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研究課題名(和文) Testing dilution versus amplification effects of primate biodiversity on parasite biodiversity

研究課題名(英文) Testing dilution versus amplification effects of primate biodiversity on parasite biodiversity

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研究成果の概要(和文)：この研究の目的は、マレーシア・ボルネオの霊長類と寄生虫の生物多様性の関係を調査することでしたが、SARS-CoV2パンデミックのため、2020年度と2021年度には計画通りに進みませんでした。2023年にフィールド調査を実施し、解析を継続中です。また、国際渡航の遅れにより、日本の屋久島で侵入種が寄生虫の多様性に与える影響も調査しています。本報告書には、両プロジェクトでの進捗状況が含まれています。ボルネオでは霊長類と寄生虫の相互作用のモデル化を進め、屋久島では侵入種であるタヌキを通じた寄生虫拡散リスクを測定しています。

研究成果の学術的意義や社会的意義

Global events like the SARS-CoV2 pandemic underscore the importance of understanding disease emergence, especially in human-wildlife interface areas. Our work shows the connections between changing biodiversity and parasitic disease risk in wildlife within sensitive ecosystems.

研究成果の概要(英文)：The original aim of this research was to test the relationship between primate biodiversity and parasite biodiversity in the biodiverse forests of Malaysian Borneo. However, because of the SARS-CoV2 pandemic, we were unable to conduct this research as planned in the first 2 years (FY2020, FY2021). We were able to conduct field surveys in 2023, and analyses are ongoing. Because of the delays associated with international travel, we expanded this project to include an investigation of how invasive species - themselves agents of biodiversity change - influence parasite biodiversity in a terrestrial island ecosystem in Japan (Yakushima). This report includes progress made in both projects.

Through the project, we were able to further distinguish parasite diversity in a primate rich region of Malaysian Borneo and are currently working on modeling primate and parasite biodiversity interactions. We are also measuring risks of parasite spread to local fauna through invasive tanuki on Yakushima.

研究分野：Primateology and Wildlife Science

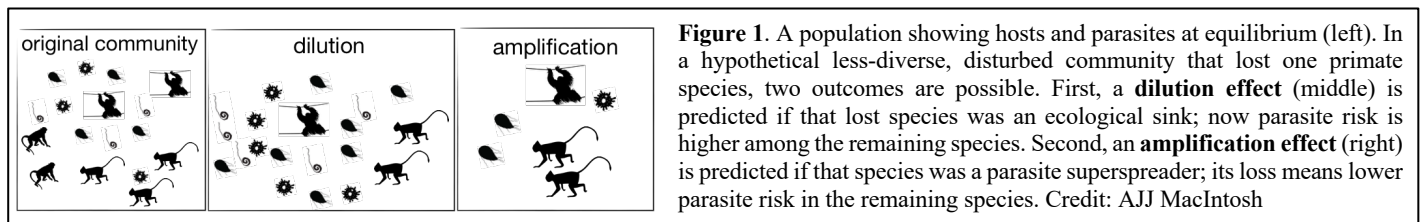
キーワード：primatology parasitology conservation epidemiology biodiversity macroecology

1. 研究開始当初の背景

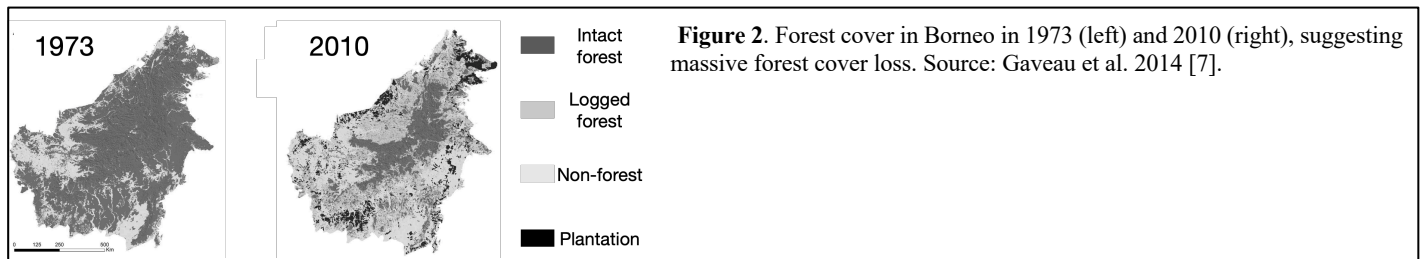
The Biodiversity Crisis: Over 60% of all animal populations are in decline (World Wildlife Fund’s Living Planet Index published in 2022). Over 60% of primates are currently listed as ‘*threatened with extinction*’ according to the IUCN Red List of Threatened Species, with 75% of populations in decline [1]. The main threats to primates are habitat loss and overharvesting, but infectious disease has emerged as another key factor that can drive vulnerable populations to extinction [2].

