more advanced stage of decay and their crowns touched other trees less compared to a matched control sample. Roost trees were also closer to villages than expected by chance. Two trees of differing species were selected that had not previously been used by the ibis, but which met its identified roosting preferences. Ten wooden decoys of white-shouldered ibis were deployed in the crowns of these trees and call playback was used to attract the birds to them. One tree (D. obtusifolius) attracted nine white-shouldered ibises over the four months of the experiment, with visits increasing towards the end of the study (September & October, 2016). Subsequently, the species was recorded during opportunistic visits to the same tree in 2018 which retained some of our wooden decoys. We suggest that decoys with or without call playback may be used to attract the white-shouldered ibis, although several months may be required before results are observed.

The global population of the Critically Endangered white-shouldered ibis is estimated at 1,000 individuals, 95% of which are located in northern Cambodia (BirdLife International, 2018). Wright et al. (2012a) showed that 74% of the Cambodian population roosted at sites outside of existing protected areas. The white-shouldered ibis roosts communally in large numbers which makes it vulnerable to hunting and protection of communal roosts is central to conserving the species.

Siem Pang Kang Lech Wildlife Sanctuary (SPKLWS) was designated by the Royal Government of Cambodia in May 2016 (Fig. 1). The wildlife sanctuary encompasses a large portion of the Western Siem Pang Important Bird Area (centred on 14°17' N, 106°27' E) and suitable deciduous dipterocarp habitat for the white-shouldered ibis. As such, the sanctuary provides an opportunity to protect key roost sites of the species, although at the time of its creation only two of the 22 roost sites known were located within SPKLWS. To address this challenge, we conducted an experiment using decoys and call playback to attract the white-shouldered ibises to suitable roosting sites within SPKLWS where the risk of hunting and disturbance are lower (Kress, 1983; Crozier et al., 2003).

To identify suitable trees for the experiment, we first assessed the characteristics of known roost trees used by the white-shouldered ibis. We recorded their species, height, diameter at breast height, percentage of crown touching another tree and decay stage. Decay stage was characterised using a six-factor level categorical variable modified from Cline et al. (1980) and Vonhof & Barclay (1996) (Table 1). To compare roost trees with other trees in the surrounding area which were available but not used, control trees were selected by walking 200 m from each roost tree on a sequential compass bearing of 0, 90,
180 or 270 degrees and selecting the closest mature tree to that location. One matched control tree was selected for each roost tree and assessed in the same way.

We assessed the roost preference of the white-shouldered ibis for different trees using non-parametric methods. A G test (Crawley, 2007) was used to compare roost and control trees. More specifically, we compared whether the frequencies of tree species used by the ibis differed significantly from the frequencies of tree species in the random control group (Table 2). Tree height, crown size and decay stage were compared using a Wilcoxon signed ranks test (R Core Team, 2016). The location of each roost tree in suitable habitat was also assessed. The distance of roost sites to villages was calculated in ArcGIS (ESRI, 2011). To determine the mean distance to villages for the forest as a whole (the control group), a regular grid of points with 100 m spacing was created for deciduous dipterocarp habitat within the study area and the distance of each point to the nearest village calculated. Due to the large difference in sample sizes between the roost treatment and control treatment, we used a permutation test with 2,000 Monte Carlo replications to compare the means of the two treatments using the perm package in R (Fay & Shaw, 2010; R Core Team, 2016).

We located 43 roost trees regularly used by the white-shouldered ibis at 22 separate sites. Numbers of roost trees per site ranged between one and 12. The species showed a significant preference for roosting in particular dipterocarp species (Table 2), especially D. intricatus (Log likelihood ratio statistic $G = 98.2$, $df = 6$, $p < 0.001$). The median height of roost trees was 17.5 m, compared with a median height of 12 m for matched control trees. This difference in tree height was significant ($V = 818.5$, $p = < 0.001$, $n = 43$). The median decay stage for roost trees was 4 and 1 for control trees. This difference in decay stage was also significant ($V = 595$, $p = < 0.001$, $n = 43$). The branches of 38 roost trees were entirely isolated from other trees (e.g., 0% of the tree crown touched other trees). The same was only true for 18 of the matched control trees (which had a median value of 10% of crown touching

![Figure 1](image-url) Siem Pang Kang Lech Wildlife Sanctuary (SPKLWS) and adjoining Siem Pang Wildlife Sanctuary (SPWS), northeastern Cambodia.

**Table 1** Tree decay stages recognized in this study (adapted from Cline et al. 1980 and Vonhof & Barclay, 1996).

<table>
<thead>
<tr>
<th>Factor level</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Live, healthy; no decay; no obvious defects.</td>
</tr>
<tr>
<td>2</td>
<td>Live, usually unhealthy; obvious defects such as broken top, cracks, or hollows present.</td>
</tr>
<tr>
<td>3</td>
<td>Recently dead; dead leaves still present, very little decay; heartwood hard.</td>
</tr>
<tr>
<td>4</td>
<td>Dead; no leaves, few twigs; top often broken; &lt;50% of branches lost; bark loose; heartwood hard; sapwood spongy.</td>
</tr>
<tr>
<td>5</td>
<td>Dead; most branches and bark lost; top broken; heartwood spongy; sapwood soft.</td>
</tr>
<tr>
<td>6</td>
<td>Dead; no branches or bark; broken off along mid-trunk; sapwood sloughing from upper bole; heartwood soft.</td>
</tr>
</tbody>
</table>

**Table 2** Tree species used for roosting by the white-shouldered ibis, compared with control tree species.

<table>
<thead>
<tr>
<th>Species</th>
<th>No. roost trees</th>
<th>No. control trees</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dipterocarpus intricatus</td>
<td>38</td>
<td>9</td>
</tr>
<tr>
<td>Dipterocarpus obtusifolius</td>
<td>3</td>
<td>7</td>
</tr>
<tr>
<td>Dipterocarpus tuberculatus</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td>Shorea obtusa</td>
<td>1</td>
<td>11</td>
</tr>
<tr>
<td>Catunaregam tomentosa</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Syzygium cumini</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Terminalia chebula</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>43</strong></td>
<td><strong>43</strong></td>
</tr>
</tbody>
</table>
Diagrams and illustrations are not included in the text. Differences between roost and control trees in the percentage of tree crown touching other trees were consequently significant ($V = 39$, $p < 0.001$, $n = 43$). The mean distance of the 22 independent roost sites to villages was 3.33 km, compared to 9.14 km for the entire area of deciduous dipterocarp forest we surveyed. This difference was also significant ($p = 0.002$).

Using the identified characteristics of roost trees, we selected two trees previously unused by the white-shouldered ibis in SPKLWS. The trees selected were as similar as possible to known roost trees (Table 3). The only limiting factor was that whereas the ibis prefers dead and decaying trees for roosting, we selected trees in a less advanced stage of decay to ensure that their upper reaches could be safely reached by climbers. This being assured, we made 10 decoys of the white-shouldered ibis from wood, painted these to resemble the plumage of the species and attached them to the upper branches of the trees to provide a visual cue to encourage the species to roost there (Feare et al., 2015; Fig. 2). The white-shouldered ibis roosts communally, vocalising frequently at dawn and dusk as it leaves and returns to roost sites.

To encourage the species to use our two decoy trees, we played call recordings of the ibis (obtained from www.xeno-canto.org) through a loudspeaker concealed near the base of each tree (Kress, 1983). Playback and monitoring was undertaken at each tree twice a week between 1700 and 1830 hrs from 14 June 2016 to 28 October 2016 to coincide with the period of communal roosting. The call recordings could be heard from 300 to 500 m away.

White-shouldered ibises were subsequently recorded at the second decoy tree (D. obtusifolius) (Fig. 2) on six separate occasions. Singletons were observed on three of these occasions, whereas two birds were observed on the remaining occasions. The frequency of visits increased towards the end of the experiment with five visits occurring during the third study month (September). Almost two years later, white-shouldered ibises were recorded during opportunistic visits to the same tree as follows: up to 30 birds on 2 and 3 July 2018 and 18 birds on 4 July 2018 (Fig. 3). Because we ceased call playback in October 2016, the birds observed in 2018 must have relied upon visual cues or memory to return to the roost tree.

### Table 3

<table>
<thead>
<tr>
<th>Species</th>
<th>Height</th>
<th>Decay stage</th>
<th>% of crown touching</th>
<th>Distance to villages (km)</th>
<th>No. decoys</th>
<th>Sampling occasions</th>
<th>WSI</th>
</tr>
</thead>
<tbody>
<tr>
<td>1  Dipterocarpus intricatus</td>
<td>16</td>
<td>2</td>
<td>0</td>
<td>6.9</td>
<td>4</td>
<td>37</td>
<td>0</td>
</tr>
<tr>
<td>2  Dipterocarpus obtusifolius</td>
<td>20</td>
<td>2</td>
<td>0</td>
<td>4.8</td>
<td>6</td>
<td>38</td>
<td>9</td>
</tr>
</tbody>
</table>

*Fig. 2* White-shouldered ibis next to a wooden decoy on the second experimental roost tree, 7 July 2016 (© BirdLife International Cambodia Programme).

*Fig. 3* Up to 30 white-shouldered ibis roosting on the second experimental roost tree, 2 July 2018 (© Jonathan Eames).
We found that the characteristics of known roost trees differed from our matched control trees, but did not attempt to infer which characteristics are most influential in roost tree selection. We simply document that white-shouldered ibises tend to select roost trees which are significantly taller than most trees in the landscape of SPKLWS and that they select *D. intricatus* at a greater frequency than would be expected by chance. They also appear to select roosts closer to villages than would be predicted by chance. It has been hypothesized that the white-shouldered ibis has an association with domestic buffalo because it feeds on invertebrates which are disturbed by the grazing and wallowing behaviour of the latter (Wright *et al.*, 2010; 2012b). Domestic livestock occur in greater density closer to villages and this might explain why roosting sites are located close to villages. Equally, ibises could prefer habitats with intermediate levels of disturbance a short distance from villages. However, we cannot exclude the possibility that our knowledge of roost trees may be spatially biased. Put simply, roost sites closer to villages are more likely to be known because these areas are more intensively used by people.

Our comparisons are limited because we were only able to employ decoys and call playback at two trees. We also acknowledge our lack of data for random control trees with similar characteristics to our experimental trees. White-shouldered ibises were not recorded at our first experimental tree which represented its preferred roost species (*D. intricatus*), but were recorded at the second tree which was larger, closer to human habitation and a different species (*D. obtusifolius*). Because optimal roost trees have likely declined in the landscape due to selective logging, it is possible that roost trees now used by the species represent the most favoured trees out of those that remain. Notwithstanding this, our partial success in attracting white-shouldered ibis to novel roost trees suggests that decoys and call playback have some utility in this respect. The increased frequency of visits towards the end of our experiment also suggests that these may need to be employed for significant periods before birds visit a desired location. However, it should be noted the effective distance from which it may be possible to divert birds using call playback will depend to some extent on the volume of the loudspeaker employed. We also acknowledge that we took a risk in conducting the study because our methods could be observed and copied by local hunters.

We are aware that decoys and call playback have been used to encourage breeding in the Critically Endangered Chinese crested tern *Sterna bernsteini* on the Mazu Islands of Taiwan (BirdLife International, 2013), and that acoustic enrichment techniques employed at the Bronx Zoo, New York, increased breeding success in a captive colony of the Critically Endangered northern bald ibis *Geronticus eremita* (Clark *et al.*, 2012). To our knowledge however, our study is the first attempt to use decoys and call playback to encourage modified roosting behaviour in the Critically Endangered white-shouldered ibis.

**Acknowledgements**

We would like to thank the Ministry of Environment and the Stung Treng Department of Environment for facilitating this research, which took place within the framework of project collaboration with BirdLife International. We would also like to thank the US Fish and Wildlife Service Critically Endangered Animal Conservation Fund, which provided financial support for this project.

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Leaf phenology effects on transpiration: leaf age-related stomatal conductance of *Dipterocarpus costatus* during the dry season in a Cambodian seasonally tropical evergreen forest

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Abstract

Leaf phenology regulates the leaf-age structure of a tree crown, which possibly governs leaf physiological activity. We evaluated the effects of leaf age on single leaf-scale transpiration in the tall dipterocarp *Dipterocarpus costatus* in a lowland seasonally tropical evergreen forest. As an index of leaf transpiration, we investigated the stomatal conductance ($g_s$) of intact leaves during the dry season in Kampong Thom Province, Cambodia. Leaf-age-dependent $g_s$ was found to peak in semi-mature leaves (17 days after unfolding), after which it gradually declined, although it remained at around 60% of the peak $g_s$ until four days before leaf fall. Ambient factors such as temperature and humidity were less strongly related to $g_s$. Our study also indicated a bimodal leaf flushing peak of *D. costatus* i.e., peak leaf flushing primarily occurred during the early dry season and secondary flushing during the late dry season. The results of our study indicate that, in addition to ambient factors, leaf-flushing phenology, as the governing driver of leaf age structure of the crown, could be a decisive factor in tree-level transpiration of *D. costatus* during the dry season.

Keywords

*Dipterocarpus costatus*, leaf phenology, lowland dry evergreen forest, porometer, transpiration, seasonal tropical forest.

Introduction

Tropical dry forests are widely distributed in pantropics where annual rainfall ranges from 600 to 1,800 mm and there is a distinct dry season of 3–8 months (Murphy & Lugo, 1986). The factors governing tropical dry forest ecosystems have attracted interest, given that seasonal variation in irradiance and water availability often do not coincide (Wright & van Schaik, 1994). Intensive research on Amazonian tropical dry forests has revealed that seasonal controls on the exchange of water and carbon rely on light availability followed by leaf-phenology (Saleska et al., 2003; Xiao et al., 2005; Huete et al., 2006; Hutyra et al., 2007). On the other hand, pantropic comparative studies have indicated lesser light control in Asian tropical forests, where there is variation in governing factors from primarily water-limited drier forests to wet evergreen forests showing slight positive trends with light availability (Huete et al., 2008).

Several studies on Southeast Asian tropical forests have clarified forest water cycles (Tanaka et al., 2008; Mizoguchi et al., 2009), where seasonally tropical evergreen forests demonstrate various transpiration patterns during the dry season. Peak transpiration was observed to occur at the end of the dry season in the less water-limited hill evergreen forests of northern Thailand (Tanaka et al., 2004), and in both the early and late dry season in the lowland evergreen forests of central Cambodia (Nobuhiro et al., 2009). In contrast, a lower transpiration rate was observed during the dry season in the water-limited evergreen forests of Sakaerat, Thailand (Pinker et al., 1980; Tanaka et al., 2008).

Hydrological process models have been used to simulate the water-cycle in Asian tropical dry forests (Tanaka et al., 2004; Kumagai et al., 2009). However, in the seasonally tropical evergreen forests in Cambodia, a model with vapor pressure deficit as the decisive factor failed to predict evapotranspiration peaks during the dry season (Nobuhiro et al., 2009). In fact, the potential evaporation rate, which is indicative of the climatic conditions that encourage evaporation from the water surface, cannot account for the peaks, in which highly variable mid-values (~5.5 ± 1.0 mm d$^{-1}$) and constant high values (~6.2 ± 0.2 mm d$^{-1}$) were reported for the early dry season and the late dry season, respectively (Chann et al., 2011). It is still not known why the peak of transpiration occurs in both the early and late dry season in seasonally tropical evergreen forests in Cambodia.

Leaf phenology, including leaf flush, maturation, senescence and fall, may be a key factor influencing ecosystem processes (Forrest & Miller-Rushing, 2010; Wu et al., 2017). However, little attention has been given to tropical evergreen forests because their leaf mass is relatively constant (Richardson et al., 2013). Leaf stomatal conductance ($g_s$) is positively and almost linearly related to transpiration (Burrows & Milthorpe, 1976; Ishida et al., 2006), although Meinzer et al. (1995) pointed out the influence of boundary layer on the relationship between $g_s$ and transpiration. Considerable changes in $g_s$ have been observed during leaf maturation (Field, 1987; Brodribb & Holbrook, 2005; Pallardy, 2010; Kenzo et al., 2016). In turn, leaf phenology regulates the leaf age structure in individuals, which may govern leaf physiological activity. Iida et al. (2013), who investigated the same study site as Nobuhiro et al. (2009), suggested that tree-scale transpiration depended on the leaf age structure of the crown for several tree species composing the low to mid layers of the canopy in seasonal tropical evergreen forest in Cambodia. We hypothesized that the peaks of
evapotranspiration during the dry season derives from high leaf-physiological activities that are associated with the leaf age structure of crowns. Based on this hypothesis, we expected that leaf age-related physiological activities would be observed for dominant tall tree species comprising the upper canopy layers of the forest. Moreover, we predicted that leaf phenology increases the physiological activities of crown layers via the exchange of old leaves with new ones during the dry season.

The objectives of our research were to investigate the leaf-age-related leaf gₘ of tree species in a seasonally tropical evergreen forest in Cambodia. We examined a tall evergreen dipterocarp, D. costatus C.F. Gaertn (Dipterocarpaceae, Poorna, 2002; Tani et al., 2007), composing the top layer of the forest. The leaf photosynthetic features of D. costatus have been documented in the forest, but only from fully mature leaves (Kenzo et al., 2012). We measured the intact leaf gₘ of two D. costatus trees during the dry season, covering both leaf-exchange and leaf-unfolding phenomena. In addition, we investigated the leaf-exchange phenology of the study trees. Based on the results, we discuss leaf phenology effects on transpiration at the tree scale. Finally, we consider the potential changes in seasonal variation in tree-scale transpiration that may accompany intermittent and multiple leaf flushings per year.

Methods

Study site and species

The study was conducted in Kampong Thom Province, central Cambodia (12.76°N, 105.48°E), in a lowland seasonally dry evergreen forest covering a flat, gently rolling alluvial plain at an altitude of 80–100 m above sea level (a.s.l.). The forest is located on extensive Quaternary sedimentary rock (Wakita et al., 2004) with a typically sandy soil that is classified as Haplic Acrisol (Alumic, Profodic) in the world reference base system (Toriyama et al., 2007, 2008). The groundwater table seasonally fluctuates from approximately 2 m to 10 m in depth (Araki et al., 2008; Ohnuki et al., 2008a, 2008b). Fine roots (<2 mm) grow in the deep soils near ~10 m, corresponding to the groundwater table at the end of the dry season (Ohnuki et al., 2008b).

