17th International Symposium *on* Primatology *and* Wildlife Science

March 7–8, 2022

Primate Research Institute founded 1967 Kyoto University

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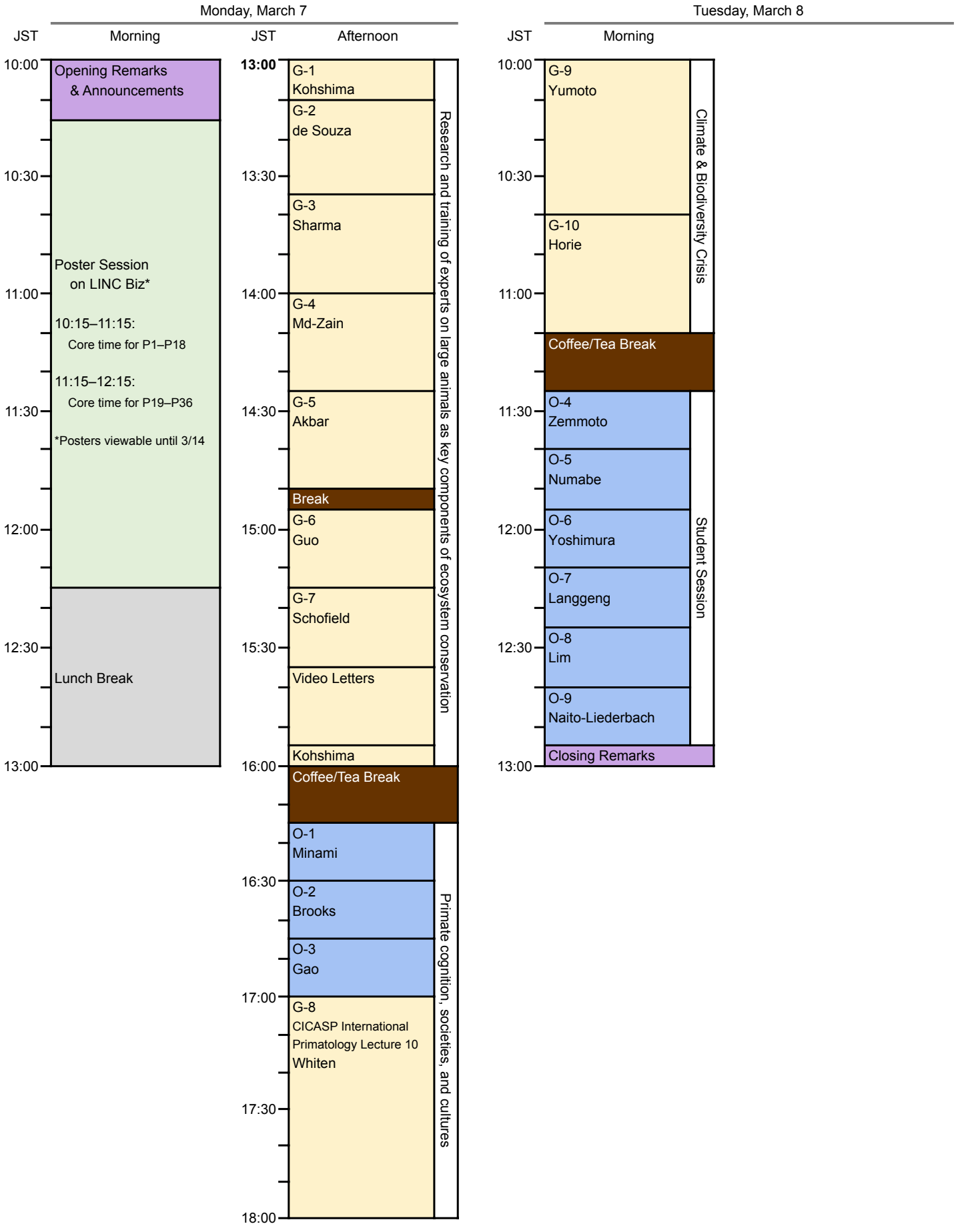
The 17th International Symposium on Primatology and Wildlife Science

Schedule at a Glance (updated Feb. 16, 2022; subject to change)



Cover Photo: Japanese macaque in Yakushima (Credit: Kenneth Keuk)

The 17th International Symposium on Primatology and Wildlife Science

PROGRAM

All times in Japan Standard Time (GMT+9)

Day 1 (Monday, March 7)