Parasites in ecosystems: Parasites organisms are a heterogeneous group, including viruses (e.g., EBOLA), bacteria (e.g., Anthrax), single-celled protozoa (e.g. malaria), multi-cellular intestinal worms (e.g., hookworm), and more. All can influence host populations, sometimes in unexpected ways. Parasites are key players in biological communities and some view them as supporting ecosystem health [3]. Monitoring the patterns and dynamics of parasitism in threatened species and ecosystems is key to understanding such threats to primates globally.

Parasites and host biodiversity: One concern is how changing host communities might impact parasite infection risk in endangered species. Ecologically speaking, there are two opposing predictions for the expected relationship between biodiversity loss and infectious disease risk (**Fig 1**). First, biodiverse communities can reduce exposure to parasites through a ‘*dilution effect*’, because species-rich communities have more hosts that act as ecological sinks for parasites, reducing their prevalence in the community [4]. The dilution effect has been cited as a key reason to conserve biodiversity, because it can reduce the disease burden on wildlife as well as on humans by reducing the chance of zoonotic transmission from animals. The second prediction arises because parasites themselves are part of the same evolved community, so if hosts are disappearing, their parasites may follow or even precede their host’s extinction [5]. Declines in free-living species diversity reduces potential amplifier hosts and therefore the exposure of remaining members of the community to parasites. This is known as the ‘*amplification effect*’, where some hosts are more important than others in transmitting certain parasites, so if they are lost, parasite transmission is reduced.



Model Systems: Many factors cause changes in biodiversity and influence host and parasite dynamics. The most common is habitat loss. The island of Borneo in Southeast Asia provides a good example of a biodiverse area with large-scale destruction of natural habitats (mostly for oil palm production; **Fig 2**). Bornean ecosystems support as many as 10 primate species [6], and most are listed as *at risk* in the IUCN Red List of Threatened Species. It is unclear how parasites and risk of parasitic disease has changed or may change in response to such conditions.



Other threats like the presence of invasive species also cause concern. Introduced species carry novel parasites and pathogens, which can emerge in local wildlife and human populations as ‘*pathogen pollution*’ [8]. An excellent example is the raccoon dog (*Nyctereutes* spp.), or tanuki in Japan, one of the most successful invasive mammals in Europe. In addition to eating and competing with local species they are also carriers of numerous transmissible diseases that they spread into new geographic regions [9]. Raccoon dogs are also invasive on two Japanese islands, but their impacts locally have yet to be assessed.

2. 研究の目的

In part one of this study, we aimed to test two competing hypotheses about biodiversity change and parasite infection risk:

- (1) decreasing biodiversity is associated with increased parasitism (**dilution effect**).
- (2) decreasing biodiversity is associated with decreased parasitism (**amplification effect**).

We worked in Sabah, Malaysia, where habitat fragmentation driven by agricultural practices and human settlements is causing changes in primate communities. We focused on gastrointestinal parasites, especially helminths. We first characterized biodiversity in primates and parasites, then assessed the relationship between the two. We built parasite sharing networks to visualize the relationships and used results to understand parasite infection risk under changes in primate biodiversity.