Annual precipitation and average monthly temperature are 1,540 mm and 27°C (Kenzo et al., 2012). Average monthly temperature ranges from 24 to 29°C (Chann et al., 2011). The seasonal tropical climate is governed by monsoons and can be divided into three seasons: an early dry season (from October or November to December) with little rain (50 mm), high groundwater levels (from ~50 to ~100 cm) and decreasing air temperature (from 25.5 to 24.5°C); a late dry season (from January to March or April) with little rain (100 mm), declining groundwater levels (from ~125 to ~200 cm) and increasing air temperature (from 25 to 28°C); and a rainy season (from April or May to September or October) providing >90% of the annual precipitation, increasing groundwater levels (from ~180 to ~50 cm) and moderate air temperature (approximately 26°C) (Kabeya et al., 2008; Chann et al., 2011). Monthly averages for meteorological data are available from the website of the Changes in Water Cycle in the Mekong River Basin research project in Cambodia (www.ffpri.affrc.go.jp/labs/cwcm/dataset/EF/EF_E.htm). Daily maximum temperature in the canopy (40 m above ground) reaches 35°C in the early dry season, while a daily minimum temperature of 20°C has been recorded (November 2009, Kenzo et al., 2012). Wind speeds at heights of 38 or 60 m range from 2.5 to 3.5 m s⁻¹ during the dry season, relatively higher than the wet season (from 2.0 to 2.5 m s⁻¹: www.ffpri.affrc.go.jp/labs/cwcm/dataset/EF/EF_E.htm). Monthly 10-m scalar wind speeds were estimated at 2.2–2.4 m s⁻¹ in January and December and 1.6–1.9 m s⁻¹ in February–March and November in surface JRA-55 reanalysis data (http://jra.kishou.go.jp/JRA-55/index_en.html) with a spatial resolution of 1.25°×1.25°. These wind speeds are far higher than 0.25 m s⁻¹, resulting in decoupling conditions between stomatal and boundary layer conductance (Meinzer et al., 1995). The seasonal difference in solar radiation is 20%, ranging from 20 kJ m⁻² day⁻¹ in April to 16 kJ m⁻² day⁻¹ in September. The two months with the lowest radiation occur in the late rainy season (Chann et al., 2011).

Dipterocarpus costatus Gaertn. f. (Chhoeuteal Bankouy in Khmer) was selected as our study plant. It is a tall evergreen tree belonging to the Dipterocarpaceae family (Poorna, 2002). This species is widely scattered in lowlands, hills and upper dipterocarp forests in India (Andaman Islands), Bangladesh, Myanmar, Thailand, Malaysia (Peninsular Malaysia), Laos, Cambodia, and Vietnam (Ashton, 1979, 1998; Toyama et al., 2013). However, D. costatus is listed as Vulnerable in the IUCN Red List of Threatened Species (Ly et al., 2017) and is a target species for logging in the study area (Kim Phat et al., 2002). Its height commonly reaches 30–35 m (Toyama et al., 2013), although it can reach heights of 45 m and stem diameters of 130 cm (Authors, unpublished data), thereby dominating the upper canopy layer of dry evergreen forests (Tani et al., 2007).
Stomatal conductance measurement

We measured intact leaf gs as an indicator of transpiration using a portable leaf porometer (SC-I: Decagon, Pullman, WA, USA). Tanaka-Oda et al. (2012) verified that gs measurements (SC-1) were significantly correlated with the values measured with a portable photosynthesis meter (LI-6400: Li-Cor, Lincoln, NE, USA), but found that gs measurements (SC-1) were lower than the values measured with the LI-6400 portable photosynthesis meter under humid conditions (relative humidity of ambient air = 65%). Thus, we first validated the gs measurements (SC-1) by comparison with values measured with a LI-6400 using 39 identical leaves from branches cut from 18 individuals of 14 tree species, including 11 leaves of D. costatus at the study site in November 2009. Complete details of the LI-6400 measurement methods can be found in Kenzo et al. (2006, 2012). The gs measurements (SC-1) were positively correlated with the maximum absolute conductance values measured using the LI-6400 ($r^2 = 0.35$ and $r^2 = 0.26$ for D. costatus), and a paired $t$-test confirmed that there was no significant difference between the measurements made by the two instruments ($p = 0.3966$). Similar correlations have been reported in several broadleaf trees (Tanaka-Oda et al., 2012).

We selected two reproductive D. costatus individuals with similar tree crown sizes for gs measurements. Tree I had a diameter at breast height (DBH) of 48 cm and height of 23.8 m, while Tree II had a DBH of 43 cm and height of 23.8 m. Tree II was located 1.5 km southwest of Tree I. Both trees were growing beside a road and so even mid-layer branches were fully exposed to the sun for part of the day. The time of sun exposure was different between the two trees because Tree I and Tree II were located on the west and east side of the road, respectively. We selected five to six branches per individual at a height of 12–13 m, and all of the leaves on the selected branches were marked for identification using a 12-m-high platform. A total of 140 leaves were measured, and the approximate flushing period of each leaf was estimated. The gs of all marked leaves was measured concurrently with air temperature and humidity as ambient factors using the SC-1. Relative photosynthetic active radiation was not measured. Measurements were conducted in March 2009 for Tree I and from December 2009 to early March 2010 for Trees I and II. All measurements took place at least 1.5 hrs after sunrise to avoid low light intensities in early morning and before 1300 hrs to avoid midday photosynthesis depression (Kenzo et al., 2003). We conducted daytime measurements to determine the degree of midday gs depression at the study site on 3–4 March 2009 for 42 leaves. Each leaf was repeatedly measured 9–14 times. Measurement time ranged from 0843 to 1620 hrs. For newly flushed leaves, measurements commenced when the leaves had unfolded sufficiently to cover the porometer measurement area (6.3 mm diameter). We defined the leaf age as day 0 on the day of the first measurement.

Leaf-unfolding observation

To investigate branch-level leaf phenology, the maturity of all leaves on selected branches was confirmed for each gs measurement using leaf colour: young (yellowish green, hairy), semi-mature (pale green, sparse hairs), mature (dark green, glabrous) and senescent (dull green with yellow or brown spots). The dates of leaf emergence and fall were recorded for all leaves on the branches studied.

Leaf size (length and width) was recorded for 36 (2 December 2009), 94 (12 December 2009) and 99 (2 March 2010) intact leaves. Leaves that fell during the measurement period ($n = 29$) were collected, and their sizes were recorded. The leaf expansion pattern was estimated from the relationship between the degree of leaf expansion relative to the full leaf size (%) and leaf age.

Litter trap measurement

The leaf exchange phenology of D. costatus was estimated from fallen leaves collected in litter traps between September 2010 and April 2011. Three litter traps covering an area of ~0.5 m$^2$ were set within a lowland evergreen forest stand located 1 km north of Tree I; therefore, the fallen leaves were not collected from Tree I or Tree II. Litter was collected three times per month, and was then oven-dried, sorted (into leaves and leaf bud scales of D. costatus and others) and weighed. Leaf bud scales cover new leaf buds and fall when the buds burst.

Statistical analysis

Relationships between mean gs values and measurement date, leaf age and ambient factors were assessed using a generalised linear model (GLM) framework, incorporating individual leaves as a random effect. Significant differences among leaf ages were evaluated using the post hoc Tukey–Kramer HSD test. To examine the degree of midday gs depression, the relationship of gs values with measurement time was assessed using a GLM incorporating individual leaves as a random effect. Regression tree analysis was also used to assess when midday gs depression occurred for each leaf. The most accurate predictive models were those with the lowest Akaike information criterion, corrected for small sample sizes (Akaike, 1974; Sugiura, 1978). Statistical analysis was conducted using JMP statistical software (ver. 10.0; SAS Institute Inc., Cary, NC, USA).
**Results**

**Daily changes in leaf stomatal conductance**

Intact leaf $g_s$ averaged 196 ± 85 mmol m$^{-2}$ s$^{-1}$ over the measurement period (Fig. 1). Average $g_s$ values differed significantly from December 2009 to early March 2010 ($p < 0.05$) between Tree I (231 ± 83 mmol m$^{-2}$ s$^{-1}$) and Tree II (178 ± 78 mmol m$^{-2}$ s$^{-1}$). In addition, high $g_s$ values were observed in different periods for Trees I and II. For Tree I, $g_s$ peaked on 22 December 2009 and was high from 16 to 23 December 2009, whereas for Tree II, $g_s$ peaked on 29 December 2009 and was high from 28 December 2009 to 12 January 2010, with another relatively high period from 19 to 26 December 2009.

**Trends in stomatal conductance with leaf development and senescence**

A clear trend was observed in $g_s$ related to leaf age (Fig. 2). In summary, $g_s$ was low but stable in newly flushed leaves, and then steadily increased to a peak, followed by a gradual decline. We divided leaf age into six phases based on a statistical interpretation: phase (i), newly flushed and expanding leaves with low-level $g_s$; phases (ii) and (iii), full size semi-mature leaves with low–mid and mid-level $g_s$, respectively; phase (iv), semi-mature leaves with peaking $g_s$; and phases (v) and (vi), mature leaves with mid and low-level $g_s$, respectively (Table 1). We identified three critical leaf ages: day 0, at leaf unfolding (mean ± SD: 113 ± 45 mmol m$^{-2}$ s$^{-1}$); day 10, at the inflection point of the increasing curve of $g_s$ (169 ± 77); and day 17, at the peak level of $g_s$ (268 ± 88). Then, leaf ages were categorised based on whether there was a difference in mean $g_s$ among the selected ages of leaves within the GLM framework. The mean $g_s$ values of phases (i) and (vi) were the same as those of 0-day leaves. Only phase (iv) leaves differed significantly from the 10-day leaves in mean $g_s$. The mean $g_s$ values of phases (iii), (iv) and (v) did not differ from that of 17-day leaves. After 85 days, significant trends were not apparent due to the small number of measurements.

No relationship was observed between $g_s$ and leaf age for leaves older than three months in our study. Of the 46 leaves measured on Tree I in March 2009, 20% remained until March 2010 (i.e., longevity >1 year). Another 40% remained on the branch for at least nine months.

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**Fig. 1** Daily changes in stomatal conductance: a) Tree I, mature individual, b) Tree II, young individual. The data are presented as box-and-whisker plots (median, 25% and 75% quartiles, range). Average $g_s$ values for measurements from December 2009 to early March 2010 differed significantly ($p < 0.05$) between Tree I and Tree II.
long-lived, mature leaves had high \( g_s \) values (186 ± 66 mmol m\(^{-2}\) s\(^{-1}\), \( n = 843 \)) even at nine months after emergence (Fig. 2).

Changes in \( g_s \) in the days before leaf fall are shown in Fig. 3. The \( g_s \) of leaves one day before fall (mean ± SD: 41 ± 18 mmol m\(^{-2}\) s\(^{-1}\), \( n = 30 \)) was significantly lower than over four days before fall (115 ± 51 mmol m\(^{-2}\) s\(^{-1}\), \( n = 25 \), \( p < 0.05 \)). Visual features of leaf senescence were clearly identified 0–2 days before leaf fall, identified with some difficulty three days before fall (88%, \( n = 8 \)), and were rarely identified four days before fall. Conductance gradually decreased in leaves from three days before fall (average <100 mmol m\(^{-2}\) s\(^{-1}\)), whereas the \( g_s \) of leaves five days before fall did not differ significantly in any leaf group (140 ± 54 mmol m\(^{-2}\) s\(^{-1}\), \( n = 24 \), \( p < 0.05 \)). The \( g_s \) values averaged 164 ± 68 mmol m\(^{-2}\) s\(^{-1}\) for mature leaves 5–70 days before leaf fall (\( n = 344 \)).

### Ambient factors related to stomatal conductance

Air temperature and humidity during the measurement period averaged 29.2 ± 1.8°C and 66 ± 6%, respectively. Neither meteorological factor alone was significantly related to \( g_s \). The effect of measurement time (-0800–1300
Leaf phenology effects on transpiration

Of the three ambient factors in a GLM incorporating leaf age and the ambient factors at midday with individual leaves as a random effect, only humidity had a significantly negative effect ($F = 42.2$, $p < 0.0001$, $df = 1$). The coefficient of humidity ($\pm$ standard error) in the selected GLM model was $-2.09 (\pm 0.32)$. Because humidity ranged from 55.2% to 79.0% in 95% of the measurements, the effect of humidity on $g_s$ was estimated to be less than 50 mmol m$^{-2}$ s$^{-1}$. For midday $g_s$ depression, a negative correlation was found between measurement time (hr) and $g_s$ ($F = 12.1$, $p < 0.001$, $df = 1$). The coefficient of measurement time ($\pm$ standard error) was $-3.27 (\pm 0.94)$, which indicates that the effect of measurement time on $g_s$ was estimated as $-3.27$ mmol m$^{-2}$ s$^{-1}$ per one-hour measurement time delay. Regression tree analysis indicated the first splitting criteria for 28 leaves (67% of total leaves measured), whereas no clear trend was observed for the remaining 14 leaves. Higher $g_s$ values were observed in earlier measurement time zones for all the 28 leaves. The first splitting criteria of measurement time averaged ($\pm$ standard error) 1329 hrs ($\pm 47$).

Intact leaf observation

Leaves of various ages were found on the crown of $D.\ costatus$, indicative of intermittent and multiple leaf flushings per year and long leaf longevity (Fig. 4). Branches could be distinguished as several sequential clusters with distinctive gaps and clear differences between new end sections and senescent base sections. Each cluster was composed of a twig and a series of leaves that flushed over short periods.

The total number of leaves on the branches studied increased during the early dry season due to the temporal overlap between new and old leaves, and then decreased at a steady rate due to the shedding of old leaves (Fig. 5). This trend was in agreement with litter trap measurements. The amount of fallen leaves of $D.\ costatus$ in litter traps peaked from late November to early December (Fig. 6a), and the amount of leaf bud scales also peaked during late November (Fig. 6b). A significant proportion of the leaves appeared to have long longevity (sometimes approximately two years). Tree I displayed leaf-flushing phenology slightly earlier than Tree II, with peaks occurring in November and December, respectively. In both study trees, leaf flushing was dominant in the early dry season. Leaf flushing in the late dry season and early wet season was more frequent on Tree I than Tree II (Fig. 5).

During the measurement period, semi-mature leaves were dominant in December, and many leaves were unfolded between November and early December (i.e. the 2009 early dry season cluster). In early March, when the final measurements were taken, bursting buds were also observed on the branches, suggesting that the next cluster would appear in late March or early April.

When a new leaf opened (i.e. the date of the first measurement), the leaf was 84–86% of its full size. Leaves

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Fig. 3 Relationship between time before leaf fall and stomatal conductance (mmol m$^{-2}$ s$^{-1}$). The data are presented as box-and-whisker plots (median, 25% and 75% quartiles, range).
unfolded rapidly, grew to 88–92% of their full size five days after opening, and reached 97–100% of their full size 8–9 days after opening. The expansion of leaf size corresponded well with leaf maturation stage as indicated by the leaf colour observations. When the leaves unfolded to their full size (~8 days after opening), these were considered to have proceeded from young to semi-mature, which corresponded with the start of the rapid increase in conductance. The age at which leaves proceeded from the semi-mature to mature stage varied. The first leaf of a cluster was often small, rapidly reached the mature stage (e.g., 35 days), and often fell earlier (e.g., leaves of the 2009 early dry season cluster on Tree II, Fig. 5b). In contrast, leaves of second order or later were larger, proceeded slowly to the mature stage (> 60 day), and had greater longevity.

Discussion

Leaf-age-related stomatal conductance pattern

We found that leaf age influenced the gₛ of D. costatus during the dry season more than air temperature, humidity, or measurement time during the morning, even though the data included cloudy conditions. Leaf-age-related gₛ displayed an overall pattern of a steep
initial rise followed almost immediately by a gradual decline (Figs 2–3). This pattern is typically found in gs at the maximum photosynthetic rate of leaves with a long lifespan (Čatský et al., 1985), such as evergreen broad-leaved trees in warm-temperate forests (e.g., Castanopsis sieboldii: Miyazawa & Terashima, 2001), in a Mediterranean climate (Escudero & Mediavilla, 2003) and in Xishuangbanna, China, the northern limit of tropical rain forest in Southeast Asia (Cai et al., 2005).

The leaf maximum photosynthetic rate in D. costatus may reach a maximum around peak gs because many tropical tree species, including dipterocarp trees, generally show a strong correlation between maximum photosynthetic rate and gs (Ishida et al., 2006; Kenzo et al., 2012, 2015, 2016; Inoue et al., 2015). Patterns related to leaf age are also found in the photosynthesis rate of the leaves of evergreen plants (Šesták et al., 1985) and deciduous broad-leaved trees in temperate forests (Koike, 1990; Pallardy, 2010). In seasonally dry tropical forests, a gradual decline in photosynthetic capacity related to leaf age has been found in upper canopy leaves in Panama, although a steep initial rise was not detected due to the long measurement span and the fact that measurements were only performed on fully expanded leaves (Kitajima et al., 1997; Kitajima et al., 2002).

The steep initial rise and peak we observed in gs (17 days after unfolding; Fig. 2) is in striking contrast to deciduous dipterocarps in a dry deciduous forest located in Kratie Province of Cambodia (Kenzo et al., 2016). Here, the gs of immature leaves that flushed in the mid-dry season was one-sixth that of mature leaves in the rainy season (6–7 months after emergence) and one-third that of mature leaves in the early dry season (~9 months after emergence) in both D. tuberculatus (Khlong in Khmer) and Shorea obtusa (Phchek in Khmer). One possible reason for the difference in gs of immature leaves between D. costatus and these deciduous dipterocarps is the difference in leaf flushing phenology and edaphic conditions related to soil water deficits. In most tree species in the dry deciduous forest in Kratie Province, except a glabrous Terminalia alata, leaves flush in the mid- to late dry season (Iida et al., 2016; Ito et al., 2017). Low gs in immature leaves of deciduous dipterocarps has been suggested as an adaptation to the severe dry conditions in the mid to late dry season (Kenzo et al., 2016), given the limited soil water available due to the shallow soil (Toriyama et al., 2010). In contrast, the deep root systems of lowland seasonally dry evergreen forests in Cambodia have access to groundwater (Araki & Ito, 2009; Toriyama et al., 2011), as do those in a Brazilian seasonally dry evergreen forests (Xiao et al., 2005). The groundwater supply may mitigate the extent of drought caused by low seasonal precipitation. Significantly lower gs in a deciduous dipterocarp during the mid dry season is indicative of a strong suppression of transpiration (Kenzo et al., 2016). After peaking, gs gradually declined over two months, and then remained at around 60% of the maximum until a few days before leaf fall (Fig. 3). The gs maintained by D. costatus may be indicative of the extent of leaf transpiration even during the dry season in the studied forest. To explicitly test the effects of drought stress on gs and transpiration, future studies should measure leaf water potential and soil moisture conditions in addition to the factors we investigated.

Temporal variation in stomatal conductance associated with leaf phenology

The relatively high gs we observed in different periods for Trees I and II (Fig. 1) was associated with semi-mature leaves flushed in late November (Tree I) or early December (Tree II), as the high gs occurred in leaves 14–30 days after unfolding (phase iv: Fig. 2, Table 1). The difference between Trees I and II in the changing pattern in gs during dry season can be explained by differences
in leaf-flushing phenology (Fig. 5). In addition, we observed bursting buds (Fig. 4) and flushing leaves (Tree I: Fig. 5) at the end of February, which were indicative of two peaks of leaf flushing in *D. costatus*. The *g*ᵣ of newly flushed leaves during the late dry season might have increased around late March to early April if there were less water stress. Deep root systems of lowland seasonally dry evergreen forests in Cambodia have access to significant groundwater (Araki & Ito 2009; Toriyama *et al.*, 2011). Such groundwater supply may mitigate the extent of water stress during the late dry season.