Time	Abstract	(min)	Title	Speaker	Affiliation
10:00–10:15		(15)	Opening Remarks & Announcements	Gen'ichi IDANI	Wildlife Research Center, Kyoto University
Online Poster Session					
10:15–12:15		(120)	Posters on LINC Biz [see pp. iii–iv for titles]		
12:15–13:00		(45)	<Lunch Break>		
[JSPS Core-to-Core Program Special Session] Research and training of experts on large animals as key components of ecosystem conservation: The past 10 years and the next 10 years			Co-Chairs: Ikki MATSUDA, Yamato TSUJI, Shiro KOHSHIMA		
13:00–13:10	G-1	(10)	Brief Introduction of Our Program	Shiro KOHSHIMA	Wildlife Research Center, Kyoto University
13:10–13:35	G-2	(25)	Ecology and Conservation of the Amazonian Manatee in Central Amazon, Brazil	Diogo DE SOUZA	Friends of the Manatee Association; Federal University of Rio Grande do Norte
13:35–14:00	G-3	(25)	More Brain than Brawn: An Overview of Elephant Cognition	Nachiketha SHARMA	Kyoto University Institute for Advanced Study
14:00–14:25	G-4	(25)	Behavioral Ecology, Genetics and Human-Monkey Conflict of the Malaysian Primates	Badrul Munir MD ZAIN	Faculty of Science and Technology, Universiti Kebangsaan Malaysia
14:25–14:50	G-5	(25)	Activity, Diet and Ranging of Silvery Lutung (*Trachypithecus cristatus*) for Ecotourism Planning at Gunung Padang	Muhammad Azhari AKBAR	Biology Department, IPB University
14:50–14:55		(5)	<Break>		
14:55–15:15	G-6	(20)	Long-Term Field Study Of Golden Snub-Nosed Monkeys: Combining Traditional Fieldwork With Innovation Techniques	Songtao GUO	School of Life Sciences, Northwest University
15:15–15:35	G-7	(20)	New Tools for Conservation: Automated Behaviour Analysis of Wild Chimpanzees using Deep Learning	Dan SCHOFIELD	School of Anthropology University of Oxford
15:35–15:55		(20)	Video Letters & Comments		
15:55–16:00		(5)	Concluding Remarks	Shiro KOHSHIMA	Wildlife Research Center, Kyoto University
16:00–16:15		(15)	<Coffee Break>		

Primate Cognition, Societies and Cultures				Co-Chairs: Yuko HATTORI, Michael HUFFMAN	
16:15–16:30	O-1	(15)	Are Neonates the Most "Infant-Like"? Early Development of Physical Features in Free-Ranging Japanese Macaques	Toshiki MINAMI	Primate Research Institute, Kyoto University
16:30–16:45	O-2	(15)	The Founder Sociality Hypothesis	James BROOKS	Wildlife Research Center, Kyoto University
16:45–17:00	O-3	(15)	Body Representations in Chimpanzees	Jie GAO	Primate Research Institute, Kyoto University
17:00–18:00	G-8	(60)	**[CICASP International Primatology Lecture]** The Discovery and Reach of Culture in Primates' Lives	Andrew WHITEN	University of St Andrews

Day 2 (Tuesday, March 8)

Time	Abstract	(min)	Title	Speaker	Affiliation
Climate & Biodiversity Crisis					Chair: Gen'ichi IDANI
10:00–10:40	G-9	(40)	The Values of Biodiversity and SDGs	Takakazu YUMOTO	Primate Research Institute, Kyoto University, Japan
10:40–11:10	G-10	(30)	Climate and Biodiversity Crises	Masahiko HORIE	Meiji University
11:10–11:25		(15)	<Coffee Break>		
Student Session					Chair: Susumu TOMIYA
11:25–11:40	O-4	(15)	Study on Adorable Two Dog Breeds' Personalities and Their Genetic Basis	Chika ZEMMOTO	Wildlife Research Center, Kyoto University
11:40–11:55	O-5	(15)	Difference in the Sensitivity to Bitter Compounds in Coffee based on TAS2R Gene Polymorphism	Rena NUMABE	Primate Research Institute, Kyoto University
11:55–12:10	O-6	(15)	Metabarcoding of Fecal DNA Indicates Frequent Plant Consumption in Wild Snow Leopard (*Panthera uncia*)	Hiroto YOSHIMURA	Wildlife Research Center, Kyoto University
12:10–12:25	O-7	(15)	Seasonal Variation of Gastrointestinal Helminth Infection in Japanese macaques of the Jigokudani Snow Monkey Park	Abdullah LANGGENG	Primate Research Institute, Kyoto University
12:25–12:40	O-8	(15)	Demographic History of the Endangered Malayan Tapir: A Whole-Genome Resequencing Approach	Qi Luan LIM	Wildlife Research Center, Kyoto University
12:40–12:55	O-9	(15)	Age but Not MHC Similarity Predict Reproductive Success of Captive Japanese Golden Eagles	Annegret Moto NAITO-LIEDERBACH	Wildlife Research Center, Kyoto University
12:55–13:00		(5)	Closing Remarks	Hiroo IMAI	Primate Research Institute, Kyoto University