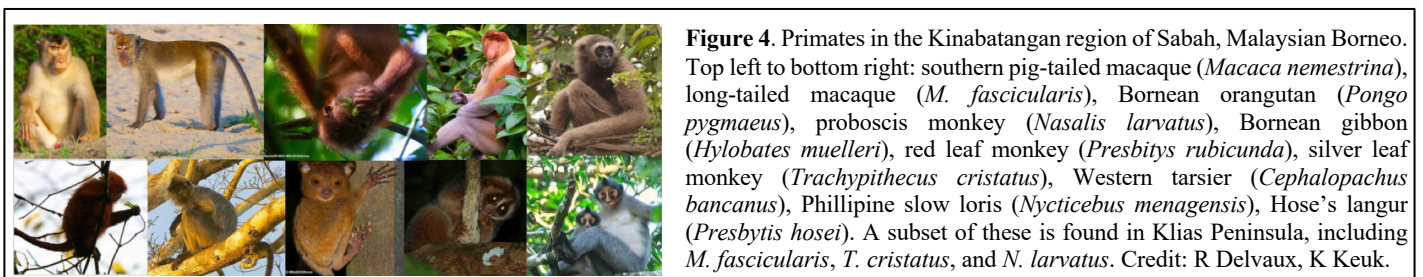
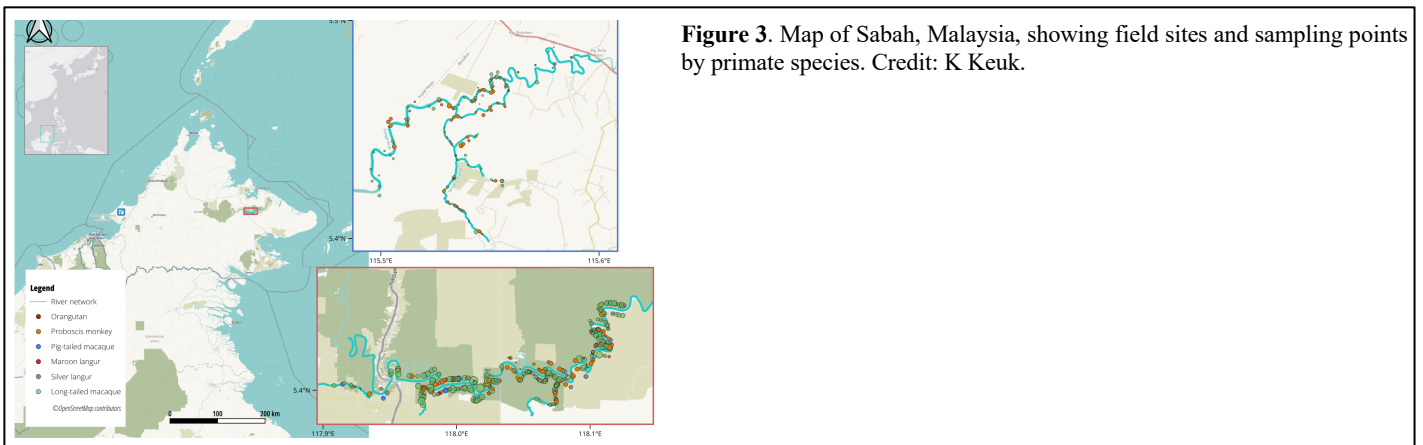
Part two of this study was done in Yakushima, Japan, where an invasive mammalian carnivore, the raccoon dog (tanuki, *Nyctereutes procyonoides*) was introduced in the 1980's and has become widespread, leading to unknown influences on local species. We are investigating their possible paths to ecosystem disruption. Here, we focused on how likely they are to spread disease, and what role their latrines (common defecation and urination sites) might play in this process.

3. 研究の方法

Study 1 – Primate Biodiversity and Parasitism.

Much of this research is part of an ongoing collaboration under the project name “*P²ARASITE*” (*Primates and PARAsites of Sabah as Indicators of a Transitioning Ecosystem*). Collaborators are researchers and/officers from the Danau Girang Field Centre and Cardiff University, the Sabah Wildlife Department, the Universiti Malaysia Sabah, Osaka City University, Oita University, the Czech Academy of Sciences and the University of Brno, and Kyoto University’s Wildlife Research Center and former Primate Research Institute. Fieldwork and laboratory analysis was largely carried out by K Keuk, with support from a team of research assistants.