When scaling between single leaf *g*ᵣ measurements and tree-level transpiration, the total leaf area within a tree canopy could be a decisive factor because *g*ᵣ is a measure of the relationship of stomatal opening to diffusion per unit area. The tree-level leaf area of *D. costatus* decreased minimally due to leaf exchange because the total number of leaves per branch remained unchanged (Fig. 5) and leaves expanded rapidly to full size before the peak in *g*ᵣ. Therefore, the relatively constant leaf area of the crown likely has minimal influence on seasonal variation in transpiration. Given the leaf age related *g*ᵣ constant leaf area in the tree canopy, and no restriction from water stress, the dynamic changes in the proportion of young and old leaves within a tree canopy could be the decisive factor influencing tree-level transpiration in the dry season, as suggested by Iida *et al.* (2013). In other words, tree-level transpiration, normalised by ambient air factors, decreases synchronously with leaf exchange, then increases as the *g*ᵣ of flushing leaves peaks, and then gradually decreases. We expect that the tree-level *g*ᵣ of *D. costatus* becomes high at 2–4 weeks after the leaf flushing peak based on the high *g*ᵣ in leaves 14–30 days after unfolding. However, because the number of trees analysed in our study was low (*n* = 2), the generality of our findings should be verified using a larger number of samples in future.

**Ambient factors related to tree-scale transpiration**

Tree-level transpiration often depends on the physical characteristics of the atmospheric environment, including solar radiation, vapour pressure deficit, air temperature and wind speed (Granier & Bréda, 1996; Granier *et al.*, 2000; Pallardy, 2010). Aside for air temperature and humidity which we measured using simple methods, the most important limitation of our study lies in the fact that we could not evaluate the relative importance of these various ambient factors on *g*ᵣ compared with the patterns of *g*ᵣ related to leaf age. Further research is required to determine the strength of environmental controls on *g*ᵣ including its daily variation.

**Conclusions**

Our study indicates that leaf-flushing phenology, as the governing driver of leaf age structure of the tree crown, could be a decisive factor in tree transpiration of *D. costatus* during the dry season. Phenology-driven physiological activity has been reported at a stand scale in a Cambodian deciduous dipterocarp forest (Iida *et al.*, 2016) and in seasonally dry eastern Amazon rainforests (Huete *et al.*, 2006; Hutyra *et al.*, 2007), where leaf flushing during the dry season is associated with an increase in transpiration. Moreover, aggregate canopy phenology, not the seasonality of climate drivers, is the primary cause of photosynthetic seasonality (Wu *et al.*, 2016, 2017). Phenology varies within communities, and the phenology of individuals has a key role in determining how ecosystems are structured and function (Cleland *et al.*, 2007). Additional research on stand-scale leaf phenology in the seasonally tropical evergreen forest is required to determine the dynamic changes in leaf age structure in the crown layers, which could improve our understanding of the eco-hydrological processes of Cambodian forest ecosystems.

**Acknowledgements**

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**References**


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Mammal records and conservation threats in Siem Pang Wildlife Sanctuary and Siem Pang Khang Lech Wildlife Sanctuary, Cambodia

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Abstract

Siem Pang Wildlife Sanctuary and Siem Pang Khang Lech Wildlife Sanctuary form part of a 11,217 km² network of protected areas in Laos, Cambodia and Vietnam, one of the largest protected landscapes in the Mekong basin. Our study contributes to filling a knowledge gap for the two wildlife sanctuaries and consisted of a systematic camera trap survey (comprising 60 camera trap stations and 1,574 trap-nights) in 2016, complemented by an opportunistic camera trap survey (comprising 47 camera trap stations and ≈1,100 trap nights) in 2012–2013 and direct observations of mammals between 2012 and 2016. Including previously published data, seventeen globally threatened medium and large-bodied mammals are recorded from the two contiguous sanctuaries, which contain a species assemblage that reflects the depressed status of mammal populations in Cambodia. Together with other contiguous protected areas, Siem Pang Wildlife Sanctuary may support nationally important populations of Asian elephants and gaur and a globally important red-shanked douc population, whereas Siem Pang Khang Lech Wildlife Sanctuary supports a globally important Eld’s deer population. Threats to the two wildlife sanctuaries include habitat fragmentation and forest conversion to agriculture which are exacerbated by road construction and hunting. The protected landscape they form part of provides sufficient land area to support viable populations of medium and large-bodied mammals. However, the continued persistence of these will require improved management supported by rigorous species and threat assessments to underpin conservation actions.

Keywords
Asian elephant, camera trapping, clouded leopard, hunting, landscape connectivity, red-shanked douc, roads, semi-evergreen forest.

Introduction

Southeast Asia is a biodiversity hotspot (Myers et al., 2000), yet the region is the most biologically threatened large continental area (Schipper et al., 2008; Duckworth et al., 2012). Despite the urgent need to conserve its biodiversity, significant knowledge gaps remain regarding species occurrence at sub-national scales (WWF, 2014; Bryant et al., 2016; Proosdij et al., 2016). This information is crucial to aid identification of viable species sub-populations (Mace et al., 2008; Pimm et al., 2014).

Siem Pang Wildlife Sanctuary (SPWS) and Siem Pang Khang Lech Wildlife Sanctuary (SPKLWS) form part of a network of protected areas in Laos, Cambodia and Vietnam, which together comprise a protected land area of 11,217 km² (UNEP-WCMC & IUCN, 2017). This is one of the largest nominally protected landscapes in the Mekong basin (Souter et al., 2016), yet the only published information on the mammals of the two wildlife sanctuaries is presented in BLCP (2012) and Eames (2014). The former report compiles mammal records which are mainly drawn from reliable unpublished sources, whereas the latter includes some additional records as part of a more general treatment on biodiversity. Prior to the present study, faunal surveys within the two sites primarily focused on the deciduous dipterocarp forests, while semi-evergreen forests remained under surveyed. This was because the deciduous dipterocarp forests support globally irreplaceable populations of five Critically Endangered bird species and a population of the Endangered Eld’s deer Rucervus eldii siamensis, and were therefore prioritised for conservation over semi-evergreen forest (which is unsuitable for these species) (BLCP, 2012). Aside from these works, no systematic surveys had been undertaken of mammals in SPWS and prior to our study only incidental mammal data existed, which comprised opportunistic photographs and sightings of a few species such as the Endangered Germain’s langur Trachypithecus germaini.

The aim of our study was therefore to contribute to filling a knowledge gap on medium and large-bodied mammals present in both wildlife sanctuaries. The study comprised a systematic camera trap survey in SPWS in 2016, complemented by opportunistic camera trap placement and direct observations of mammals in SPWS and SPKLWS in 2012–2013 and 2012–2016 respectively. We report the results of this work and detail conservation threats observed during the field surveys, incorporate previous records of medium and large-bodied mammals in BLCP (2012) and Eames (2014), and discuss the findings in relation to the regional significance of selected mammals and conservation threats present at both sites.
Methods

Study sites

Siem Pang Wildlife Sanctuary (SPWS) covers 66,932 ha and is centred on 14°17’ N, 106°27’ E (Fig. 1). First designated in 2014, SPWS connects a patchwork of contiguous protected areas including Xe Pian National Protected Area, Nam Ghong Provincial Protected Area, Dong Ampham National Protected Area in Laos, Virachey National Park, Veun Sai-Siem Pang National Park, Siem Pang Kang Lech Wildlife Sanctuary in Cambodia and Chu Mom Ray National Park in Vietnam (here collectively referred to as ‘the protected landscape’). SPWS supports mainly semi-evergreen forest at low elevations e.g., <150 m above sea level (a.s.l.). Maximum elevations (350 m a.s.l.) are found close to the border with Virachey National Park. The adjoining Siem Pang Kang Lech Wildlife Sanctuary encompasses 65,389 ha, centres on 14°11’N, 106°13’E and supports mainly deciduous dipterocarp forest with numerous trapeangs (water-holes) and riverine semi-evergreen forest along larger rivers. Minimum elevations of 60 m a.s.l. occur near the Sekong River which increase to 400 m a.s.l. next to the Laos border in the north-western area of the sanctuary (United States Army Map Service, 1967). Our field work at these sites comprised a systematic camera trap survey in SPWS in 2016, complemented by opportunistic camera trap placement in 2012–2013 and direct observations of mammals in SPWS and SPKLWS between 2012 and 2016.

Systematic camera trap survey

Our systematic camera trap survey of medium and large-bodied ground dwelling mammals was undertaken during the dry and early wet seasons (between March and June) in 2016. Camera trapping focused on the semi-evergreen habitat, east of the Sekong River in SPWS. The survey covered an area 106 km² of semi-evergreen forest out of the total of 600 km² of semi-evergreen forest (Fig. 1).
Deployment of camera traps followed principles of the Terrestrial Ecology and Monitoring Network’s vertebrate camera trapping protocol; positioning of camera traps 30 cm above the ground, thereby targeting medium and large-bodied species (Jansen et al., 2014). Sixty camera trap (Bushnell 12MP Natureview Cam Essential HD) sampling points were deployed in three grids of 20 cameras for four weeks per grid. A systematic sampling grid was developed in ArcGIS 10.4.1 (ESRI, 2015), deploying cameras at a density of one camera every 2 km². Camera traps were positioned within 800 m of planned locations, and targeted at animal trails so as to maximize the likelihood of photographing ground-dwelling, trail-following mammal species (Jansen et al., 2014), although this biased against non-ground-dwelling, trail-following mammals. No baits or lures were used. Cameras were configured to take three consecutive photographs per trigger and operated 24 hours per day. The cameras shot in colour during daylight, and in black and white during darkness. Sightings, faeces and photographic documentation of other species were also recorded opportunistically along trails at dawn and during travel to and from camera trap locations.

We calculated the encounter rate per 100 camera trap days for each species photographed by dividing the total number of notionally independent events across all camera stations by the total number of camera trapping days, and multiplying by 100 (Carbone et al., 2001; O’Brien et al., 2003). For a given species, notionally independent capture events at the same camera station were arbitrarily defined as being separated by more than 30 minutes from the previous photograph of what could have been the same ‘event’ (O’Brien et al., 2003). The number of camera trap stations where each species was captured was also recorded.

For the most commonly trapped species—those with more than an arbitrary number of 20 detection events—detection/non-detection at a camera trap station were modelled as a function of environmental covariates collected at the same spatial scale-using binomial generalized linear models (GLMs). A square root transformation was applied to distance to river and relascope scores to reduce skew. Variance Inflation Factors (VIF) and Pearson correlation coefficients were used to assess collinearity among seven predictor variables (Appendix 1). In the event that explanatory variables showed high collinearity (Pearson $r \geq 0.7$ and / or VIF $\geq 3$), the variable with the strongest univariate relationship with the response variable was retained (Zuur et al., 2010). GLMs included the log of camera station-specific effort in days as an offset to account for differences in the number of days that cameras were in operation. Model selection was undertaken using backwards-forwards selection with Akaike’s information criterion (Akaike, 1974; Murtaugh, 2009).

**Opportunistic camera trap survey**

Our opportunistic camera trap survey was undertaken between 2012 and 2013 in SPWS and SPKLWS and consisted of 47 camera trap stations and approximately 1,100 camera trap nights. Encounter rates were not calculated. Camera traps were located across a 75 km² area in riverine semi-evergreen forest along the northern tributaries to the Sekong River, specifically along the smaller O’Khampa and Stung Tin Hiang rivers, and in the block of semi-evergreen forest that extends to the border with Laos. These were placed in natural clearings in the forest, at forest pools (although it was unknown whether these were seasonal or permanent), salt/mineral licks and next to carnivore latrines or where there were signs of wild cattle. Our opportunistic camera trap survey did not focus on trails. Cameras were set approximately one metre above ground throughout the 24 hr cycle and no baits were used.

**Incidental mammal records and threat assessment**

We collated data on mammals observed at SPWS and SPKLWS from 2012 to 2016 and included these in the present study for completeness. These do not include reports from villagers or footprint data. Records of other medium to large sized mammal species from both wildlife sanctuaries in BLCP (2012) and Eames (2014) were also collated. An indication of threats facing mammal populations at both sites was derived from 1) protected area enforcement team reports arising from patrols using Spatial Monitoring And Reporting Tool software (SMART, 2014), 2) quantification of levels of habitat disturbance observed at camera trap stations following Wearn et al. (2013) (Appendix 1), 3) description of the extent of anthropogenic forest use identified from camera trap photographs (per Azlan, 2006; Hossain et al., 2016), 4) opportunistic threat monitoring and documentation of evidence of hunting activity observed during the fieldwork in SPWS.

**Results**

Including data presented in BirdLife International Cambodia Programme (2012) and Eames (2014), 38 species of medium and large-bodied mammal are confirmed to occur in SPWS and SPKLWS (Table 1). This figure includes 28 taxa recorded in BLCP (2012) and Eames (2014) and 10 taxa exclusively recorded during our surveys. It also comprises one Critically Endangered,
Table 1: Medium and large-bodied mammal species recorded to date at Siem Pang Wildlife Sanctuary (SPWS) and Siem Pang Kang Lech Wildlife Sanctuary (SPKLWS). Taxonomy follows IUCN (2018) except for northern yellow-cheeked crested gibbon Nomascus annamensis (after Vu et al., 2010). Record: CT = camera trap record, P = photograph (non-camera trap record of wild animal), S = sighting (without photograph). Source: A = BLCP (2012), B = Eames (2014), C = This study.

<table>
<thead>
<tr>
<th>Species</th>
<th>Status</th>
<th>Record</th>
<th>Source</th>
<th>Site</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sunda pangolin Manis javanica</td>
<td>CR</td>
<td>P¹</td>
<td>A</td>
<td>SPWS?</td>
</tr>
<tr>
<td>Northern treeshrew Tupaiia belangeri</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Northern slender-tailed treeshrew Dendrogale murina</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Pygmy slow loris Nycticebus pygmaeus</td>
<td>VU</td>
<td>P</td>
<td>C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Northern pig-tailed macaque Macaca leonina</td>
<td>VU</td>
<td>S</td>
<td>A, C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Long-tailed macaque Macaca fasicularis</td>
<td>LC</td>
<td>S, CT</td>
<td>A, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Germain’s langur Trachypithecus germainii</td>
<td>EN</td>
<td>S, P², CT</td>
<td>A, B, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Red-shanked douc Pygathrix nemaeus</td>
<td>EN</td>
<td>P</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Northern yellow-cheeked crested gibbon Nomascus annamensis</td>
<td>EN</td>
<td>S</td>
<td>A, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Golden jackal Canis aureus</td>
<td>LC</td>
<td>P</td>
<td>A, B</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Dhole Cuon alpinus</td>
<td>EN</td>
<td>CT</td>
<td>B</td>
<td>SPWS</td>
</tr>
<tr>
<td>Sun bear Helarctos malayanus</td>
<td>VU</td>
<td>CT</td>
<td>B</td>
<td>SPWS</td>
</tr>
<tr>
<td>Yellow-throated marten Martes flavigula</td>
<td>LC</td>
<td>P</td>
<td>C</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Greater hog badger Arctonyx collaris</td>
<td>VU</td>
<td>P³</td>
<td>C</td>
<td>Origin unclear</td>
</tr>
<tr>
<td>Smooth-coated otter Lutrogale perspicillata</td>
<td>VU</td>
<td>CT</td>
<td>B</td>
<td>SPWS</td>
</tr>
<tr>
<td>Large-spotted civet Viverrea megaspiata</td>
<td>EN</td>
<td>CT</td>
<td>B</td>
<td>SPWS</td>
</tr>
<tr>
<td>Small Indian civet Viverrica indica</td>
<td>LC</td>
<td>CT</td>
<td>C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Common palm civet Paradoxurus hermaphroditus</td>
<td>LC</td>
<td>CT</td>
<td>A, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Small-toothed palm civet Arctogalidia trivirgata</td>
<td>LC</td>
<td>S, P</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Leopard cat Prionailurus bengalensis</td>
<td>LC</td>
<td>CT, P</td>
<td>A, B, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Asiatic golden cat Catopuma temminckii</td>
<td>NT</td>
<td>CT</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Mainland clouded leopard Neofelis nebulosa</td>
<td>VU</td>
<td>CT</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Asian elephant Elephas maximus</td>
<td>EN</td>
<td>P⁴</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Wild pig Sus scrofa</td>
<td>LC</td>
<td>CT, P</td>
<td>A, B, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Lesser oriental chevrotain Tragulus kanchil</td>
<td>LC</td>
<td>CT</td>
<td>A, C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Sambar Rusa unicolor</td>
<td>VU</td>
<td>CT</td>
<td>B, C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Eld’s deer Rucervus eldii</td>
<td>EN</td>
<td>P, S</td>
<td>B, C</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Northern red muntjac Muntiacus vaginalis</td>
<td>LC</td>
<td>CT, P, S</td>
<td>A, B, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Gaur Bos gaurus</td>
<td>VU</td>
<td>CT, P, S</td>
<td>B, C</td>
<td>SPWS, SPKLWS</td>
</tr>
<tr>
<td>Banteng Bos javanicus</td>
<td>EN</td>
<td>CT</td>
<td>B</td>
<td>SPWS</td>
</tr>
<tr>
<td>Black giant squirrel Ratufa bicolor</td>
<td>NT</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Variable squirrel Callosciurus finlaysonii williamsoni</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Cambodian striped squirrel Tamiops rodolphi</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Red-cheeked squirrel Dremomys rufigenis</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Berdmore’s squirrel Menetes berdmorei</td>
<td>LC</td>
<td>S</td>
<td>A</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>Giant flying squirrel sp. Petaurista sp.</td>
<td></td>
<td>S</td>
<td>A³</td>
<td>SPKLWS</td>
</tr>
<tr>
<td>East Asian porcupine Hystrix brachyura</td>
<td>LC</td>
<td>CT</td>
<td>C</td>
<td>SPWS</td>
</tr>
<tr>
<td>Siamese hare Lepus peguensis</td>
<td>LC</td>
<td>S</td>
<td>A, C</td>
<td>SPKLWS</td>
</tr>
</tbody>
</table>

¹ Skins and confiscated animals, probably originating from SPWS. ² Photograph of dead animal. ³ Two skins photographed in house in Siem Pang on 9 December 2016. ⁴ Originally identified as P. philippensis on range grounds but P. petaurista is now known to occur in Cambodia as well and therefore cannot be ruled out.
eight Endangered, eight Vulnerable, two Near Threatened and 18 Least Concern mammal species.

**Systematic camera trap survey**

Eleven mammal species were recorded over the course of the systematic camera trap survey in SPWS in 2016 (Table 2). With respect to notionally independent detection events, northern red muntjac *Muntiacus vaginalis* (Least Concern) was the most commonly recorded mammal, followed by lesser oriental chevrotain *Tragulus kanchil* (Least Concern), gaur *Bos gaurus* (Vulnerable) and wild pig *Sus scrofa* (Least Concern).