Online Poster Session

on LINC Biz

Monday, March 7, 10:15–12:15

**During their core presentation times, presenters are requested to be available on LINC Biz for live chats with the audience*

Core presentation time* 10:15–11:15		
P-1	Naoto TOYODA	Evolutionary Patterns of the Facial Part of the Cranium in Strepsirrhines: Combining Geometric Morphometrics and Phylogenetic Comparative Methods
P-2	Christen LIN	Emotional Contagion in Chimpanzees and Bonobos
P-3	Elio BORGHEZAN	Effect of Light Bias on Male Mating Signal and Female Mate Choice in a Sexually Dimorphic Amazonian Fish, the Sailfin Tetra (*Crenuchus spilurus*)
P-4	Sok Hwan LEE	Effect of a Novel Skill in Social Rank of Chimpanzees and Bonobos
P-5	Takuto SUGIMOTO	Do Horses Evaluate Humans Based on Skillfulness?
P-6	Kushaal SELVARAJAH	Comparative Study on the Social Behavior of Sambar Deer (*Rusa unicolor*) in Three Selected Captive Facilities in Peninsular Malaysia
P-7	Chisato TANAKA	Evaluating Stressors in Captive Tsushima Leopard Cats (*Prionailurus bengalensis euptilurus*) Based on Stress Hormone Concentration and Behavioral Changes
P-8	Kana ARAI	Application of Methylation-Sensitive High-Resolution Melting (MS-HRM) to Estimate Epigenetic Age in Captive Asian Elephants (*Elephas maximus*)
P-9	James BUKIE	Activity Budget of Semi-Captive Drill Monkeys at Drill Ranch, Cross River State, Nigeria
P-10	Shenwen XU	Generalization of Video-Referent Correspondence in Chimpanzees
P-11	Yeongju LEE	Social Associations of Mothers and Offsprings in Free-Roaming Horse Groups
P-12	Yuko KURIYAMA	How Can We Get Information from Old Fecal Samples of Iriomote Cats?: Methods for Species Identification, Sex Determination, and Hormone Measurements
P-13	Heping LI	Influences of Oceanographic Features on the Northbound Migratory Movement of Northern Fur Seals (*Callorhinus ursinus*)
P-14	Ena ONISHI	Contagious Urination among Captive Chimpanzees: Investigation of Social Centrality of Initiator and Follower
P-15	Alexander HENDRY	Are Semi-Arboreal Felids at Greater Risk from Habitat Disturbances than Terrestrial Felids? The Ecology of the Marbled Cat in Southeast Asia
P-16	Yige PIAO	Do Pit Vipers Assess Their Venom? Defensive Tactics of *Deinagkistrodon acutus* Shift with Changed Venom Reserve
P-17	Seda KAVLAK	The Social Networks and Hierarchy of a Captive Group of Orphaned Infant Chimpanzees
P-18	Kenneth KEUK	Studying the Effects of a Raccoon Dog Invasion of Yakushima on the Ecology of Disease, from the Ground Up (to the Sky): a Pilot Study