Study Site & Subjects: We chose two study sites within Sabah, the Lower Kinabatangan Wildlife Sanctuary and Klias Peninsula (**Fig 3**), which have large differences in primate biodiversity (10 versus 3 species, respectively). **Fig 4** shows the primates found in Sabah.



Primate Censuses and Biological Sampling: We used boat surveys [10] to identify primate groups and measure primate diversity along the river. We then collected primate feces from the ground below sleeping trees where it was concentrated. We used molecular techniques to confirm the identity of the donor species before parasitological analysis.

Parasite Identification and Monitoring: We used microscopic and molecular techniques to analyze our parasitological samples, either laboratories at Kyoto University’s Inuyama Campus or at laboratories based in Sabah. Molecular analyses allowed us to assess host-parasite associations and phylogenetic diversity in parasite lineages, based on previous work we conducted in Sabah [11,12].

Study 2 – Invasive Tanuki and Parasitism.

Study Site & Subjects: The second part of the project focused on invasive tanuki (**Fig 5**) on Yakushima. In fact, tanuki are invasive on two Japanese islands, the other being Chiburijima in the Oki Dozen Archipelago (National Institute for Environmental Studies, Japan). Both islands are home to UNESCO World Natural Heritage Sites. We have launched projects to investigate their ecological impacts on both.



Figure 5. A pair of tanuki emerge to eat cow feed on Chiburijima, Shimane, Japan. Credit AJJ MacIntosh.

Tanuki Distribution, Behavior, and spread of parasites: To monitor tanuki activity and parasite spread we focused on conducting latrine surveys. Tanuki are a latrine forming species, likely using them for communication within the population. Latrines are also probable sites of parasite transmission, especially for environmentally transmitted parasites such as intestinal worms. We surveyed Yakushima’s western coastal forest for latrine sites, mapped them, and monitored each site for fresh feces semi-daily during field seasons. Latrines were mapped using a combination of QGIS and R, and we used Kernel Density Estimation (KDE) to quantify ‘hot spot’ latrines and test whether parasite abundance correlates with latrine popularity, which would indicate increased infection risk. We also set camera traps to monitor tanuki activity at latrines and identify individuals in the population. We aimed to attach GPS units and accelerometers to tanuki to assess their home range and behavior at higher resolutions, but deployment proved challenging as trapping was unsuccessful for most of the field time.

4. 研究成果

With the SARS-CoV2 pandemic affecting much of FY2020 and FY2021, we were able to conduct fieldwork in Sabah only during FY2022 and FY2023. During the two field seasons, we collected over 600 fecal samples from 5 primate species in the two Sabah study sites. Combined with previous data, our data set now includes over 1800 fecal samples from 9 primates, and we have identified at least 11 species of intestinal helminths in the community (**Fig 6**). We have also shown the interconnectedness of hosts and parasites in this community, as shown in bipartite networks in **Fig 7**. Such results clearly show the value of using molecular techniques, as parasite stages observed in feces cannot often be distinguished among closely related species.