Modelling of species detection/non-detection showed that the presence of gaur at a camera station was most strongly predicted by the variables semi-evergreen forest habitat (%$D = 36$, $p = 0.002$) and camera trapping grid number (%$D = 12$, $p = 0.008$), with the more northerly grid further from settlements detecting gaur significantly more frequently than the more southerly grids. Similarly, detection of the common palm civet *Paradoxurus hermaphroditus* (Least Concern) was significantly predicted by increasing distance to settlements (Table 3). In contrast, wild pigs were detected significantly more frequently at camera trap stations with higher levels of forest disturbance (Table 3). There were no significant factors influencing detection of northern red muntjac or lesser oriental chevrotain.

Aside from the globally Vulnerable gaur, one globally threatened species not previously confirmed in SPWS or SPKLWS was also recorded during the systematic survey: mainland clouded leopard *Neofelis nebulosa* (Vulnerable). This was camera-trapped in a dry streambed within semi-evergreen forest (14°19’32” N, 106°24’43” E) on 6 April 2016.

**Opportunistic camera trap survey**

Fourteen mammal species were recorded during the opportunistic camera trap survey in SPWS and SPKLWS in 2012–2013. These included four Endangered species (Germain’s langur, dhole *Cuon alpinus*, large-spotted civet *Viverra megaspila* and banteng *Bos javanicus*). At least nine Germain’s langurs were camera-trapped together at a salt/mineral lick along the Stung Tin Hiang River on 30 December 2013 (approximately 14°20’51” N, 106°16’21” E). Dholes were camera-trapped twice at two camera trap stations set along the Stung Tin Hiang River (approximately 14°20’51” N, 106°16’21” E), both on the 25 December and 29 December 2013. One photograph on the 29 December 2013 contained two animals. One large-spotted civet was camera-trapped on 20 January 2013 along the Stung Tin Hiang River (approximately 14°20’51” N, 106°16’21” E) and at least three bantengs comprising one adult male, an adult female and a juvenile were camera-trapped on 7 April 2013 between the Stung Tin Hiang and O’Kul rivers (14°19’53.8” N, 106°16’25.2” E).

**Table 2** Medium and large-bodied mammal species recorded during the systematic camera trap survey in Siem Pang Wildlife Sanctuary. The number of events is the number of notionally independent captures. Encounter rate is the number of events across all camera stations divided by the total number of camera trapping days, multiplied by 100.

<table>
<thead>
<tr>
<th>Species</th>
<th>No. of events</th>
<th>Encounter rate</th>
<th>No. of stations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Northern red muntjac <em>Muntiacus vaginalis</em></td>
<td>100</td>
<td>6.35</td>
<td>40</td>
</tr>
<tr>
<td>Lesser oriental chevrotain <em>Tragulus kanchil</em></td>
<td>35</td>
<td>2.22</td>
<td>10</td>
</tr>
<tr>
<td>Gaur <em>Bos gaurus</em></td>
<td>31</td>
<td>1.97</td>
<td>7</td>
</tr>
<tr>
<td>Wild pig <em>Sus scrofa</em></td>
<td>22</td>
<td>1.4</td>
<td>17</td>
</tr>
<tr>
<td>Common palm civet <em>Paradoxurus hermaphroditus</em></td>
<td>15</td>
<td>0.95</td>
<td>14</td>
</tr>
<tr>
<td>East Asian porcupine <em>Hystrix brachyura</em></td>
<td>10</td>
<td>0.64</td>
<td>7</td>
</tr>
<tr>
<td>Northern pig-tailed macaque <em>Macaca leonina</em></td>
<td>5</td>
<td>0.32</td>
<td>4</td>
</tr>
<tr>
<td>Leopard cat <em>Prionailurus bengalensis</em></td>
<td>2</td>
<td>0.13</td>
<td>2</td>
</tr>
<tr>
<td>Long-tailed macaque <em>Macaca fascicularis</em></td>
<td>2</td>
<td>0.13</td>
<td>2</td>
</tr>
<tr>
<td>Mainland clouded leopard <em>Neofelis nebulosa</em></td>
<td>1</td>
<td>0.06</td>
<td>1</td>
</tr>
<tr>
<td>Sambar <em>Rusa unicolor</em></td>
<td>1</td>
<td>0.06</td>
<td>1</td>
</tr>
</tbody>
</table>
Four Vulnerable species were also recorded, namely smooth-coated otter *Lutrogale perspicillata*, sun bear *Helarctos malayanus*, and sambar *Rusa unicolor*. Photographs of a smooth-coated otter, possibly the same animal, were obtained from one camera-trap station on 26 and 28 November 2013 along the Stung Tin Hiang River (approximately 14°20'51" N, 106°16'21" E). Sun bears were camera-trapped on 4 and 5 January 2013 and 23 February 2013 along the O'Kul, west of the Sekong River (approximately 14°17'24.42" N, 106°16'20.60" E). Following the systematic camera trapping, sun bears were also recorded in opportunistically deployed camera traps east of the Sekong River on 21 July and 5 November 2016 (14°21'87" N, 106°22'89" E). Gaur were camera-trapped during the opportunistic survey as follows: one on 27 January 2012, four on 20 April 2013, one on 11 May 2013 and three on 22 May 2013 (all at approximately 14°20'51" N, 106°16'21" E). Following the opportunistic survey, one gaur was photographed on 27 January 2012, four on 20 April 2013, one on 11 May 2013 and three on 22 May 2013 (all at 14°18'37" N, 106°18'19" E). Sambar were photographed at nine camera trap stations (at approximately 14°20'51" N, 106°16'21" E) between December 2012 and January 2013.

Incidental mammal records

Nineteen species were incidentally recorded during fieldwork in the two wildlife sanctuaries between 2012 and 2016 (Table 1), including four Endangered taxa: red-shanked douc *Pygathrix nemaeus*, Germain’s langur, northern yellow-cheeked crested gibbon *Nomascus annamensis* and Asian elephant *Elephas maximus*. Two Vulnerable taxa, pygmy slow loris *Nycticebus pygmaeus* and northern pig-tailed macaque, were also recorded.

Red-shanked doucs were identified from photographs of a dead individual (14°20'59. 42" N, 106°20'36.12" E within the camera trap block, east of the Sekong River) found on 14 April 2016 and a live captive juvenile observed at the 101 Army station within SPWS on the west bank of the Sekong River (14°19'24.41" N, 106°18'36.16" E: Fig. 2). Douc langurs, identified as red-shanked, were also observed (two adults and one juvenile) in semi-evergreen habitat close to the Stung Mulu River (14°19'34.86" N, 106°22'59.19" E) on 11 April 2016, as were two feeding in association with a northern yellow-cheeked crested gibbon (14°20'59.36" N, 106°20'35.95" E) on 13 April 2016. Footprints and dung boluses of Asian elephant were detected in three areas between 10 and 12 June 2016 (exact locations are not reported here due to security concerns). Troops of Germain’s langurs were observed and photographed on most field excursions between 2012–2016 along the Sekong River, including one on 2 January 2013 and three together on 22 October 2012. The largest was a troop of 40 observed on 29 March 2015 (at 14°18'21.08"N, 106°20'1.32"E). Pygmy slow lorises were sometimes confiscated and released by the protected area enforcement team including one released on 30 May 2015 and four in March 2016. Two northern pig-tailed macaques were observed along the Sekong River on 29 March 2015 (near 14°18'21.08" N, 106°20'1.32" E).

Conservation threats

Threats to mammals documented in SPWS and SPKLWS during the study period included habitat loss and fragmentation due to road construction and subsequent forest conversion to agriculture and settlement along roads and riverbanks, together with hunting and live animal capture.

Connectivity within the protected landscape is threatened by a road construction project that began in 2016 along the Cambodia-Laos border between SPWS and Xe Pian National Protected Area in Laos. By January 2018, construction of a road bridge across the Sekong River (at 14°26'5. 11" N, 106°19'48. 98" E) was well advanced and road construction continues along the international frontier along the northern boundary of Virachey National...
Park (Fig. 1). Construction of a second road bridge ca. 3 km south of this point began in 2018. A road has also been built from Siem Pang town northwards and almost parallel in places to the Sekong River through SPKLWS and SPWS to both bridge sites. Thus, SPKLWS and the portion of SPWS west of the Sekong are now completely encircled by roads.

A dispute with Laos in 2017 associated with border road construction has resulted in a greater number of Cambodian army personnel being stationed in SPWS. Army personnel have cleared forest to establish a camp along the road (at 14°26’N 106°19’E) and proposed that ca. 5,000 ha of land within SPKLWS be set aside for military facilities. SMART reports for SPKLWS show a consistently high level of attempted land demarcation and clearance along these roads and the Sekong River, with 101 instances reported from 352 patrols in 2015, 83 instances from 397 patrols in 2016 and 122 instances from 379 patrols in 2017.

Hunting activity recorded by field teams during the systematic camera trapping survey in SPWS included gunshots heard on all six deployment expeditions and two snare lines comprising 22 snares were encountered and removed by field teams. People not associated with the systematic survey were camera-trapped at 14 of the 60 camera trap stations and were detected across all three camera trapping grids. These were in the company of hunting dogs in 19 out of 33 notionally independent records, including one photograph of people in possession of a dead lesser oriental chevrotain. Field teams during the systematic survey also encountered a community member with hunting dogs in possession of a dead common palm civet and a group in possession of a dead wild pig. People were also recorded regularly during the opportunistic camera trap survey in 2013, although none were accompanied by hunting dogs. Snare lines and other traps were not recorded during the opportunistic survey, nor during the earlier camera trap survey.

Live mammal capture, documented through opportunistic threat monitoring by field teams, included the first confirmed case of a red-shanked douc being held in captivity in SPWS (Fig. 2). On two occasions in 2015, single juvenile long-tailed macaques were observed on public transport from Siem Pang town. A pygmy slow loris and a juvenile leopard cat *Prionailurus bengalensis* were also confiscated by the protected area enforcement team in 2015 (BLCP, 2015). The same enforcement team also confiscated a live juvenile male sun bear from villagers who reportedly obtained the animal in SPWS in July 2018.

**Discussion**

The severe threats facing Southeast Asia’s mammal community have resulted in many sites losing their top native predators and other medium and large-bodied mammals persisting at very low densities (Steinmetz et al., 2006). This pattern was found in SPWS with tiger *Panthera tigris* and leopard *P. pardus* both unrecorded in our survey, although both are reported to have been present historically in SPKLWS (Mem Mai, pers. comm. 2018). These absences are consistent with recent camera trap studies of other large protected landscapes in Cambodia (Gray et al., 2017; Suzuki et al., 2017), although the latter were neither extensive or of sufficient duration to rule-out their continued presence. Despite significant survey effort, other threatened mammals including Asian elephant and mainland clouded leopard were only recorded on a single occasion despite some 600 km² of seemingly suitable semi-evergreen forest habitat in SPWS. As such, SPWS contains an assemblage of medium and large-bodied mammal species which reflects the depressed status of mammal populations in Cambodia. However, the continued presence of 17 globally threatened medium and large-bodied mammal species and the geographic location of SPWS and SPKLWS within a continuous network of 11,217 km² of protected areas...
suggests these sites have potential regional importance as a refuge for wide ranging mammals.

Remarks on selected mammal species

The Sunda pangolin *Manis javanica* (Critically Endangered) was not detected in our camera trapping surveys. This may be a reflection of the high hunting pressure pangolins face throughout their ranges, partly arboreal, non-trail-following nature, camera trap placement and/or the duration of the survey. Previously skins had been recorded in houses in Siem Pang town (BLCP, 2012) and from live animals confiscated from hunters in SPKLWS (which was assumed to be the source of the animals).

Our records of the red-shanked douc extend its global range west from the first documented record for Cambodia by 50 km. We identified the species based on photographs of the remains of an individual found shot and a juvenile held in captivity, both with pronounced red pelage on the legs (Fig. 2), but acknowledge that they might be hybrid forms of unknown genetic provenance. The largest known population of red-shanked doucs are in Laos (Coudrat et al., 2012). The species was recorded until 1999 in Nam Ghong Provincial Protected Area and Dong Ampham National Protected Area in Laos (Timmins & Duckworth, 1999), part of the protected landscape considered in our study. Recent reports of the species from these protected areas are not available, although Coudrat et al. (2012) suggest they may retain a fairly large population characterized by relatively high genetic diversity, which could include introgression with other douc species. Given that these sites are connected to two protected areas in Cambodia where the genus is confirmed, the protected landscape may be an important stronghold for the red-shanked form in the southern part of its range. Further surveys across the landscape are required to confirm the status of this population.

Germain’s langur appears to be restricted to riverine tracts of semi-evergreen forest in SPWS and SPKLWS and has yet to be documented beyond this ecotone. The species is largely arboreal but does come to the ground to visit mineral licks. For example, Eames (2014) includes a photograph of a Germain’s langur at a mineral lick on the junction of the Sekong and O’Khampa rivers. The species is known from 17 protected or proposed protected areas including six in Cambodia (Mittermeier et al., 2013). Our study recorded sightings of northern yellow-cheeked crested gibbon (Endangered), which was previously documented in SPWS (BLCP, 2012) and is known from along the Stung Tin Hiang River. It has also been recorded from several points along the recently constructed border road in the north-west of SPWS (Jonathan C. Eames, unpublished data). The species remains widely distributed in SPWS, both west and east of the Sekong River and is known from 18 protected or proposed protected areas including Virachey National Park in Cambodia (Mittermeier et al., 2013).

We erroneously anticipated our systematic camera trapping might record dhole and large-spotted civet as both inhabit semi-evergreen forest (in addition to deciduous dipterocarp forest and mixed deciduous forest in the case of dhole). Their detection east of the Sekong River could possibly have been prevented by a naturally low density or reduced density as a result of snaring, disease transmission (rabies and distemper) from domestic dogs, or the short duration of the camera trapping study. Our few records preclude assessment of the potential significance of local populations of these species. Detection of the mainland clouded leopard in SPWS was anticipated due to extensive areas of suitable habitat and as snaring levels appear to remain relatively low. The species has experienced declines across its range, including Myanmar, Vietnam (Wilcox et al., 2014; Grassman et al., 2016) and Laos (Duckworth et al., 2014). In Cambodia, the species has recently been camera-trapped in Virachey National Park (McCann & Pawlowski, 2017), in three protected areas in the eastern plains of Cambodia (Gray et al., 2012) and in the Cardamom Mountains and Chhep Wildlife Sanctuary (Gray et al., 2017; Suzuki et al., 2017). Further survey work is required to determine the status of the species nationally.

Asian elephants were previously documented in Siem Pang District, although it is unknown whether this record was from the area now encompassed by SPWS (Desai et al., 2002). It was also recently recorded in the Yak Yeuk grassland area near the Laos border in Veun Sai District, Stung Treng Province and at other locations in Virachey National Park, which adjoins SPWS (Pawlowski & McCann, 2017). We are aware of historical reports of captive Asian elephants brought through Xe Pian National Protected Area up to and over the Cambodia border in 1992–1993 (William Duckworth, pers. comm. 2018), but know of no current or historical records of captive elephants held in Siem Pang District. Since elephants have not been used for logging in Cambodia within the authors memory, we regard the recent records from Virachey National Park as wild elephants and because Veun Sai District adjoins Siem Pang District, our records likely represent the first confirmed use of SPWS by wild elephants. A minimum area for long-term elephant conservation is 4,400 km² (Sukumar, 1992) and four protected landscapes meet this requirement in Cambodia: the Eastern Plains Landscape (300 individuals), the Cardamom Mountains (175), the...
Northern Plains Landscape around Chhep wildlife Sanctuary (5) and Virachey-Siem Pang protected landscape (unknown). The total wild population in Cambodia is estimated at 400–600 individuals, whereas 500–1,000 are estimated in Laos, 70–150 in Vietnam and 2,500–3,200 in Thailand (Sukumar, 2003; Maltby & Bourchier, 2011; Gray et al., 2014). Thus, while the little-known population in the Virachey-Siem Pang area may have limited regional significance, it may be important for supporting the precariously low national population.

Although not recorded by our camera trapping surveys, Eld’s deer has been regularly recorded in SPKLWS since 2003 when a population likely larger than 50 animals was reported (BirdLife International Cambodia Programme, 2012). This is somewhat expected as the species largely occurs in deciduous dipterocarp forest within its Southeast Asian range and SPKLWS contains large tracts of this habitat. Eld’s deer are reported regularly from areas of open, or park-like deciduous dipterocarp forest in the central and southern parts of the site (BLCP, 2015–2018) and are most conspicuous during the rut in April. Fifty different animals were recorded between 16–19 April 2015, lesser numbers in 2016 and 2017 and a survey in 2018 counted 39 different animals (Eames, 2018). While no conclusions can be drawn regarding population trends due to inconsistencies in survey methods, we regard SPKLWS as a priority site for the Eld’s deer on the basis that no site in Cambodia is known to hold more animals. A systematic study of the population at SPKLWS is therefore a conservation priority and research has recently begun.

Because banteng show a preference for deciduous dipterocarp and mixed deciduous forests in the Mekong Basin, we would not have expected to detect the species in the semi-evergreen forests of SPWS. Our camera trap records in 2013 remain the most recent for the site. Gaur were the third most frequently trapped mammal during our systematic camera trap survey. This species was previously camera-trapped in SPWS west and east of the Sekong River, and in SPKLWS. The only daytime photographic record in SPKLWS is of a male on 28 May 2015 (Anon. 2015). Although detected by camera trapping studies across several protected areas in the Cardamom Mountains landscape (e.g., Gray et al., 2017), the conservation status of the Cambodian population has yet to be determined. Sambar was only detected once during the 2016 camera trap study, which is surprising given the higher detection rate of gaur. However, it was detected at nine camera trap stations during the 2012–2013 camera trap survey.

Conservation threats

Roads can have devastating impacts on biodiversity, acting as new frontiers for development and fragmenting landscapes (Perz et al., 2008; Laurence et al., 2009). Once complete, the road now being built along the Cambodia-Laos border will separate Xe Pian National Protected Area from SPWS and Virachey National Park (Khy, 2017; McCann, 2017). As such, road construction poses the single greatest threat to mammal populations in SPWS and SPKLWS. This is due to the cascade of secondary threats, which result from increased access to the forest, including unregulated settlement and land clearance (Cropper et al., 2001).

The combination of 1) extensive anthropogenic presence in SPWS, 2) hunting activity, 3) species modelling of commonly detected species (gaur and common palm civet) showing higher detection rates further from villages, and 4) absence of hunting sensitive species such as large predators, suggest that some species are being hunted at unsustainable levels. While detections of wild pig did not show the same spatial pattern, this is likely due to its high reproductive rate which makes it more resilient to hunting pressure (Oliver & Leus, 2008).

Our data provides a cursory insight into hunting behaviour. We recorded firearm use, which is of particular concern for large-bodied mammals and primates. For instance, red-shanked doucs are commonly hunted for consumption at the 101 Army station on the west bank of the Sekong River in SPWS (Eames, 2014). On 5 May 2018, a report was received of a dead gaur in SPWS, which upon investigation was found to have been shot by army personnel (Anon. 2018).