Core presentation time* 11:15–12:15		
P-19	Boyun LEE	Asymmetry of Early Social Interactions between Infants and Non-Mother Individuals in Yakushima Japanese Macaque (*Macaca fuscata yakui*)
P-20	Akihiro ITAHARA	Tracking Visual Attention of Crows Using a Motion Capture System
P-21	Hayate TAKEUCHI	Comparative Transcriptome Analysis in Stranded Cetaceans Provides Novel Insights into the Evolution of Lipid Utilization
P-22	Asuka NISHIJIMA	Transcriptome Changes of Maternal Immune System Related to Embryonic Diapause in Brown Bears
P-23	Hinako KATSUSHIMA	Possibility of Gustation Being Used during Breastfeeding by Cetacean Infants
P-24	Kohei KANETSUNA	Comprehensive Analysis of the Gut Microbiome in Gum-Eating Mammals
P-25	Ayumu SANTA	Training of Matching to Sample Task for Killer Whales (*Orcinus orca*)
P-26	Tamao MAEDA	Comparing the Social Relationships of Feral Horses during Breeding and Non-breeding Season Using Drones and GPS Tags
P-27	Hideki SUGIURA	Exploratory Vegetation Survey by UAV in Yakushima Island
P-28	Akiho MURAMATSU	Longitudinal Working Memory Study in Chimpanzees
P-29	Qi Luan LIM	Population Genetic Structure of the Malayan Tapirs in Peninsular Malaysia Revealed by Nine Cross-species Microsatellite Markers: Genetic Diversity Compared to the Japanese Ex-situ Population
P-30	Yoshiyuki TABUSE	Rest Site Selection Analysis in Japanese Macaques
P-31	Juno SHIMADA	Molecular Evolution of Sweet Taste Receptor Genes in Elephants
P-32	Shiina SAKAMOTO	Genetics, Behavior, and Evolution of the Visual Adaptation in Egg-Laying Mammals
P-33	Gema PALACINO-GONZALEZ	The Role of Intentional Communication in a Cooperative Activity in Horses (*Equus caballus*)
P-34	China KOBAYASHI	How Do Cats Move Through Forests? : the Analysis of Capture Rate by Camera Traps
P-35	Katherine MAJEWSKI	All-You-Can-Eat: A Preliminary Study of Invasive Raccoon Dog (*Nyctereutes procyonoides*) Predation of Endemic Species and Dietary Competition on Yakushima Island, Japan
P-36	Satsuki SUZUKI	Estimating Epigenetic Age in Okinawa rail (*Hypotaenidia okinawae*) using Methylation-Sensitive High-Resolution Melting (MS-HRM)

PWS Symposia Special Session for JSPS Core-to-Core program of WRC

RESEARCH AND TRAINING OF EXPERTS ON LARGE ANIMALS AS KEY COMPONENTS OF ECOSYSTEM CONSERVATION

- THE PAST 10 YEARS AND THE NEXT 10 YEARS -

7th March 2022 13:00～16:00
Online broadcast (Registration required)

Outcomes of the program to-date

Ecology and conservation of the Amazonian manatees in central Amazon, Brazil
Diogo Alexandre de Souza
Federal University of Rio Grande do Norte

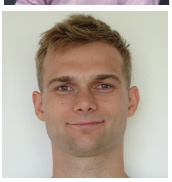
More brain than brawn: An overview of elephant cognition
Nachiketha Sharma
Kyoto University

Behavioral ecology, genetics and human monkey conflicts of the Malaysian primates
Badrul Munir Md-Zain
Universiti Kebangsaan Malaysia

Activity, diet and ranging of silvery lutung for conservation management planning at Gunung Padang
Muhammad Azhari Akbar
IPB University

The future of conservation research: applying innovative technologies

Long-term field study of golden snub-nosed monkeys: combining traditional fieldwork with innovative techniques
Songtao Guo
Northwest University



Automated behaviour analysis of wild chimpanzees using deep learning
Daniel Schofield
University of Oxford

Coorganized by
JSPS core-to-core programs, CCT-Bio (2012-2017) & CET-Bio (2017-2022)
JSPS Primatology and Wildlife Science (PWS)
Supported by
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Chubu University Academy of Emerging Sciences (CUAES)

Brief Introduction of Our Program

Shiro Kohshima
Wildlife Research Center, Kyoto University, Kyoto, Japan

Our program, the JSPS Core-to-Core Program “International Core of Excellence for Tropical Biodiversity Conservation Focusing on Large Animal Studies” started from April, 2017. It is a five-years program supported by the Japan Society for the Promotion of Science (JSPS) to promote academic exchange on tropical biodiversity conservation among seven countries; Japan, Brazil, India, Malaysia, China, Indonesia and UK. We started this program to extend and develop the international network established by our previous JSPS core-to-core program ”International Network for Tropical Biodiversity Conservation Focusing on Large Animal Studies” that promoted academic exchange among Japan, Brazil, India and Malaysia for 5 years from April, 2012.

In our program, we focus on large animal studies because many of them are umbrella species and flagship species important for biodiversity conservation though we never eliminate the studies on other organisms. We have promoted collaborative studies on various wildlife as equal partners. Especially, we have promoted development and adaptation of new research technologies for wildlife studies, such as genome analysis, environmental DNA, bio-logging, sound/image analysis and drone use. We have also held training courses for wildlife studies every year inviting young researchers from the participant countries to Yakushima-island and Kyoto, Japan.