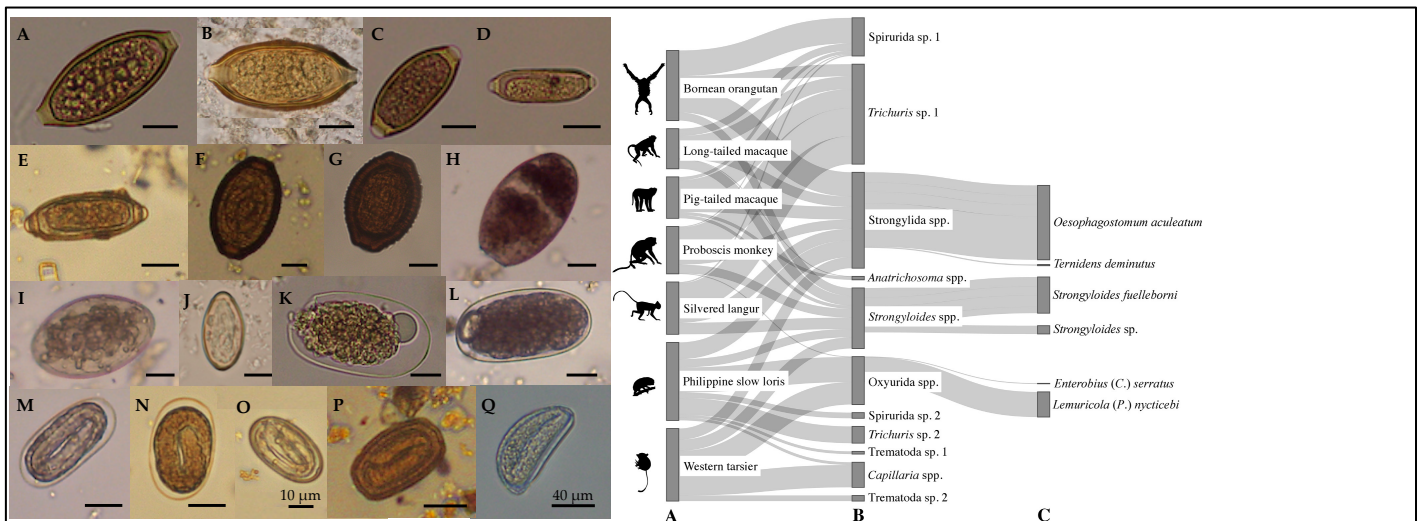


Figure 6 (above left). Helminth eggs found in the feces of Bornean primates. Source: Frias et al (2021) [13].

Figure 7 (above right). Primate-parasite associations, showing (A) primate hosts, (B) observed parasite species richness per host species (detected through microscopy), and (C) hidden parasite species richness per parasite taxonomic group (detected through molecular analyses). Widths of bars correspond to parasite species richness for each primate host (A; range = 4–7) and parasite prevalence (B; range for observed prevalence = 0.6–48.5%). Genetic studies conducted in parallel have revealed higher parasite species richness (C) than that observed by microscopy alone [11,12]. Source: Frias et al. 2021 [13].

We were able to extend our molecular work by using Next Generation Sequencing (NGS) to better assess diversity in one common intestinal worm suborder – Strongylida – which includes potentially pathogenic species [14]. We used the Illumina MiSeq platform to perform ITS-2 strongylid metabarcoding on samples from Sabahan primates and from Japanese macaques for comparison. We found that *Oesophagostomum aculeatum* is the dominant strongyle parasite in all

communities, suggesting these parasite communities in Asian primates are less diverse than those in Africa (Fig 8). This foundational knowledge can assist ongoing monitoring of health threats to primates in different regions.

Current work is now testing the key question, whether primate biodiversity is linked to parasite biodiversity, again using a metabarcoding approach. Answers to our questions are forthcoming and will be published when analyses are completed.