In contrast to the high incidence of firearm use, our seemingly low (although anecdotal) detection of snares compared to other sites in Southeast Asia (Harrison et al., 2016) may explain the continued persistence of relatively healthy populations of mammals commonly snared, particularly gaur. This may not actually be the case however because as most law enforcement patrols are undertaken on motorbike, snare lines set in cover are likely to be missed. In addition, while collection of non-timber forest products is permitted within the wildlife sanctuaries, more than half of the people photographed in SPWS were accompanied by hunting dogs. While it was not possible to infer from our photographs whether the dogs had been actively hunting, this raises a cause for
concern and suggests research into hunting practices and social norms is needed.

Study limitations

Our systematic camera trap sampling strategy was biased towards ground-living medium to large bodied mammal species that use trails (Wearn et al., 2013). It was recognised at the design stage that our approach would not capture the full mammal community of SPWS and would exclude arboreal species, most semi-arboreal species, species that actively avoid trails and most ambush predators. For example, our methods were not suitable for recording primates. We also acknowledge that the short duration of the survey limited detection of species, especially taxa that avoid trails. An alternative approach would have set more cameras in a wider variety of microhabitats over a longer period, although this was by limited funds available. Nonetheless, some species may have been under reported, or reported as absent when they actually occur at very low density.

Opportunistic camera trapping in 2012 and 2013 recorded several globally threatened mammal species which were not detected by the systematic camera trapping in 2016. Although variations in hunting pressure and habitat may explain some of these differences, camera trap placement was likely also a significant factor. For example, we would not have detected smooth-coated otter had a camera-trap not been attached to a wooden stake that was then driven into a sandbank facing a latrine. Such techniques are rarely used in conventional systematic camera trapping, although targeted placement reflecting the known ecology of a species is valuable where the objective is to document rare and low-density taxa.

Finally, our threat data is largely anecdotal and provides an indication of the threats present, rather than a rigorous threat assessment. To compare the severity of different threats, their recorded incidences should be standardised in terms of observer research effort, such as number of incidents per patrols. This could be achieved by more systematic planning of threat data collection and analysis using SMART software. This would also facilitate comparisons with other sites in the region.

Prognosis

The assemblage of medium and large-bodied mammal species present in SPWS and SPKLWS reflects the depressed status of most remaining mammal populations in Cambodia. The severe threats facing the site have likely reduced mammal species in-situ to remnant populations.

Although a report on the environmental impacts of the Cambodia-Laos border road has been submitted to the Government of Cambodia (Eames, 2016), road development has not halted. Conservation action to limit the impact of this road must therefore employ a triage approach following principles of the mitigation hierarchy (SCBD & UNEP-WCMC, 2012). Priority mitigation actions would be 1) ban settlement in SPWS and SPKLWS to maintain connectivity across the landscape; and 2) identify and prioritise protection of core areas used by of Asian elephant, gaur and Eld’s deer within the protected landscape to minimise the impact of human presence on their movement (Songhurst et al., 2016).

The presence of military installations in SPWS makes the army a powerful actor that must be constructively engaged to reduce encroachment and hunting and improve protected area management. Strengthened enforcement operations are required to combat encroachment and the prevalent use of firearms (Lunstrum, 2014; Gray et al., 2016). Army border patrol teams could provide one such source of support. Hunting poses an increasing threat to mammals and other wildlife in SPWS and SPKLWS. Understanding and dismantling local trade networks—where villagers sell bush-meat to local middlemen who in turn sell this, which is then sold on Siem Pang and Stung Treng markets—is also required. Indigenous Kavet communities living along the border of SPWS and Virachey National Park engage in hunting and have a tradition of animist beliefs and culture closely associated with the forest. Consequently, understanding social norms, particularly hunting behaviour and meat-consumption preferences will be key to developing management strategies aligned to local values that successfully build community support for conservation (Infield, 2001; Steinmetz et al., 2006).

Sufficient land area is now included in the protected landscape and nationally in Cambodia to conserve viable populations of remaining medium and large-bodied mammals. However the protected estate is becoming increasingly degraded and depleted of harvest sensitive species (Petersen et al., 2015). To strengthen mammal conservation, attention should turn from expansion of the protected area estate to increased investment in existing protected areas (Watson et al., 2014). Ultimately, the persistence of medium and large-bodied mammals across the protected landscape will depend on building a coalition of actors with the political weight, economic resources and vision needed to conserve the integrity of the landscape.
Acknowledgements

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**Appendix 1 Habitat variables recorded at each sampling location during the systematic camera trap survey in Siem Pang Wildlife Sanctuary**

<table>
<thead>
<tr>
<th>Variables</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Distance to settlements</td>
<td>Straight line distance to the nearest permanent settlement calculated in ArcGIS 10.4.1 (ESRI, 2015).</td>
</tr>
<tr>
<td>Distance to river (km)</td>
<td>Straight line distance to the Sekong river calculated in ArcGIS 10.4.1 (ESRI, 2015).</td>
</tr>
<tr>
<td>Grid</td>
<td>Three camera trap grids were established running from south to north. Each camera trap station was assigned to a grid.</td>
</tr>
<tr>
<td>Habitat type</td>
<td>Four habitat types were recognised: semi-evergreen forest, deciduous dipterocarp forest, bamboo stands, dry river beds. Each habitat type was categorised as a separate binary variable (present/absent).</td>
</tr>
<tr>
<td>Average tree height (m)</td>
<td>A laser rangefinder (Nite Hawk Pin Predator 400) was used to estimate the height of four trees at each listening station. Tree selection was randomized by selecting tree stems closest to bearings of north, south, east and west from the camera trap station. The average of these trees was then calculated.</td>
</tr>
<tr>
<td>Tree density (m²/ha)</td>
<td>A relascope (Gove et al. 2001) was held at eye level 53cm from the researcher whilst a 360° rotation was made about the central position of the spool track. The number of trees viewed as larger than the 1cm opening in the relascope was counted and the number multiplied by two to give an estimate of tree basal area per hectare (m²/ha).</td>
</tr>
<tr>
<td>Forest disturbance score</td>
<td>A four level categorical variable from low to high forest disturbance: 1 = No evidence of recent logging; 2 = Occasional single tree stumps and single trees felled; 3 = Frequently encounter felled trees, some evidence of trails cleared for vehicle access; 4 = Frequently encounter felled trees grouped into piles, recent trail clearance for vehicle access, land clearance and evidence of forest conversion with intent of settlement, recent/active logging camps.</td>
</tr>
</tbody>
</table>
Aquatic Polyphaga (Insecta: Coleoptera) from Kampong Speu Province, Cambodia

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Abstract

We present the results of a faunistic inventory undertaken on aquatic Polyphaga beetles as part of a workshop organised by the BIO-PHIL project in Cambodia. Three small rivers and a pond in Kampong Speu Province were sampled in July 2018. Twenty-one taxa were collected during the sampling and identified to species or morphospecies level. These are described with comments on habitat, distribution and further remarks, where applicable. A checklist of the genera and species of aquatic Dryopidae, Elmidae, Hydraenidae, Hydrophilidae, Psephenidae and Spercheidae of Cambodia is presented which includes new records for the country.

Keywords

Biodiversity survey, checklist, Dryopidae, Elmidae, Hydraenidae, Hydrophilidae, Psephenidae, Spercheidae, taxonomy, water beetles.

Introduction

Cambodia forms part of the Indo-Chinese sub-region of the Indo-Burma biodiversity hotspot (Wikramanayake et al., 2002). Most of its territory belongs to the ‘Greater Mekong’ region, so called because the river crosses the country in a north-south direction. Until recently, Cambodia was rarely the focus of entomological research. This changed after the country became an ASEAN member state, as endeavours such as the Cambodian Entomology Initiatives (CEI) at the Royal University of Phnom Penh (RUPP) began surveying the presumably rich insect diversity in 2015. Short-term projects such as the Scientific Consortium for Interdisciplinary Biodiversity Research have also supported the creation of scientific networks with Cambodian scientists, with results in aquatic entomology and taxonomy (Kodada et al., 2017; Zettet et al., 2017). Recently, the CEI team joined the Biodiversity Teaching in a Philippine-Cambodian-German Network (BIO-PHIL) funded by the German Academic Exchange Service.

A BIO-PHIL workshop on field sampling and morpho-taxonomy was delivered at the RUPP in July 2018 (Fig. 1A). Twenty students and instructors from affiliated institutions participated. The practical sessions, including a rapid field survey, focused specifically on aquatic beetles because occurrence and distribution data of Cambodian aquatic beetles are very scarce compared to the neighbouring countries of Laos, Thailand and Vietnam. For example, among the major families of aquatic Polyphaga, only 22 species of Hydrophilidae and no species of Dryopidae, Elmidae, Psephenidae or Hydraenidae were reported from Cambodia (Hansen, 1998, 1999; Short & Hebauer, 2006; Short & Fikacek, 2011; Jäch et al., 2016). This contrasts with more than 87 species of Hydrophilidae, Hydraenidae and Elmidae recorded from Thailand, approximately 130 from Vietnam and 107 from the Philippines (Hansen, 1998, 1999; Short & Hebauer, 2006; Short & Fikacek, 2011; Freitag et al., 2016; Jäch et al., 2016; Vidal et al., 2017). This is very unsatisfactory given the fact that many aquatic beetles have high potential as bioindicators (Freitag et al., 2016) and for use in environmental impact assessments (Balke et al., 1997).

This paper presents new faunistic data for Cambodia on aquatic Polyphaga collected during field sampling undertaken as part of the above workshop.

Methods

Field sampling

Aquatic beetles were collected at four main sites in Kampong Speu Province (Figs 1B–D & 2) between 11–13 July 2018, as follows:

- Cam3P—Vegetated pond, 0.1 km north of Chambok Community, secondary vegetation (90 m above sea level (a.s.l.), 11°23’00” N, 104°06’50” E);
- Cam3R—Chambok River, 1.83 km from Chambok Community, secondary forest (240 m a.s.l., 11°21’58” N, 104°06’17” E);
- Cam4—Kokie Waterfall, secondary forest remnants (110 m a.s.l., 11°12’11” N, 104°03’49” E);
- Cam5—Waterfall at Kirirom National Park, rural area partly with secondary forest (640 m a.s.l., 11°20’26” N, 104°02’14” E).

The beetles were obtained by checking various microhabitats with hand-nets and manual examination of submerged substrates (Fig. 1D) as outlined in Freitag (2015). Specimens collected in this way are indicated by ‘M’ at the end of the label and the microhabitats sampled were as follows: a—sand/gravel deposits in littoral run areas (with distinct current) of a stream; b—sand/gravel deposits in calm littoral pool areas of a stream; c—bottom gravel in a run/riﬄ e; e—leaf litter/ coarse particulate organic matter (CPOM) in side pools adjacent to a stream; f—submerged wood; g—submerged rock surface in a run/riﬄ e; h—littoral root packs/grass bunches in a run/riﬄ e; p—rock surface in calm pool areas of a stream.

Black light traps were also used to sample between 1800 and 2100 hrs at the Cam3P and Cam3R sites. Specimens collected with these traps are indicated by ‘L’ at the end of the label. An emergence trap, as described in Freitag (2004), was also installed at the Cam3R sampling site (Fig. 1B) for one week, but did not yield any specimens as the device was flooded and partly damaged by heavy monsoon rains.

Specimen preservation and identiﬁ cation

Specimens were preserved in 96% ethanol or glued on entomological card-mounts when dissected for identification. The material we list is deposited at the CEI collection in the RUPP. Original descriptions or re-descriptions of taxa, regional faunal guides (Komarek, 2003; Shepard & Sites, 2016) and comparisons to type material at the Natural History Museum of Vienna, Austria (NMW) were used to identify specimens to species when possible. Specimens were examined with Olympus SZ60 stereomicroscopes. Specimens of selected taxa were photographed under a Zeiss Axio Zoom V16 microscope using diffuse LED lighting and a Canon 5D Mark II SLR. Photographs were taken at various focus layers and subsequently stacked using Zerene Stacker software (Zerene Systems, Washington, USA).
Fig. 1 Laboratory work and collecting sites: A) BIO-PHIL workshop participants working on sample identification at Royal University of Phnom Penh, B) Cam3R sampling site, C) Cam4 sampling site, D) Participants sampling at Cam5 site.

Fig. 2 Collecting localities in Kampong Speu Province, Cambodia.
## Results

Twenty-one taxa were collected during the sampling and identified to species or morphospecies level. These are described with comments on habitat, distribution and additional remarks, where applicable.

### HYDRAENIDAE (Minute Moss Beetles)

**Hydraena (Hydraenopsis) spp.** (Fig. 3A)

**Material:** Sp. 1, one male (Cam3R, b, 11.7.2018, M), one female (Cam4, p, 12.7.2018, M); Sp. 2: one male (Cam3R, b, 11.7.2018, M).

**Habitat:** Our specimens were collected from littoral sandy deposits and rock surfaces in calm pool sections of streams, respectively. *Hydraena* Kugelann, 1794 species are most commonly collected from such shallow littoral microhabitats in slow-flowing or stagnant waters, but also among leaf litter and other CPOM deposits. However, some species are also adapted to special habitats like helocrenes, hygropetric rocks and even trapped leaf litter in fast-flowing streams (Freitag, 2015).

**Remarks:** These two species of the subgenus *Hydraenopsis* Janssens, 1972 are unnamed and our report is the first for Cambodia. The genus is mega-diverse in the Oriental Realm (Jäch & Balke, 2008), but commonly overlooked due to species’ small size, patchy distribution and hidden habitats (Freitag, 2014). It would not be surprising if *Hydraena* turns out to be the most speciose genus of aquatic beetles in Cambodia, as in the Philippines (Freitag *et al.*, 2016). However, only six species of *Hydraena* are known from the Indo-Chinese sub-region, all of them from Vietnam (Hansen, 1998). We refrain from formally describing our material until more is available.

### HYDROPHILIDAE (Water Scavenger Beetles)

**Hydrophilinae**

**Berosus (Euoplurus) chinensis Knisch, 1922** (Fig. 3B)


**Material:** One male, one female (Cam3P, 12.7.2018, L).

**Distribution:** *Berosus chinensis* is widely distributed from Afghanistan in the West to China (Hong Kong) in the East including Thailand and Vietnam (Hansen, 1999). Our record is the first from Cambodia.

**Habitat:** Our two specimens were caught by a black light trap near a pond which is a typical microhabitat for *Berosus* spp., which generally inhabit sun-exposed, stagnant water bodies (Freitag, 2015).

**Berosus (s.str.) pulchellus MacLeay, 1825** (Fig. 3C)


**Material:** One male, two females (Cam3P, 12.7.2018, L).

**Distribution:** *Berosus pulchellus* is widely distributed in the Oriental Realm and neighbouring areas of the Australasian, Afrotropical and Palearctic Realms (Hansen, 1999). This is the first record from Cambodia.

**Habitat:** See respective remarks for *Berosus chinensis*.

**Paracymus mimicus Wooldridge, 1977**

*Paracymus mimicus* Wooldridge, 1977, 123 (orig. descr.).

**Material:** One male (Cam3P, 12.7.2018, L).

**Distribution:** *Paracymus mimicus* is known from Laos and Thailand (Wooldridge, 1977). This is the first record from Cambodia.

**Material:** The single specimen was caught by a black light trap near a vegetated pond and its preferred microhabitat is therefore unknown. *Paracymus* spp. are usually found in shallow littoral sand and gravel deposits or under stones in stagnant water bodies and pool sections of streams (Freitag, 2015).

**Pelthydrus cf. vitalisi Orchymont, 1926** (Figs 3D–E)


**Material:** One male (Cam4, f, 12.7.2018, M).

**Distribution:** The presumed complex of species currently recognized as *Pelthydrus vitalisi* s.l. is known from China (Hong Kong & Taiwan), Vietnam, Thailand, Malaysia and Indonesia (Sumatra, Java, Bali & Lombok). The type locality is in northern Vietnam. This is the first record of *P. vitalisi* s.l. from Cambodia.

**Diagnosis of the sole male from Cambodia:** Body ovoid elongate and dorsally moderately convex, calculated length of pronotum and elytra (CL) 2.2 mm, body length approximately 2.4 mm, maximum elytral width (EW) 1.1 mm, 2.0 times as long as wide (CL/EW).

Dorsal colouration (Fig. 3D) predominantly chestnut brown. Head rectangular, distinctly narrower than pronotum, head width including eyes 0.65 mm; interocular distance 0.50 mm; lateral portion indistinctly microreticulate. Pronotum sub-rectangular (Fig. 3D), pronotum length along midline (PL) 0.5 mm, maximum pronotum width (PW) 1.0 mm, 2.0 times as long as wide (PL/PW); lateral margin finely dentate, evenly convex, broadest at
middle. Elytra (Fig. 3D) elongate, lateral margin finely dentate, elytral length (EL) 1.7 mm, ca. 3.0 times as long as wide (EL/EW), widest at anterior 0.3; apices separately rounded. Venter predominantly with plastron pubescence. Aedeagus as in Fig. 3E.

Remarks: Our specimen is very similar to other material from the sub-region determined as *P. vitalisi* by Schönmann (material in NMW). However, it varies slightly from the redescription and from the holotype by some characters: aedeagus (Fig. 3E) ca. 400 μm long (vs. 500 μm); median lobe, although conical, relatively broader in upper half, more distinctly tapered apically and with a more roundly pointed tip (vs. almost sub-parallel in apical fourth); corona very distally localized (vs. in apical forth (Schönmann, 1995)). Median lobe apically extended by a membranous sac. Certainly, the specimen belongs to the *P. vitalisi* complex of species. We are planning to investigate the species delimitation with mtDNA analysis before attempting a formal description of the presumed new species, since Schönmann (1995) suggested more detailed studies to clarify if *P. vitalisi* s.l. comprises one or several species.

Habitat: The specimen was collected from submerged wood in fast flowing water. For many species of the genus, it is somewhat typical to dwell in runs and ripples of streams. This is quite exceptional among hydrophilids, which are usually adapted to slow-moving or stagnant waters.

**Chaetaarthrinae**

**Chaetaarthria sp.**

**Material:** One female (Cam5, e, 13.7.2018, M).

Habitat: The specimen was collected in the same site and microhabitat as *Notionotus notaticollis* (see respective remarks for that species).
Acidocerinae

**Agraphydrus (s.str.) coomani** (Orchymont, 1927)


**Material:** One male (Cam3P, 12.7.2018, L).

**Distribution:** _Agraphydrus coomani_ is widely distributed from Vietnam, peninsular Malaysia across Papua New Guinea up to Australia (Hansen, 1999). This is the first record from Cambodia.

**Habitat:** The single specimen was attracted to a light trap placed near a vegetated pond. _Agraphydrus_ species are usually found in littoral deposits of stagnant water bodies or calm stream sections (Freitag, 2015).

**Agraphydrus sp.**

**Material:** Sp. 1, one female (Cam5, e, 13.7.2018, M).

**Habitat:** Our specimen was collected from a shallow, isolated side pool with CPOM and leaf litter near a small river in secondary forest.

**Remarks:** Except for the previous record, no species of _Agraphydrus_ has been recorded from Cambodia (Hansen, 1999; Short & Hebauer, 2006; Short & Fikacek, 2011). We did not attempt to identify our single female specimen. The genus is currently undergoing a taxonomic revision by Albrecht Komarek (NMW).