We have organized an international workshop every year in one of the member countries. This workshop included a symposium and a study tour not only to share the results of collaborative studies but also to develop and share the idea of “Field Museum”, a new concept of next generation Zoo/Aquarium tightly connected with the natural habitats of local wildlife. Field Museum can contribute to the study, conservation and education of the wildlife in that area. It can also contribute to the local economy through eco-tourism, for example. In each workshop, we have deepened and shared the understanding on the study and conservation of wildlife in the host country and discuss how to realize Field Museum in each country. Based on this academic exchange by our program, the first Field Museum in Amazon was realized in Manaus, Brazil in 2019. In the end of this special session, we will discuss how to develop our international collaboration for the study and conservation of wildlife in next 10 years.

Ecology and Conservation of the Amazonian Manatee in Central Amazon, Brazil

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Amazonian freshwater ecosystems are undergoing large-scale alteration through widespread land cover changes, construction of hydropower dams, overfishing, mining and pollution, making the Amazonian manatee (*Trichechus inunguis*) among the most vulnerable species. In addition, direct human activities (e.g., hunting and fisheries-related accidental mortality) constitute an unprecedented set of threats to the persistence of this endemic species. However, a major obstacle to reduce the effects of these threats and support conservation strategies has been the scarce information about ecology and population data for the species. *T. inunguis* is extremely difficult to observe in the wild, and the limitation of sampling methods has been a challenge for the conservationists. In this talk, I will provide a short view about the initiatives to reduce the ecological information gap for the Amazonian manatee and to promote its conservation in a Sustainable Development Reserve, located in the Purus River basin, Brazilian Central Amazon. Three main topics are addressed: (i) mortality of manatees and cultural perception of local people about the conservation prospects of species; (ii) advances and challenges to study their ecology in the wild, and (iii) what is being done to conserve the species. A multi-methodological approach has been used to collect data: interviews with fishermen and former manatee hunters; direct/indirect observations of the species analyzed with hierarchical models based on imperfect detection to estimate their occupancy, and release and monitoring of manatees. First, direct capture using harpoons continues to be the main cause of Amazonian manatee mortality. However, there has been a decrease in mortality events after the year 2000, which coincides with the creation of sustainability initiatives taken by communities such as management of fishing resources. Second, within 265h of effort, 91 manatee's observations were recorded. The manatee's detection probability had never been estimated before: surprisingly, it was high ($p=0.50$, $SD=0.05$) and positively related with macrophyte coverage of the sampling sites. Contrary to our prior hypothesis, the results suggest that the studied communities resident impact is not affecting the manatee occupancy, with greatest probabilities closer to human settlements, possibly related to greater connection of this region with other water bodies and Purus river main channel. Third, released manatees are successfully adapting to the wild, including the confirmation of pregnancy from one female manatee; movements, seasonal habitat use and home range of the individuals monitored by radiotelemetry will also be informed. This study is a timely opportunity to provide insights and scientific information to gain a better understanding of the manatee ecology, and provide subsidies for managers and local communities to establish an effective protection plan for the species in the Reserve. Our experience shows that a combination of releases and environmental education activities is an effective conservation tool for the *T. inunguis*.

More Brain than Brawn: An Overview of Elephant Cognition

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The extant proboscideans are the largest terrestrial animals weighing around 4-6 tons. They are long lived social species with strong bonds between individuals, display complex communication (from short distance tactile to long distance auditory, olfactory) systems and show high fission-fusion dynamics. Along with the societal complexity, the absolute brain size (5.5-6.5 kgs depending on the species) of elephants is largest among land animals. These unique features make elephants one of the best model species to study cognition. Understanding the elephant cognition has multiple advantages. Purely from the perspective of comparative psychology, we can gain deeper insights on the evolution of cognition in phylogenetically distant taxa such as elephants, primates, birds and several other species. From the practical point of view, elephant cognition can play a huge role in the welfare, management and conservation of both captive and wild elephants. In this talk, I will provide a broad overview of our understanding of elephant cognition by highlighting a few classic studies conducted in both wild and captive settings. I will also briefly discuss possible challenges faced while studying elephant cognition and its conservation applications.