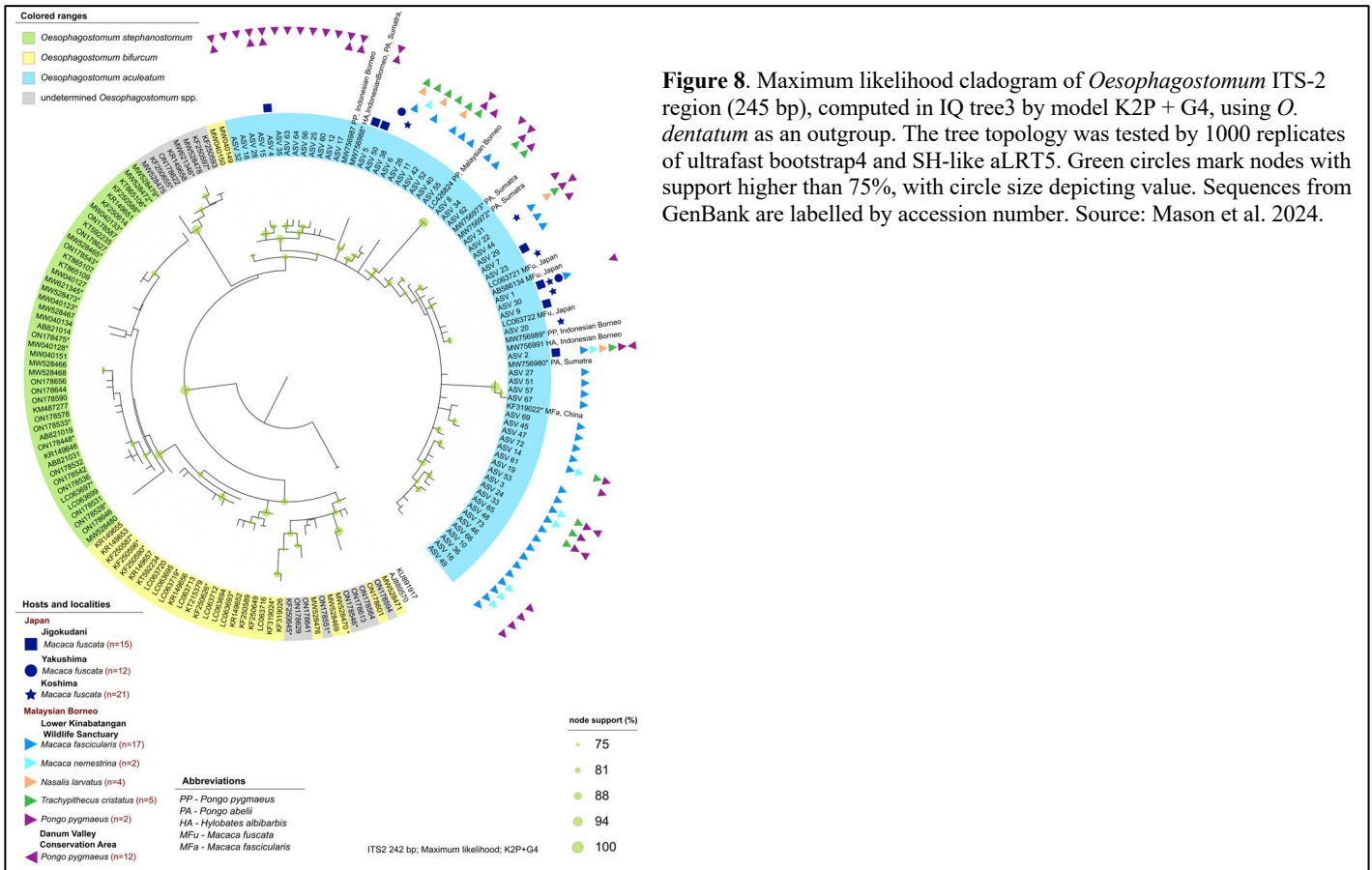


Figure 8. Maximum likelihood cladogram of *Oesophagostomum* ITS-2 region (245 bp), computed in IQ tree3 by model K2P + G4, using *O. dentatum* as an outgroup. The tree topology was tested by 1000 replicates of ultrafast bootstrap4 and SH-like aLRT5. Green circles mark nodes with support higher than 75%, with circle size depicting value. Sequences from GenBank are labelled by accession number. Source: Mason et al. 2024.

Regarding invasive tanuki and their likelihood to spread disease to local fauna in Yakushima, we have collected over 500 fecal samples from latrines at our main study sites and are currently building a machine learning algorithm to automatically detect and count parasites. Preliminary analyses examining the intensity of latrine use and the abundance of the common helminth parasite *Toxocara* sp. do not suggest hotspots of parasite transmission (Fig 9). The next stages of this work will test which parasites might be spread from tanuki to local fauna, another likely outcome of shifting biodiversity’s impacts on ecosystem health.