**Helochares (Hydrobaticus) sp.** (Fig. 4A–B)

**Material:** Three males, two females (Cam3P, 12.7.2018, L).

**Habitat:** See respective remarks for _Agraphydrus coomani_.

**Remarks:** Only the following species of _Helochares ( Hydrobaticus)_ MacLeay, 1871 were previously known from Cambodia: _H. anchoralis_ Sharp, 1890; _H. lentus_ Sharp, 1890; _H. neglectus_ (Hope, 1845); and _H. salvazai_ Orchymont, 1919. Our specimens do not agree with the descriptions of any of these or other comparative material from Southeast Asia and is probably new to science. It is easily recognizable by its aedeagus: the long and slender parameres and internal fibula that can be outstretched presumably during copulation (Fig. 4B).

Two of our males have identical features, but their genitalia are retracted and conically aligned. Comparisons with species known from the sub-region suggests this is a new species but further study is needed to confirm this and allow for formal description.

**Helochares (s.str.) pallens** (MacLeay, 1825)

_Enhydрус pallens_ MacLeay, 1825, 140 (orig. descr.); _Helochares pallens_ (MacLeay, 1825), Orchymont, 1932, 688; _Helochares (s.str.) pallens_ MacLeay, 1825, Hansen 1999, 162 (cat.); Freitag, 2013, 18.

**Material:** One female (Cam3P, 12.7.2018, L).

**Distribution:** _Helochares pallens_ is widely distributed from New Guinea to the Afrotropical region and the southern Palaearctic (Hansen, 1999). Several records are known from Thailand (Hansen, 1999), but this is the first record from Cambodia.

**Habitat:** See respective remarks for _Agraphydrus coomani_.

**Enochrinae**

**Enochrus sp.**

**Material:** One male (Cam3P, 12.7.2018, L).

**Habitat:** See respective remarks for _Agraphydrus coomani_.

**Remarks:** _Enochrus (Methydrus) haroldi_ (Sharp, 1884) is the only species of the genus recorded from Cambodia (Satô & Chujô, 1961). The available literature and lack of immediately accessible types do not allow us to identify the species at present.

**Notionotus notaticollis** Hebauer, 2001


**Material:** Two males, two exemplars (Cam5, e, 13.7.2018, M); three males, two females (Cam3R, e, 11.7.2018, M).

**Habitat:** Most specimens were collected at the same site and microhabitat as _Agraphydrus_ sp.

**Distribution:** _Notionotus notaticollis_ is known from Laos, Vietnam and China (Hebauer, 2001). This is a new record for Cambodia.

**Sphaeridiinae**

**Coelostoma sp.** (Fig. 4C)

**Material:** One male (Cam4, f, 12.7.2018, M); one female (Cam3R, a, 11.7.2018, M).

**Habitat:** Our two specimens were found in wet semi-submerged wood in the sprayzone of a waterfall.
and in a shallow littoral in slow-flowing water, respectively. Shallow flowing or stagnant waters are generally a common habitat for *Coelostoma* Brullé, 1835 and specimens are often observed on hygropetric rocks (H. Freitag, unpublished data).

**Remarks:** The following species of *Coelostoma* are recorded from Cambodia: *C. (Hamnocelostoma) salvezai* Orchymont, 1919; *C. (Lachnocoelostoma) phallicum* Orchymont, 1940; and *C. (Lachnocoelostoma) vagum* Orchymont, 1940. Seven more species are known from neighbouring countries (Hansen, 1999). However, the specimens from our collection do not agree well with any of those, especially in the male aedeagus, and they are probably new to science. Further study needed to confirm this assessment.

**ELMIDAE (Riffl e Beetles), Elminae**

*Graphelmis boukali* Čiampor, 2004 (Fig. 4D)


**Material:** One female (Cam3R, f, 11.7.2018, M).

**Habitat:** As with most representatives of the genus, this taxon was found on submerged wood in rapidly flowing water.

**Distribution:** Our sole female agrees well with the original description and type material at NMW. *Graphelmis boukali* was previously only known from Laos, Malaysia and Thailand (Čiampor, 2004). This is a new record for Cambodia.

*Grouvellinus* sp.

**Material:** One larva (Cam3R, g, 11.7.2018, M).

**Habitat:** A single larva was collected from a rock surface in a forest river.

**Remarks:** The general habitus of *Grouvellinus* larvae is known from Kodada *et al.* (2016a), Hayashi & Sota (2010) and works of the first author in Borneo and the Philippines (H. Freitag, unpublished data). In Indochina, the species *Grouvellinus pelacoti* Delève, 1970, *G. setosus* Delève, 1970 and *G. tonkinus* (Grouvelle, 1889) are known from Vietnam, and *G. punctatostriatus* Bollow 1940 and *G. sculptus* Bollow 1940 from Myanmar, respectively (Jäch *et al.*, 2016). Due to the lack of larval descriptions, our specimen cannot be identified. This is the first record of the genus from Cambodia.

**Stenelmis** sp.

**Material:** One female (Cam5, c, 13.7.2018, M).

**Habitat:** Our specimen was collected from gravel deposits in a stream riffle section.

**Remarks:** Species of *Stenelmis* (s.l.) are usually very common in Southeast Asian streams (H. Freitag, unpublished data). However, many of them are undescribed and the genus needs revision, as it may comprise several unrelated genera.

**Larinae**

*Potamophilinus* sp. [*? longipes*]

**Material:** Three males, three females (Cam3R, h, 11–12.7.2018, M).

**Habitat:** Our specimens were collected from littoral grass bunches.

**Remarks:** *Potamophilinus longipes* Grouvelle, 1892 is only known from Myanmar. Due to the brief original description (Grouvelle, 1892) and lack of immediate access to type material, our specimens cannot be identified with certainty. There is an obvious sexual dimorphism on the sub-basal section of the elytra: females display distinct rounded humps between the 2nd and 3rd elytral striae, similar to the spiny humps in females of *P. bispinosa* (Bollow, 1938, 172–174: Fig. 31) for which material has been checked at NMW for comparison. The genus is recorded from Cambodia for the first time.

**DRYOPIDAE (Longtoed Water Beetles)**

*Ceradryops* sp.

**Material:** Two exemplars (Cam4, f, 12.7.2018, M).

**Habitat:** Our specimens were collected from semi-submerged wood in rapidly-flowing water below a waterfall. We presume they were not really submerged.

**Remarks:** This species is unknown to science. Several species of the genus are known from India and Sri Lanka. Within the region adjacent to Indochina, only one species, *Ceradryops matei* Kodada & Boukal 2003, is known from Hong Kong (China) which obviously differs in lacking a dense setaceous body cover underlying the erected setae (Kodada & Boukal, 2003: Fig. 12) and by possessing a slenderer pronotum and smaller overall size (ca. 1.8 mm). This interesting discovery will be described in a separate paper. Whether or not the genus represents a true dryopid is currently questionable (Kodada *et al.*, 2016b).
Elmomorphus sp.

**Material:** Eight exemplars (Cam3R, f, 11.7.2018, M).

**Habitat:** Our specimens were collected from fully submerged wood and leaf litter in rather slow-flowing, shallow water.

**Remarks:** The Elmomorphus genus is common and wide-spread and is currently being revised by David Ziak & Jan Kodada of the Comenius University Bratislava. We therefore do not attempt a detailed diagnosis.

PSEPHENIDAE (Waterpenny Beetles), Eubrianacinae

Jinbrianax sp.

**Material:** One larva (Cam4, g, 20.7.2018, M).

**Habitat:** Our specimen was collected from rock surface in torrential water below a waterfall.

**Remarks:** Due to the lack of larval descriptions at the species level, our specimen cannot be identified further. The species Jinbrianax metallicus (Pic, 1922) (West Malaysia, Thailand & Vietnam) and J. schillhammeri Lee, Satô & Yang, 1999 (Laos) are known from the sub-region (Lee et al., 1999). Our report is the first record of the genus from Cambodia.

Discussion

Although our study was based on just three days of fieldwork, we were able to record 21 taxa of aquatic Polyphaga (seven confidently identified species, eight treated as morphospecies and six presumably new species) from four study sites. This figure might have been much higher with better weather conditions, because heavy monsoon rain and flooding occurred before and during our sampling. At least 16 (76%) of our taxa (excluding unidentified specimens of genera already recorded in the country) have never been recorded from Cambodia. This illustrates the lack of historical survey activity and reflects the current incompleteness of knowledge regarding Cambodian water beetles and aquatic macroinvertebrates in general.

Jäch & Balke (2008) predicted that 39% of Oriental water beetles have yet to be described. While our knowledge does not yet allow for precise estimates (Appendix 1), many new species likely still await discovery and scientific description, some of them within our collections. This is exemplified by our first records of the Dryopidae, Elmidae, Hydraenidae and Psephenidae families from Cambodia, although we only provide genus and morpho-species records here, except for Graphestalis bokuali.

Notwithstanding this increase in faunistic data, knowledge on Cambodian water beetles remains far from complete. Further sampling and rigorous identifications are needed to provide a basis for ecological studies. Sampling is especially needed in areas of high-endemcity such as mountain ranges, as well as sampling in different seasons. A high variety of microhabitats and various environmental parameters should be included in the data collection (Freitag, 2015; Freitag et al., 2016: 207–208). Different trapping methods should also be employed, such as emergence traps and drift nets, which have proven useful in several studies (Freitag, 2015 and references therein).

Acknowledgements

We would like to especially thank the German Academic Exchange Service (DAAD) for funding the BIO-PHIL project (FZ: 57393541) that enabled this collaborative study and practical biodiversity training. Thanks are also due to the Chambok Community and especially to Mr Touch Morn, community leader of Chambok Ecotourism Site, for helping to facilitate the field sampling. We also gratefully thank Mr Chhin Sophea for creating the map of Kampong Speu Province and study sites. The authors from Museum für Naturkunde and the first author would like to thank the curator at the NMW, Dr Manfred A. Jäch, as well as Dr Jan Kodada of the Comenius University in Bratislava who gave helpful advice and provided uncomplicated access to the Coleoptera collections and technical facilities for the study of comparative material. Two anonymous reviewers are sincerely thanked for their helpful comments and suggestions.

References


Aquatic Polyphaga from Kampong Speu
Appendix 1 Checklist of aquatic Polyphaga (families Spercheidae, Hydrophilidae, Hydraenidae, Elmidae, Dryopidae, Psephenidae) known to occur in Cambodia

The list below compiles data from this study and relevant catalogues (Hansen 1998, 1999; Short & Hebauer 2006; Short & Fikacek, 2011; Jäch et al., 2016). Representatives of genera which are recorded for the first time are included, even if not identified to species level. Unidentified species of genera previously recorded in Cambodia are excluded, as are taxa which are considered terrestrial (e.g., Sphaeridium Fabricius, 1775 and Coelofletium Orchymont, 1925 (Hydrophilidae)).

\( N = \) new record from this study, \( P = \) previously published record.

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<tr>
<th>#</th>
<th>Taxon</th>
<th>Record</th>
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<td><strong>SPERCHEIDAE</strong></td>
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<td>1</td>
<td><em>Spercheus</em> stangii Schwarz &amp; Barber, 1918</td>
<td>P</td>
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<tr>
<td>2</td>
<td><em>Allocotoccerus</em> myronius (Orchymont, 1939)</td>
<td>P</td>
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<tr>
<td>3</td>
<td><em>Allocotoccerus</em> nigellus Chujô &amp; Satô, 1964</td>
<td>P</td>
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<tr>
<td>4</td>
<td><em>Amphiops</em> mater Sharp, 1873</td>
<td>P</td>
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<tr>
<td>5</td>
<td><em>Berosus siamensis</em> Schodel, 1992</td>
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<tr>
<td>6</td>
<td><em>Berosus</em> (Euoplurus) chinensis Knisch, 1922</td>
<td>N</td>
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<td>7</td>
<td><em>Berosus</em> (s.str.) <em>pulchellus</em> MacLeay, 1825</td>
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<td><em>Hydrobiomorpha</em> (s.str.) <em>cambodiensis</em> (Régimbart, 1903)</td>
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<td><em>Hydrobiomorpha</em> (s.str.) <em>malaisica</em> Mouchamps, 1959</td>
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<td><em>Hydrophilus</em> (s.str.) <em>bilineatus caschmirensis</em> Redtenbacher, 1844</td>
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<td><em>Hydrophilus</em> (s.str.) <em>cavisternum</em> (Bedel, 1891)</td>
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<td>12</td>
<td><em>Regimbartia attenuata</em> (Fabricius, 1801)</td>
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<td><em>Paracyrnus mimicus</em> Woolridge, 1977</td>
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<tr>
<td>14</td>
<td><em>Pelthydrus vitalisi</em> Orchymont, 1926 (s.l.)</td>
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<td>15</td>
<td><em>Sternolophus inconspicuus</em> (Nietner, 1856)</td>
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<td><em>Sternolophus rufipes</em> (Fabricius, 1792)</td>
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<td><em>Chaetarthria</em> sp.</td>
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<td><strong>HYDROPHILIDAE, Acidocerinae</strong></td>
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<td><em>Agraphydrus</em> coomani (Orchymont, 1927)</td>
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<tr>
<td>19</td>
<td><em>Chasmogenus abnormalis</em> (Sharp, 1890)</td>
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<td><em>Helochares</em> (Hydrobaticus) <em>anchoralis</em> Sharp, 1890</td>
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<td>21</td>
<td><em>Helochares</em> (Hydrobaticus) <em>lentus</em> Sharp, 1890</td>
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<td>22</td>
<td><em>Helochares</em> (Hydrobaticus) <em>neglectus</em> (Hope, 1845)</td>
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<td>23</td>
<td><em>Helochares</em> (Hydrobaticus) <em>salvazai</em> Orchymont, 1919</td>
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<td><em>Helochares</em> (s.str.) <em>pallens</em> (MacLeay, 1825)</td>
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<td><em>Helochares</em> (s.str.) <em>taprobanicus</em> Sharp, 1890</td>
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<td>26</td>
<td><em>Helochares</em> (s.str.) <em>vitalisi</em> Orchymont, 1919</td>
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<td><em>Notionotus notaticollis</em> Hebauer, 2001</td>
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<td><em>Coelostoma</em> (Hammacoelostoma) <em>salvazai</em> Orchymont, 1919</td>
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<td><em>Coelostoma</em> (Lachnocoelostoma) <em>phallicum</em> Orchymont, 1940</td>
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<td><em>Coelostoma</em> (Lachnocoelostoma) <em>vagum</em> Orchymont, 1940</td>
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<td>33</td>
<td><em>Graphelmis</em> boukali Ciampor, 2004</td>
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<td>34</td>
<td><em>Grouvellinus</em> sp.</td>
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<td><em>Stenelmis</em> sp.</td>
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<td><em>Potamophilinus</em> sp.</td>
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<td><strong>DRYOPIDAE</strong></td>
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<td>37</td>
<td><em>Ceradryops</em> sp.</td>
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<td>38</td>
<td><em>Elmomorphus</em> sp.</td>
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<td>39</td>
<td><em>Jinbrianax</em> sp.</td>
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Impact of experimental domestic water buffalo *Bubalus bubalis* grazing on waterhole dynamics in north-eastern Cambodia

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Abstract

The open deciduous dipterocarp forests of northern and eastern Cambodia are globally irreplaceable for biodiversity conservation. The substantial declines occurring in herbivore biomass within the eco-region due to hunting of wild ungulates and changing animal husbandry patterns are likely impacting the structure of these forests. These impacts may be particularly pertinent at seasonal waterholes where it is hypothesized that grazing by ungulates is important for maintaining micro-habitat diversity utilized by globally threatened large water birds such as the Critically Endangered giant ibis *Thaumatibis gigantea* and white-shouldered ibis *Pseudibis davisoni*. To test this, we experimentally grazed eight waterholes in the globally significant Siem Pang Wildlife Sanctuary in northeastern Cambodia with two herds.
of domestic water buffaloes over three dry seasons and compared their micro-habitats with 10 control waterholes. During the dry season, the physical characteristics of all waterholes changed predictably and consistently, irrespective of whether they were controls or grazed by our buffaloes. We found few consistent differences between control waterholes and those grazed by buffaloes, although there was some evidence that grazed waterholes retained water and saturated mud, the latter a critical habitat for ibis, for longer into the dry season. However, we could not arrive at definitive conclusions about the effect of water buffalo activity on waterhole structure in deciduous dipterocarp forests in Cambodia from our data and it is possible that the densities of buffaloes we employed were not sufficient to significantly or detectably influence this.

**Keywords**
Conservation management, dry forest, ecosystem services, re-wilding, wild cattle.

**Introduction**

Large herbivores play a critical role in ecosystem functioning and the significant declines in herbivore populations globally are likely to have had major impacts on many aspects of ecosystem structure and thus biodiversity (Ripple *et al.*, 2015). The deciduous dipterocarp forests (DDF) of insular Southeast Asia are a prime example of an ecosystem whose functioning is likely to have been significantly impacted by large herbivore declines (Miles *et al.*, 2006; Tordoff *et al.*, 2012). These savannah forests were historically described as one of the great game lands of the world and supported a unique assemblage of four species of wild cattle (banteng *Bos javanicus*, gaur *B. gaurus*, kouprey *B. sauveli*, wild water buffalo *Bubalus arnee*), combined with abundant deer and widespread Asian elephant *Elephas maximus*. As such, herbivory, trampling and wallowing by herbivores, notably Asian elephant and wild water buffalo, likely played a key role in structuring these forests and creating suitable habitat mosaics including waterholes for water birds.

The largest extent of DDF remaining globally occurs in Cambodia in a series of protected areas in the northern and eastern provinces of Mondulkiri, Stung Treng, Ratanakari and Preah Vihear. These are globally irreplaceable for conservation of DDF and its characteristic biodiversity (Gray *et al.*, 2012; Tordoff *et al.*, 2012). Despite declines in their mammalian megafauna, these forests still support important populations of globally threatened birds including the giant ibis *Thaumatibis gigantea* and white-shouldered ibis *Pseudibis davisoni* (both Critically Endangered), greater adjutant *Leptoptilos dubius* (Endangered), lesser adjutant *Leptoptilos javanica* and Asian woollyneck *Ciconia episcopus* (both Vulnerable) and three species of vultures (all Critically Endangered) (Gray *et al.*, 2014; Ty *et al.*, 2016). White-shouldered and giant ibises are largely DDF specialists, with the former a near-endemic and the latter now endemic to DDF in the Mekong basin, and both are dependent on seasonal waterholes within the forest for feeding (Keo, 2008). During the dry season, dried mud and short vegetation at waterholes have been identified as essential foraging habitat for the white-shouldered ibis (Wright *et al.*, 2010), whereas the giant ibis prefers saturated mud (Wright *et al.*, 2012). In eastern Cambodia, Pin *et al.* (2018) found that water depth and size of the open area of low vegetation surrounding waterholes were positively correlated with use by threatened water birds.

It has been suggested that wallowing and grazing by domestic water buffaloes *Bubalus bubalis* may play an important role in maintaining the ecological integrity of waterholes in the absence of wild ungulates such as wild water buffaloes and Asian elephants. Due to agricultural modernisation, buffalo ownership in many areas of DDF is decreasing. For example, in the Siem Pang and Siem Pang Khan Lech Wildlife Sanctuaries (hereafter collectively referred to as Western Siem Pang Wildlife Sanctuary), which support the largest known population of white-shouldered ibis globally, 80% of buffalo owners have expressed a desire to replace buffaloes with hand-drawn tractors in the near future due to problems with livestock disease (BirdLife International Cambodian Programme, unpublished data). Wright *et al.* (2013) hypothesised that the absence of domestic buffaloes from this site would increase sedimentation and vegetation at waterholes and reduce their suitability for species such as the white-shouldered ibis. As such, maintaining extensive rearing of domestic water buffaloes could be important for conservation of the ibis.

The aim of our study was to examine the impact of targeted stocking and grazing of domestic water buffaloes on waterholes in Western Siem Pang Wildlife Sanctuary, north-eastern Cambodia. We hypothesised that increased levels of grazing by domestic water buffaloes would increase the extent of low vegetation and dry mud preferred by white-shouldered ibises for foraging during their dry season breeding period and that such micro-habitats would be more extensive in experimental waterholes receiving managed grazing by domestic water buffaloes than at control waterholes.
Methods

Study area

Our study was conducted within Siem Pang Wildlife Sanctuary (66,932 ha) and Siem Pang Khang Lech Wildlife Sanctuary (65,389 ha) (Fig. 1). The latter was added to Cambodia’s protected area system in May 2016 (Souter et al., 2016) and the two sites are now treated as a single management unit. These are collectively referred to as Western Siem Pang Wildlife Sanctuary in this paper. Treated as such, Western Siem Pang Wildlife Sanctuary contains the largest known subpopulation of white-shouldered ibises globally, with a minimum of 346 birds (Wright et al., 2013), together with a breeding population of 50 giant ibises (Ty et al., 2016) and Cambodia’s largest remaining population of vultures which comprise breeding red-headed vultures *Sarcogyps calvus*, white-rumped vultures *Gyps bengalensis* and slender-billed vultures *G. tenuirostris*. Despite a significant (ca. 50–100 individuals) population of Eld’s deer *Rucervus eldii* (a wallowing species), the landscape supports few mammals larger than wild pigs *Sus scrofa* (also a wallowing species), with Asian elephants extirpated in Siem Pang Kang Lech Wildlife Sanctuary and very low numbers of banteng and gaur remaining.

The landscape of Western Siem Pang Wildlife Sanctuary comprises DDF, which mainly occurs west of the Sekong River, and semi-evergreen forest to the north and east of the Sekong River (Fig. 1). The Sekong River and its associated riverine forests bissect the protected area. Waterholes (*trapeang* in Khmer) ranging in size from 0.001–3.4 ha occur throughout the DDF and strongly seasonal rainfall creates spatiotemporal variation in water levels and substrate moisture (Wright et al., 2010). The rainy season occurs in May–October with maximal monthly rainfall (mean 333 mm) in September. The dry season occurs in November–April, with minimal monthly rainfall (mean 0.9 mm) in January (Thuon & Chambers, 2006).

Waterhole manipulation

We selected 18 seasonal waterholes in Western Seam Pang and allocated these to two types: i) control waterholes (*n* = 10); ii) experimental waterholes (*n* = 8) (Fig. 1). The waterholes were not randomly selected because clusters relatively short distances apart were preferable to allow us to move our buffalo herds safely with minimal stress to the animals. As such, the waterholes might not have been independent with respect to ecological processes affecting the study site, although we anticipated that factors such as rainfall would influence all waterholes to the same extent. Distances between waterholes varied from 20 m to 1,343 m. The mean dimensions of the waterholes were 46 m x 64.4 m, with an average area of 2,967 m². Differences in the dimensions of control and experimental waterholes were not significant (Appendix 1; One-Way ANOVA on normally distributed data: *F* = 0.33, *p* = 0.72).

BirdLife International purchased 24 domestic water buffaloes within Seam Pang District in 2014 on the assumption that locally procured stock would be adapted to DDF. This number grew to 28 because many of the females were pregnant when purchased and calved during the experiment. Our initial plan was to establish two herds of domestic water buffalo: i) a large herd comprising 16 individuals; and ii) a small herd comprising eight individuals. Four experimental waterholes were to be grazed by the large herd for seven days per waterhole/month, and four more were to be grazed by the small herd for seven days per waterhole/month on a continuous rotation. However, 14 of the buffaloes died during a drought caused by the 2016 El Niño event. As a consequence, four waterholes were grazed by between 8–16 buffaloes (depending on study year) for seven days each month (high intensity treatment) and four were grazed by eight buffaloes for seven days per...
month (low intensity treatment). The decision to deploy these grazing intensities was subjectively made. Prior to the experiment, the waterholes selected were judged to have sufficient vegetation for the chosen grazing intensity for the study duration. We anticipated our grazing intensities might mimic a natural grazing regime, but had no baseline to guide us due to the lack of literature on natural grazing levels in DDF.

When grazing at waterholes, buffalo herds were restricted to less than 20 m from the waterhole edge with electrical fencing (solar-powered, single-strand white tape) (Fig. 2). Grazing at waterholes only occurred during the dry season (November through February) and outside of this period, the buffalo were kept in electric-fenced areas nearby or at the closest village. The eight experimental waterholes were grazed by buffalo for three successive dry seasons: 2014–15, 2015–16 and 2016–17.

Waterhole microhabitats

The extent of different microhabitats was recorded at all 18 waterholes at approximately monthly intervals during each dry season. In the 2014–15 dry season, waterholes were visited between December and April four times (excluding January 2015). During the 2015–16 and 2016–17 dry seasons, each waterhole was visited monthly between November and April for a total of six visits per waterhole. Microhabitats recorded were based on distinctions in moisture level and were those identified as being important for ibis foraging by Wright et al. (2012). These comprised pools of water and saturated, wet or dry substrates (Fig. 3). Mud was classified in increasing order of dryness as saturated, wet, or dry based on criteria in Wright et al. (2012). During each visit the extent of each microhabitat at waterholes was sketched following Wright et al. (2012). We then digitized the sketch maps and calculated the area of each microhabitat in ArcGIS 9.3 (ESRI, 2015). Because the size of waterholes was variable, although consistent between treatments (Appendix 1), we converted these area measurements into a percentage of the total waterhole area for analysis. Vegetation height at each waterhole was calculated as the mean height of vegetation measured at four randomly selected points (Fig. 4).

A protocol was developed to monitor waterhole use by white-shouldered ibises, giant ibises and free-ranging domestic water buffaloes. Two observers visited each waterhole twice a month during the 2014–15, 2015–16 and 2016–17 dry seasons. These visits were separate to trips made to measure microhabitats at each waterhole. The date of the visits was random but one occurred in
the first two weeks of each month and the second in the second two weeks of each month. Each visit lasted up to 20 minutes and occurred between 0700–1000 hrs or 1400–1700 hrs.

Data analysis

We calculated mean (± SD) percentage cover of microhabitats and mean vegetation height at every waterhole for each survey month (i.e., November until April) during the study period. To estimate the effect of buffalo grazing on the microhabitat structure of waterholes, we compared the microhabitat data from the final study month (April 2017) across the 18 waterholes using a multivariate analysis of variance (MANOVA), with treatment type as the dependent variable and microhabitat as the response variable. This analysis was conducted in R (vers. 3.5.1; R Core Team, 2018) using the dplyr package. Prior to inclusion in the MANOVA, microhabitat variables were tested for normality using the Shapiro-Wilk normality test in the mvnormtest package. The same analysis was done on microhabitat data for waterholes at the beginning of the study (November/December 2014).

Results

A total of 334 survey visits were conducted to measure microhabitats at the 18 waterholes between November 2014 and April 2017. During this period, the physical characteristics of all waterholes changed predictably and consistently in the dry season, largely irrespective of treatment type (Fig. 5). More specifically, the area of water at each waterhole declined precipitously (Fig. 5A), the extent of saturated mud declined (Fig. 5B), the amount of wet mud increased before declining in January (Fig. 5C), the extent of dry mud increased (before plateauing at > 70% of waterhole area: Fig. 5D), whereas mean vegetation height declined (Fig. 5E).

While there were few clear or consistent differences in waterholes between the three treatments (i.e., control, high intensity grazing and low intensity grazing), grazed waterholes retained water and saturated mud for longer in the dry season compared to controls (Fig. 5A–B; Appendix 2). The Shapiro-Wilk normality test on micro-

Fig. 5 (right) Changes in the microhabitat composition of waterholes between the three treatments in Western Siem Pang from November to April: A) % of water, B) % of saturated mud, C) % of wet mud, D) % of dry mud, E) mean vegetation height. The figure shows mean monthly values for each microhabitat across the three study dry seasons.
habitat data for waterholes during the final study month (Appendix 3) indicated that three variables (water area, dry mud area and mean vegetation height) were normally distributed ($W > 0.72, p > 0.05$). These were included as response variables in the MANOVA which indicated no significant differences in microhabitat structure between the three treatments ($F = 1.55, p = 0.2; $ Appendix 4). A similar analysis on the same, normally distributed variables during the first project month (Appendix 3) also found no significant differences in microhabitat structure between treatments ($F = 1.45; p = 0.23$).

The giant ibis was not detected during the study period. However, white-shouldered ibises were detected on 56 survey visits. While the latter appeared to be detected more frequently at waterholes grazed by our buffalo herds (Table 1), it is difficult to interpret this finding as the waterholes were non-randomly allocated between treatments. Free-ranging domestic water buffaloes were detected on 34 survey visits with a mean herd size of 11 individuals (Appendix 5). Control waterholes appeared to be visited by domestic livestock less frequently than experimental waterholes (Appendix 5).

**Discussion**

Seasonal waterholes are critical for maintaining the conservation value of DDF in Indochina. This value includes providing foraging habitat for globally threatened water birds such as the giant ibis and white-shouldered ibis and water for large mammals including banteng and Eld’s deer (Gray et al., 2015). Our study demonstrates that the microhabitat characteristics of waterholes changes extensively during the dry season in DDF. This environmental change clearly influences the natural history of the landscape’s fauna. For instance, Wright et al. (2012) demonstrated that the dry season breeding period of the white-shouldered ibis is linked to the drying of waterholes, which increases access to resource-rich dry mud for foraging. In contrast, the giant ibis (which breeds in the wet season) preferentially forages in wet and saturated mud, which our results show are increasingly scarce habitats as the dry season progresses.

Despite the hypothesized influence of large ungulates on the microhabitat structure of waterholes, we failed to find significant physical differences between experimentally grazed waterholes and controls. Seasonality (e.g., month in dry season) was a much stronger predictor of waterhole dynamics than our experimental grazing. However, the experimentally grazed waterholes did appear to hold water and saturated mud, key habitats for wildlife in Cambodian DDF (Wright et al., 2012; Pin et al., 2018), for longer into the dry season than controls (Fig. 5). We analysed the percentage cover of each micro-habitat rather than their absolute area. While the total area of a substrate may be an important foraging cue for threatened water birds, given the lack of significant differences in the waterhole size between treatments, we regarded percentage cover as suitable for indicating microhabitat availability for large water birds and the impact of grazing on waterhole structure.

There may be several reasons why our experimental grazing did not cause more substantial changes to the microhabitat structure of waterholes. First, numbers of buffaloes in our experimental herds (8–16) were low and approximately half of what we had planned due to deaths that occurred during the study. Compared with historical densities of ungulates in Southeast Asian dry forests (Wharton, 1968; Eisenberg & Seidensticker, 1976), the density of individuals and biomass of grazers at our experimental waterholes were extremely low. Second, free-ranging domestic water buffalo were observed at control waterholes on 10 visits, with a mean group size of 13.6 buffalo (range 2–55), although free-ranging domestic buffaloes were detected more frequently at the experimental waterholes. For a more effective experiment, we should have maintained larger buffalo herds and fenced control waterholes to prevent grazing and wallowing by free-ranging domestic animals, although this would have been logistically difficult to implement. Restricting access to control waterholes for three dry seasons might also have affected local populations of Eld’s deer, other wild large mammals and domestic livestock. However, it ultimately seems likely that levels of grazing between our three treatments did not differ significantly and that our experimental waterholes did not experience sufficient grazing, trampling or wallowing to cause major changes in their microhabitat structure.

Given these issues, we cannot make definitive conclusions on the impact of water buffalo activity on waterhole function in Cambodian DDF. Excluding all grazing

<table>
<thead>
<tr>
<th>Treatments</th>
<th>No. of visits with WSI detections</th>
<th>% of visits with WSI detections</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control ($n=10$ waterholes)</td>
<td>23</td>
<td>7.9</td>
</tr>
<tr>
<td>High intensity grazing ($n=4$)</td>
<td>15</td>
<td>13.4</td>
</tr>
<tr>
<td>Low intensity grazing ($n=4$)</td>
<td>18</td>
<td>16.1</td>
</tr>
</tbody>
</table>

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animals from waterholes over multiple years may provide greater insights into the role that large ungulates play in structuring microhabitats used by threatened large water birds in the landscape. As noted above however, such an exclusion could impact wild ungulate populations and should therefore be carefully considered prior to implementation. Our results suggest that re-wilding efforts in DDF will require larger numbers of herbivores than we employed to achieve measurable and ecologically significant results. Given existing and ambitious plans for re-wilding Indochinese DDF, including the reintroduction of tigers Panthera tigris, which may require stocking of domestic water buffaloes to recover ungulate biomass and provide sufficient prey (Gray et al., 2018; WWF Cambodia, pers. comm. 2018), understanding the impact of herbivores on waterholes is important. As such, the extent to which water buffalo numbers affect waterhole dynamics and microhabitats used by Cambodia’s globally significant populations of large water birds should remain a key research subject.

Acknowledgements

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References


### Appendix 1  Size of waterholes between treatments in Western Siem Pang Wildlife Sanctuary

<table>
<thead>
<tr>
<th>Treatment</th>
<th>n</th>
<th>Mean ± SD (m²)</th>
<th>Range (m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10</td>
<td>4,111 ± 7,628</td>
<td>408 – 25,507</td>
</tr>
<tr>
<td>High intensity grazing</td>
<td>4</td>
<td>4,932 ± 4,377</td>
<td>1,837 – 8,027</td>
</tr>
<tr>
<td>Low intensity grazing</td>
<td>4</td>
<td>1,833 ± 1,482</td>
<td>563 – 4,558</td>
</tr>
</tbody>
</table>

### Appendix 2  Monthly composition of waterhole microhabitats across the three treatments in Western Siem Pang Wildlife Sanctuary

Values given are mean (± SD)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Month</th>
<th>Control</th>
<th>High intensity grazing</th>
<th>Low intensity grazing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water (%)</td>
<td>Nov</td>
<td>78.4 (21.4)</td>
<td>77.8 (15.6)</td>
<td>84.4 (17.8)</td>
</tr>
<tr>
<td></td>
<td>Dec</td>
<td>46.4 (32.5)</td>
<td>46.3 (27.9)</td>
<td>58.6 (33)</td>
</tr>
<tr>
<td></td>
<td>Jan</td>
<td>23.3 (24.4)</td>
<td>21.8 (18)</td>
<td>36.1 (30.1)</td>
</tr>
<tr>
<td></td>
<td>Feb</td>
<td>8.4 (12.2)</td>
<td>9.0 (10.7)</td>
<td>22.3 (25.3)</td>
</tr>
<tr>
<td></td>
<td>Mar</td>
<td>5.88 (9.98)</td>
<td>5.28 (9.5)</td>
<td>20.4 (22.4)</td>
</tr>
<tr>
<td></td>
<td>Apr</td>
<td>8.08 (13.7)</td>
<td>19.5 (22.6)</td>
<td>11.7 (14.4)</td>
</tr>
<tr>
<td>Saturated mud (%)</td>
<td>Nov</td>
<td>8 (11.8)</td>
<td>8.2 (7.1)</td>
<td>6.7 (5.8)</td>
</tr>
<tr>
<td></td>
<td>Dec</td>
<td>5.7 (4.6)</td>
<td>6.7 (4.9)</td>
<td>5.9 (5.2)</td>
</tr>
<tr>
<td></td>
<td>Jan</td>
<td>3.2 (4.6)</td>
<td>3.6 (3.6)</td>
<td>6.4 (6)</td>
</tr>
<tr>
<td></td>
<td>Feb</td>
<td>3.5 (7.6)</td>
<td>3.7 (5.8)</td>
<td>3.7 (4.0)</td>
</tr>
<tr>
<td></td>
<td>Mar</td>
<td>4.4 (8.4)</td>
<td>3.1 (5.9)</td>
<td>5.5 (4.9)</td>
</tr>
<tr>
<td></td>
<td>Apr</td>
<td>1.67 (2.98)</td>
<td>19.5 (22.6)</td>
<td>11.7 (14.4)</td>
</tr>
<tr>
<td>Wet mud (%)</td>
<td>Nov</td>
<td>2.85 (3.85)</td>
<td>4.48 (7.95)</td>
<td>4.6 (7.25)</td>
</tr>
<tr>
<td></td>
<td>Dec</td>
<td>6.5 (5.2)</td>
<td>7.4 (7.5)</td>
<td>5.1 (4.9)</td>
</tr>
<tr>
<td></td>
<td>Jan</td>
<td>3.21 (3.31)</td>
<td>3.5 (4.2)</td>
<td>5.1 (3.9)</td>
</tr>
<tr>
<td></td>
<td>Feb</td>
<td>4.01 (5.89)</td>
<td>4.43 (4.9)</td>
<td>3.6 (4.5)</td>
</tr>
<tr>
<td></td>
<td>Mar</td>
<td>2.89 (4.2)</td>
<td>3.06 (5.1)</td>
<td>3.49 (2.8)</td>
</tr>
<tr>
<td></td>
<td>Apr</td>
<td>3.42 (4.92)</td>
<td>2.9 (5.8)</td>
<td>4.67 (4.4)</td>
</tr>
<tr>
<td>Dry mud (%)</td>
<td>Nov</td>
<td>10.7 (17)</td>
<td>9.6 (8.3)</td>
<td>4.3 (10.7)</td>
</tr>
<tr>
<td></td>
<td>Dec</td>
<td>41.4 (36.2)</td>
<td>39.6 (27.2)</td>
<td>30.3 (34.2)</td>
</tr>
<tr>
<td></td>
<td>Jan</td>
<td>67.7 (33.5)</td>
<td>71.1 (23.6)</td>
<td>52.4 (35.3)</td>
</tr>
<tr>
<td></td>
<td>Feb</td>
<td>84.1 (18.3)</td>
<td>82.9 (15.4)</td>
<td>70.4 (28.4)</td>
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<tr>
<td></td>
<td>Mar</td>
<td>86.8 (15.3)</td>
<td>88.6 (13.2)</td>
<td>70.7 (26.7)</td>
</tr>
<tr>
<td></td>
<td>Apr</td>
<td>86.8 (20.5)</td>
<td>72.7 (31.5)</td>
<td>80 (17.9)</td>
</tr>
<tr>
<td>Vegetation height (cm)</td>
<td>Nov</td>
<td>52.3 (38.7)</td>
<td>56.8 (42.6)</td>
<td>72.5 (42.7)</td>
</tr>
<tr>
<td></td>
<td>Dec</td>
<td>60.8 (78.8)</td>
<td>53.9 (62.6)</td>
<td>61.2 (48.2)</td>
</tr>
<tr>
<td></td>
<td>Jan</td>
<td>50.4 (70.3)</td>
<td>37 (53.4)</td>
<td>40.8 (43.5)</td>
</tr>
<tr>
<td></td>
<td>Feb</td>
<td>34.4 (47)</td>
<td>54.9 (72.7)</td>
<td>38.5 (36.2)</td>
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<tr>
<td></td>
<td>Mar</td>
<td>28.2 (29)</td>
<td>14.7 (14.9)</td>
<td>33.5 (35.1)</td>
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<tr>
<td></td>
<td>Apr</td>
<td>23.5 (24.3)</td>
<td>22.6 (21.9)</td>
<td>29.5 (27.3)</td>
</tr>
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</table>
Appendix 3  Composition of waterhole microhabitats (mean ± SD) across the three treatments at the start and end of the study in Western Siem Pang Wildlife Sanctuary