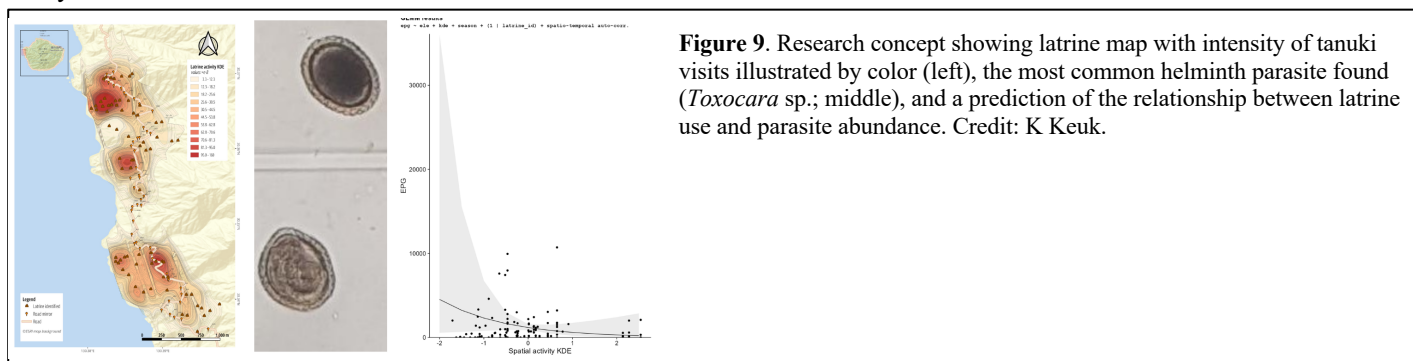


Figure 9. Research concept showing latrine map with intensity of tanuki visits illustrated by color (left), the most common helminth parasite found (*Toxocara* sp.; middle), and a prediction of the relationship between latrine use and parasite abundance. Credit: K Keuk.