Values given are mean (± SD)

<table>
<thead>
<tr>
<th>Month</th>
<th>Treatment</th>
<th>Water (%)</th>
<th>Saturated Mud (%)</th>
<th>Wet Mud (%)</th>
<th>Dry Mud (%)</th>
<th>Vegetation Height (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>December 2014</td>
<td>Control</td>
<td>66 (17)</td>
<td>8 (5)</td>
<td>5 (4)</td>
<td>22 (14)</td>
<td>34 (27)</td>
</tr>
<tr>
<td></td>
<td>High intensity grazing</td>
<td>70 (10)</td>
<td>16 (6)</td>
<td>3 (0.2)</td>
<td>11 (4)</td>
<td>52 (54)</td>
</tr>
<tr>
<td></td>
<td>Low intensity grazing</td>
<td>73 (41)</td>
<td>9 (4)</td>
<td>6 (5)</td>
<td>13 (16)</td>
<td>50 (41)</td>
</tr>
<tr>
<td>April 2017</td>
<td>Control</td>
<td>16 (16)</td>
<td>3 (4)</td>
<td>7 (5)</td>
<td>73 (22)</td>
<td>21 (17)</td>
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<tr>
<td></td>
<td>High intensity grazing</td>
<td>39 (5)</td>
<td>10 (2)</td>
<td>6 (8)</td>
<td>45 (2)</td>
<td>28 (27)</td>
</tr>
<tr>
<td></td>
<td>Low intensity grazing</td>
<td>15 (18)</td>
<td>5 (2)</td>
<td>8 (3)</td>
<td>72 (20)</td>
<td>39 (22)</td>
</tr>
</tbody>
</table>

Appendix 4  Multivariate analysis of variance (MANOVA) analysis comparing final and initial microhabitat characteristics between grazed waterholes and controls in Western Siem Pang Wildlife Sanctuary

<table>
<thead>
<tr>
<th>Variable</th>
<th>April 2017</th>
<th>December 2014</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$F$</td>
<td>$p$</td>
</tr>
<tr>
<td>Area of water (%)</td>
<td>1.58</td>
<td>0.24</td>
</tr>
<tr>
<td>Area of dry mud (%)</td>
<td>1.87</td>
<td>0.19</td>
</tr>
<tr>
<td>Mean vegetation</td>
<td>1.44</td>
<td>0.27</td>
</tr>
<tr>
<td>All three variables</td>
<td>1.55</td>
<td>0.20</td>
</tr>
</tbody>
</table>

Appendix 5  Number of visits to waterholes in Western Siem Pang Wildlife Sanctuary with detections of free-ranging domestic water buffalo

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Number (%) of visits with buffaloes present</th>
<th>Mean (range) number of buffaloes per visit</th>
<th>Mean herd size when buffaloes present</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10 (7)</td>
<td>0.9 (0–55)</td>
<td>13.6</td>
</tr>
<tr>
<td>High intensity grazing</td>
<td>11 (38)</td>
<td>4.5 (0–40)</td>
<td>11.9</td>
</tr>
<tr>
<td>Low intensity grazing</td>
<td>13 (16)</td>
<td>1.2 (0–17)</td>
<td>7.5</td>
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Recent literature from Cambodia

This section summarizes recent scientific publications concerning Cambodian biodiversity and natural resources. The complete abstracts of most articles are freely available online (and can be found using Google Scholar or other internet search engines), but not necessarily the whole article. Lead authors may be willing to provide free reprints or electronic copies on request and their email addresses, where known, are included in the summaries below.

Recent literature

Documents that use the Digital Object Identifier (DOI) System can be opened via the website http://dx.doi.org (enter the full DOI code in the text box provided, and then click Go to find the document).

If you or your organisation have recently published a technical paper, report or conference abstract that you wish to be included in the next issue, please send an electronic copy, summary or web-link to: Editor.CJNH@gmail.com or Editor.CJNH@rupp.edu.kh

New species & taxonomic reviews


This study presents faunistic records for 29 flesh fly species, including 12 species that are new records for Cambodia. Based on critical review of published records and study data, an updated checklist including 16 species of flesh fly is provided for Cambodia, together with lists for India, Taiwan, Thailand and Vietnam. Author: bartak@af.czu.cz


The authors describe a new species to science (Hergovitsia longivirga n. sp.) from Cambodia which also represents the first country record for the genus. They compare their new species with H. magnifica Buscek, 2012 and provide a key to the Hergovitsia genus on the basis of external morphology and male genitalia.


This study describes three new species in the Diduga genus from Cambodia (D. dubatolovi n. sp., D. kohkongensis n. sp. and D. bispinosa n. sp.) along with five recorded species: D. albicosta Hampson, 1891, D. barlowi Holloway, 2001, D. amoena Buscek, 2012, D. annulata Hampson, 1900 and D. alternata Buscek, 2014. A key to Cambodian species in the genus is provided, including illustrations of adults and genitalia.


The authors describe a new species of Medaurioidea Zompro, 2000 (M. romantica sp. nov.) from the Phnom Tnout mountain, Preah Vihear Province. The new species is the first record for this genus in Cambodia. They also propose taxonomic revisions for several species and bring the total number of stick insect taxa formally recorded from Cambodia to three genera and three species. Author: joachimbresseel@gmail.com


Lestes nigriceps Fraser, 1924 was described from India in 1922 and has not been reported since. In June 2018, a population was found in eastern Cambodia, Mondulkiri Province. This paper describes the Cambodian males, their variation and for the first time, the true female of L. nigriceps. Author: kosterin@bionet.nsc.ru


This study describes a new subspecies of dragonfly (Macromidia genialis buusraaensis) from 10 males and two females collected at three localities in Mondulkiri Province and provides a brief overview of the Macromidia genus. Author: kosterin@bionet.nsc.ru


The authors describe a new species of damselfly (Amphicnemis valentini sp. nov.) from Ream Peninsula in Cambodia and Phú Quốc Island, Vietnam. The new species is similar to A. gracilis Krüger, 1898 which occurs in Peninsular Malaysia and Sumatra, but differs in possessing a long process on the male prothorax. Author: kosterin@bionet.nsc.ru


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Cambodian Journal of Natural History 2018 (2) 110–113
The authors report Tetrablemmidae O. Pickard-Cambridge, 1873 from Cambodia for the first time and describe two new species to science from Kep Province and Battambang Province: Tetrablemma kepense sp. n. and Tetrablemma sokense sp. n. (respectively). Author: lisq@ioz.ac.cn


The authors present a comparative phylogeographic study which used molecular, morphological and morpho-metric methods to address systematic issues in the Kerivoula hardwickii complex in Asia. They also describe one new bat species to science (K. donghuangana sp. n) based on specimens from Vietnam and Cambodia. The new species is currently known only from the Annamite Mountains of Vietnam, Laos and Cambodia. Author: vttu@iebr.ac.vn

Biodiversity inventories


This paper presents data on Odonata collected in 2014–2018 from the coastal foothills of the Cardamom Mountains in Koh Kong Province. These data include the first record of Tetrathemis flavescens Kirby, 1889 nationally and the first record of Risiophlebia guentheri Kosterin, 2015 from southwestern Cambodia. Author: kosterin@bionet.nsc.ru


This note provides details of 12 camera trap photographs obtained in late 2014 and early 2015 of animals identified as red-shanked douc Pygathrix nemaeus in Virachey National Park. Author: greg.mccann1@gmail.com


The author provides an annotated list of 11 species of Tettigidae for Cambodia, including seven species recorded from the country for the first time. Author: storozhenko@biosoil.ru


The Vulnerable fishing cat faces a perilous future in Southeast Asia and was last sighted in Cambodia in 2003. The authors deployed camera traps at four sites in southern Cambodia in January–May 2015 to determine if it was still present. Eight records of the fishing cat were obtained from mangroves in Peam Krasop Wildlife Sanctuary and one from Ream National Park, but none were obtained from Botum Sakor National Park or Prey Nup. Several other globally threatened species were also photographed in Peam Krasop Wildlife Sanctuary. Author: nicholas.souter@alumni.adelaide.edu.au


The authors report 16 species of the fungi for the first time from Cambodia. One of these, Tetrigomycetes indeterminatus Thaxt., represents the first record since its description. Author: alter.rossi@univaq.it


The authors present a checklist of Eulophidae in Cambodia, which includes 26 species in 15 genera and nine additional species identified to genus. These include the first records for Southeast Asia of 18 species. Author: zyefremova@post.tau.ac.il

Species ecology & status


Cave-roosting bats in Southeast Asia are vulnerable to increasing pressure from climate change, habitat loss and human disturbance in the region. To guide their conservation management, the authors evaluated the reproductive phenology and body condition of Chaerephon plicatus for 14 months in 2015–2016 and quantified guano harvesting at the largest colony in Cambodia in 2011–2016. Two annual breeding cycles were documented and body condition declined significantly between the late wet season and late dry season, suggesting that the bats experience increasing energetic stress as the latter progresses. Annual guano harvests increased over the study period but could not be used as a proxy for monitoring population size. Author: neil.m.furey@gmail.com

Cambodia supports populations of three Critically Endangered vulture species. These are believed to be isolated from other populations globally and remained stable until recently. This study presents a population analysis which shows that populations of two species (white-rumped vulture *Gyps bengalensis* and red-headed vulture *Sarcogyps calvus*) have declined since 2010, while another (slender-billed vulture *Gyps tenuirostris*) may have begun to decline in 2013. These trends are supported by evidence of reduced nesting success. The authors suggest there is an urgent need to tackle use of carbamate pesticides in hunting and propose conservation actions.

Author: robin.loveridge@gmail.com


Deciduous dipterocarp forests provide crucial habitat for globally threatened species in Southeast Asia and water availability in these forests is limited to perennial rivers and waterholes during the dry season. This study investigated waterhole use by six globally threatened species through camera-trapping at 54 waterholes in Srepok Wildlife Sanctuary during the 2015–2016 dry season and measuring waterhole and landscape characteristics. Water depth and the area of water at the start of the dry season were the main factors influencing waterhole use, although waterholes further from villages were also used more frequently. The authors suggest that enlarging and deepening waterholes could enhance habitat for a range of species, although patrols would also be needed to ensure these are not targets for hunting.

Author: chanrata.pin@gmail.com

Coasts, wetlands and aquatic resources


Payment for ecosystem services may enable local participation in conservation if these are acceptable to communities. This study used contingent valuation methods to determine an appropriate level of incentives for local communities to take part in an ongoing nest protection scheme for six water bird species in the Sekong, Sesan and Srepok river basin of Cambodia.


The usefulness of the ecosystem services framework to emphasize relationships between agriculture and ecosystems has received little attention. This study combines an ecosystem services and dis-services approach with agrarian system analysis and diagnosis methodology to identify ecosystem services and and dis-services provided by rice production systems on the Tonle Sap Lake floodplain. Its findings suggest show that organic rice systems do not perform well economically or ecologically in ecosystem service provisions, whereas wet-season rice and floating rice in particular provide the most ecosystem services.

Author: nmalyne@rua.edu.kh


Molluscs are important for ecological function, livelihoods and fisheries, but are often forgotten in research and management. The authors report intra-annual variation in the landing and growth patterns of three mollusc species (*Corbicula moreletiana*, *Pila virescens* and *P. amplilacca*) in Kampong Chhnang Province of Tonle Sap Lake. Their results suggest that molluscs in the lake i) are important resources for people’s livelihoods, ii) respond differently to intra-annual variation in temperature, precipitation and hydrology, and iii) are being intensively exploited with significant reductions in growth rate.

Author: pengbun.ngor@gmail.com


The Sekong, Sesan and Srepok (3S) rivers support many threatened species and provide diverse natural resources and ecosystem services that support millions of people. The authors conducted a knowledge, attitudes and practices survey of selected communities along the 3S rivers to understand perceptions about an ongoing nest protection scheme for waterbirds there. According to local communities, waterbird populations increased and threats decreased as a result of the programme. They also had positive impressions of the programme and believed it provided them with significant livelihood benefits.

Author: seak.sophat@rupp.edu.kh

Livelhoods of people in the Tonle Sap Lake area of Cambodia are complex combinations of rice-based cropping, fishery systems, mixed cash crops/home gardens, natural pond culture/aquaculture, cattle and livestock, collection of flooded forest products, and non-farm and off-farm activities. This paper provides an overview of these through economic and mapping analysis of two districts of Battambang Province. It also identifies types of household activities and their economic performance, changes in farming systems, agro-ecological zones, levels of poverty and resilience, and the country’s related gain and loss in gross domestic production. Author: sokkim-chhin@gmail.com


Marine and coastal areas in Cambodia cover 17,237 km² and provide important resources for human well-being. Rapid expansion of fisheries in these areas has caused economic and environmental concerns about their management. This study used secondary data to explore trends in marine and coastal activities and associations with socioeconomic variables over the last two decades. It concludes that marine and coastal areas face stresses from increases in a variety of human activity, rapid expansion of fisheries to meet demands for food consumption and declining mangrove forests. This is reflected in a decreased catch per unit effort which is associated with the rise in fishing activity. Author: pao.srean@gmail.com

### Forests and forest resources


The use of smartphones for data collection has opened up new opportunities for communities wishing to engage in monitoring. While information and communication technology (ICT) can systematize data collection, it can also present challenges for community members. The authors investigated the effectiveness of having local forest monitors use ICT to collect data on forest crimes in Prey Lang, Cambodia. Their findings suggest that communities with little formal education are able to monitor forest crimes and forest resources cost-effectively using ICT, although maintenance of software and hardware and data validation will continue to require external support. Author: idat@ifro.ku.dk


This study used a variety of social research methods to examine whether extraction of non-timber forest products (NTFPs) would encourage pro-conservation behaviour among people in Phnom Prich Wildlife Sanctuary. Its findings suggest that extraction of NTFPs does encourage local participation in forest conservation and that the annual value of NTFPs extracted in the wildlife sanctuary is approximately US$ 0.95 /ha or US$ 95 /km². Author: chou.phanith@d.mbox.nagoya-u.ac.jp


Dead organic matter (DOM) plays an important role in forest ecosystem functions, although little data exists for the seasonal forests of Indochina. The authors monitored deadwood and litter masses at 1–2 years intervals over 10 years in 22 permanent plots in evergreen and deciduous forest across Cambodia. They found that large loss events of aboveground biomass, which were probably caused by logging, increased deadwood mass and DOM carbon stock, but did not necessarily affect litter masses. As a whole, the forests were characterized by a relatively small deadwood mass, possibly due to anthropogenic removal of deadwood and dying trees. Author: kiono@ffpri.affrc.go.jp


Community forests play an important role in preserving forests in Cambodia, which has experienced rapid deforestation in recent decades. This study used remote sensing data to compare temporal variation in forest structure between six community forests in Phnom Kulen National Park and a wider study area. Their findings suggest that while community-protected forests can improve conservation outcomes to some extent, more actions are needed to curb illegal selective logging of valuable timber trees. Author: minerva_singh@yahoo.co.in

The Recent Literature section was compiled by Neil Furey, with contributions from Tom Gray, Oleg Kosterin and Valter Rossi.
Instructions for Authors

Purpose and Scope

The Cambodian Journal of Natural History (ISSN 2226–969X) is an open access, peer-review journal published biannually by the Centre for Biodiversity Conservation at the Royal University of Phnom Penh. The Centre for Biodiversity Conservation is a non-profit making unit, dedicated to training Cambodian biologists and the study and conservation of Cambodia’s biodiversity.

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* Cambodian or foreign scientists on any aspect of Cambodian natural history, including fauna, flora, habitats, management policy and use of natural resources.
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The Journal especially welcomes material that enhances understanding of conservation needs and has the potential to improve conservation management in Cambodia.

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The following types of manuscripts are accepted:

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* Short communications (300–2,000 words, excluding references)
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* Letters to the editor (<650 words)

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Full Papers (2,000–7,000 words, excluding references) and Short Communications (300–2,000 words, excluding references) are welcomed on topics relevant to the Journal’s focus, including:

* Research on the status, ecology or behaviour of wild species.
* Research on the status or ecology of habitats.
* Checklists of species, whether nationally or for a specific area.
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* Conservation management plans for species, habitats or areas.
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Concise reports (<300 words) on news of general interest to the study and management of Cambodia’s biodiversity.

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* Summaries of important news from an authoritative published source; for example, a new research technique, or a recent development in conservation.

Letters to the Editors

Informative contributions (<650 words), usually in response to material published in the Journal.

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Copies or links to recent (<18 months) scientific publications concerning Cambodian biodiversity and the management of natural resources. These may include journal papers, project technical reports, conference posters and student theses.
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The order of sections in the manuscript should be: cover page, main text, references, short biography of each author, tables and figures (including photographs). All pages should be numbered consecutively.

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Title: A succinct description of the work, in no more than 20 words.

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Cambodian authors are strongly encouraged to submit a Khmer translation of the English abstract.

Keywords: (Full papers only). Up to eight pertinent words, in alphabetical order.

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Main text: (Full papers). This should comprise the following sections in order: Introduction, Methods, Results, Discussion and Acknowledgements. Subsections may be included in the Methods, Results and Discussion sections if necessary. Conclusions and recommendations should be included in the Discussion.

References: These should be cited in the text in the form of Stuart & Emmett (2006) or (Lay, 2000). For three or more authors, use the first author’s surname followed by et al.; for example, Rab et al. (2006) or (Khou et al., 2005).

Multiple references should be in chronological order, for example, Holloway & Browne (2004); Kry & Chea (2004); Phan (2005); Farrow (2006).

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The present issue was also supported by a major foundation that chooses to remain anonymous.

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The Editors are grateful to our reviewers and to Chhin Sophea, Hun Seiha, Thi Sothearen, Srey Saovina, Regine Weckauf and Yim Raksmeay for their kind assistance with the production of this issue.
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