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5. 主な発表論文等

〔雑誌論文〕 計19件（うち査読付論文 16件 / うち国際共著 19件 / うちオープンアクセス 15件）

1. 著者名 Sarabian Cecile, Wilkinson Anna, Sigaud Marie, Kano Fumihiro, Tobajas Jorge, Darmaillacq Anne Sophie, Kalema Zikusoka Gladys, Plotnik Joshua M., MacIntosh Andrew J. J.	4. 巻 na
2. 論文標題 Disgust in animals and the application of disease avoidance to wildlife management and conservation	5. 発行年 2023年
3. 雑誌名 Journal of Animal Ecology	6. 最初と最後の頁 na
掲載論文のDOI (デジタルオブジェクト識別子) 10.1111/1365-2656.13903	査読の有無 有
オープンアクセス オープンアクセスとしている (また、その予定である)	国際共著 該当する
1. 著者名 Costa Raquel F. P., Romano Valeria, Pereira Andre S., Hart Jordan D. A., MacIntosh Andrew, Hayashi Misato	4. 巻 5
2. 論文標題 Mountain gorillas benefit from social distancing too: Close proximity from tourists affects gorillas' sociality	5. 発行年 2022年
3. 雑誌名 Conservation Science and Practice	6. 最初と最後の頁 na
掲載論文のDOI (デジタルオブジェクト識別子) 10.1111/csp2.12859	査読の有無 有
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2. 論文標題 TO DRAW OR NOT TO DRAW: UNDERSTANDING THE TEMPORAL ORGANIZATION OF DRAWING BEHAVIOR USING FRACTAL ANALYSES	5. 発行年 2023年
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1. 著者名 Cheron Marion, Kato Akiko, Ropert-Coudert Yan, Meyer Xavier, MacIntosh Andrew J.J., Raelison Lea, Brischoux Francois	4. 巻 254
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1. 著者名 Xu Zhihong、MacIntosh Andrew J.J.、Castellano-Navarro Alba、Macan?s-Mart?nez Emilio、Suzumura Takafumi、Duboscq Julie	4. 巻 10
2. 論文標題 Linking parasitism to network centrality and the impact of sampling bias in its interpretation	5. 発行年 2022年
3. 雑誌名 PeerJ	6. 最初と最後の頁 e14305 ~ e14305
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1. 著者名 Romano Valeria、Sueur Cedric、MacIntosh Andrew J. J.	4. 巻 -
2. 論文標題 The tradeoff between information and pathogen transmission in animal societies	5. 発行年 2021年
3. 雑誌名 Oikos	6. 最初と最後の頁 -
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2. 論文標題 Mountain Gorillas benefit from social distancing too: Close proximity from tourists affects gorillas' sociality	5. 発行年 2022年
3. 雑誌名 EcoEvoRxiv	6. 最初と最後の頁 -
掲載論文のDOI (デジタルオブジェクト識別子) 10.32942/osf.io/ztreq	査読の有無 無
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2. 論文標題 Communication Network Reflects Social Instability in a Wild Siamang (<i>Symphalangus syndactylus</i>) Population	5. 発行年 2021年
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1. 著者名 Xu Zhihong, MacIntosh Andrew J. J., Castellano-Navarro Alba, Macan?s-Mart?nez Emilio, Suzumura Takafumi, Duboscq Julie	4. 巻 -
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1. 著者名 Romano Valeria, MacIntosh Andrew J.J., Sueur Cedric	4. 巻 35
2. 論文標題 Stemming the Flow: Information, Infection, and Social Evolution	5. 発行年 2020年
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1. 著者名 Romano Valeria, Sueur Cedric, MacIntosh Andrew J.J.	4. 巻 -
2. 論文標題 The trade-off between information and pathogen transmission in animal societies	5. 発行年 2020年
3. 雑誌名 EcoEvoRxiv Preprints	6. 最初と最後の頁 -
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1. 著者名 HASEGAWA HIDEO, FRIAS LIESBETH, PETER SURDENSTEEVE, HASAN NOOR HALIZA, STARK DANICA J., LYNN MILENA SALGADO, SIPANGKUI SYMPHOROSA, GOOSSENS BENOIT, MATSUURA KEIKO, OKAMOTO MUNEHIRO, MACINTOSH ANDREW J. J.	4. 巻 4722
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3. 雑誌名 Zootaxa	6. 最初と最後の頁 287 ~ 294
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1. 著者名 Frias Liesbeth, MacIntosh Andrew J. J.	4. 巻 -
2. 論文標題 Global Diversity and Distribution of Soil-Transmitted Helminths in Monkeys	5. 発行年 2020年
3. 雑誌名 Neglected Diseases in Monkeys	6. 最初と最後の頁 291 ~ 322
掲載論文のDOI (デジタルオブジェクト識別子) 10.1007/978-3-030-52283-4_13	査読の有無 有
オープンアクセス オープンアクセスではない、又はオープンアクセスが困難	国際共著 該当する

1. 著者名 Balasubramaniam Krishna N., Sueur Cedric, Huffman Michael A., MacIntosh Andrew J. J.	4. 巻 -
2. 論文標題 Primate Infectious Disease Ecology: Insights and Future Directions at the Human-Macaque Interface	5. 発行年 2020年
3. 雑誌名 The Behavioral Ecology of the Tibetan Macaque	6. 最初と最後の頁 249 ~ 284
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1. 著者名 Mason Bethan, Cervena Barbora, Frias Liesbeth, Goossens Benoit, Hasegawa Hideo, Keuk Kenneth, Langgeng Abdullah, Majewski Kasia, Matsumoto Takashi, Matsuura Keiko, Mendonca Renata, Okamoto Munehiro, Peter Steve, Petrzalkova Klara J., Sipangkui Symphorosa, Xu Zhihong, Pafco Barbora, MacIntosh Andrew J.J.	4. 巻 151
2. 論文標題 Novel insight into the genetic diversity of strongylid nematodes infecting South-East and East Asian primates	5. 発行年 2024年
3. 雑誌名 Parasitology	6. 最初と最後の頁 514 ~ 522
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〔図書〕 計0件

〔産業財産権〕

〔その他〕

MacIntosh Laboratory Website
<https://www.macintoshlab.com>

6. 研究組織

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7. 科研費を使用して開催した国際研究集会

〔国際研究集会〕 計0件

8. 本研究に関連して実施した国際共同研究の実施状況

共同研究相手国	相手方研究機関			
チェコ	Czech Academy of Sciences			
マレーシア	Universiti Malaysia Sabah			