Behavioral Ecology, Genetics and Human-Monkey Conflict of the Malaysian Primates

Badrul Munir Md Zain[1*], Mohd Faudzir Najmuddin[2], Muhammad Abu Bakar Abdul-Latif[2], Siti Norsyuhada Kamaluddin[1], Norlinda Mohd-Daut[1], Farhani Ruslin[1] and Ikki Matsuda[3,4,5,6]

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Behavioral ecology, genetics and human-monkey conflict studies were conducted on Malaysian langurs, long-tailed macaque and Bornean orangutan. We presented here some findings on selected Malaysian primate involving research collaboration between Malaysian and Japanese primatologists. Feeding ecology study of *Presbytis femoralis* indicated that over 75% of fruit feeding involved consuming cultivar plants planted in human settlement. Feeding ecologies of two sympatric primates, the long-tailed macaque and the dusky langur, in a mixed landscape consisting of urban and agro-forested areas and forest fragments in Malaysia showed that 59 consumed plant species common to both species, in which the dietary overlap was the highest for fruits. We also found that provisioning had negative effects on the ecology of silvery langur in the Bukit Malawati Kuala Selangor. Our observation on daily behavior of the Bornean orangutans at Bukit Merah Orang Utan Island indicated that they engaged in less feeding but more resting, and show less postural diversity. We also managed to genetically identify captive Bornean orangutans at subspecies level for captive management purposes. Overall, a proper management with conservation intervention are needed to manage primates that are living near human settlements.

Activity, Diet and Ranging of Silvery Lutung (*Trachypithecus cristatus*) for Ecotourism Planning at Gunung Padang

Muhammad Azhari Akbar[1*], Dyah Perwitasari-Farajallah[1,2], Rizaldi[3], Ani Mardiastuti[4], Try Surya Harapan[5] and Yamato Tsuji[6]

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Ecotourism has a positive synergy which combine tourism activity and biodiversity conservation. With appropriate management, it also sustains the well-being of the local people, involves interpretation and education. We present the preliminary results of an initiative of ecotourism at Gunung Padang, West Sumatra, Indonesia, with special attention to silvery lutung (*Trachypithecus cristatus*), as one of primate species inhabiting natural forest there. We conducted behavioral observations of the lutungs between 2018 and 2019 (total observation time: 482 hr), and recorded activity budgets, dietary profile, and ranging patterns, all of which would be used as a baseline for an ecotourism planning. We found the subject group spent most of time resting (average 45% of the daytime). Resting peaked in the morning, also in the afternoon while moving and feeding decreased in this period. In several seasons they spent longer time feeding. We recorded 74 items from 37 different plant species as the lutungs' diet. The lutungs mainly fed on young leaves (68%), but they seasonally fed much on ripe and unripe fruits. Home range size of the target group was 7.3 ha, which represented approximately 36.8% of the total of study site. Monthly range size has increased between January -July 2019. Gunung Padang has a high potential as ecotourism area through this scientific study which conserve the lutungs as a protected primate species by education and public awareness, also promote social and economic benefits for local communities by sharing the daily life of local people in coastal area at Gunung Padang which can coexist well with primates and primate-watching program. It may also aid reducing conflicts between human (in term of local people and tourist) and primates, which has become a major problem at Gunung Padang.

Keywords: activity, diet, ecotourism, range, instantaneous scan sampling

Long-Term Field Study of Golden Snub-Nosed Monkeys: Combining Traditional Fieldwork with Innovation Techniques

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The reporter' study is conducted on the Golden snub-nosed monkey (GSM) in the Qinling Mountains, mainly engaged in wildlife behavior, ecology and conservation. We follow the monkeys for more than 20 years in field observation and research and try to understand their behavior, society and adaptation. We discovered some knowledge of diet, home range and population structure with traditional field investigation method, as well we recently also borrow new techniques to reveal more knowledge of them than before: 1) we developed an individual face identification system based on Tri-AI technology for GSM and other mammal species; 2) we used unmanned aerial vehicle drones with thermal camera to carry out field surveys for GSM in the Qinling Mountains, and achieve the requirements of quickly detecting and measuring the size of the wild primate populations in mountainous areas; 3)through thermal physiology and nutritional ecology methods, we have systematically revealed the feeding strategy, nutritional compensation and energy balance mechanism for GSM in the cold environment;4) we use genetic method study genetic kin relationships and social structure of wild groups of GMS, and revealed the mating system and population stability mechanism for them; 5) through meta-genomics analysis, we find out how gut microbes help host digest high fiber food; except these basic biological and ecological study, we have also participated in the conservation project titled "Forest Ecology Restoration Project in the Qinling Mountains" jointly funded by Northwestern University and COSMO Petroleum Fund of Japan, planting trees in abandoned areas of forest farms and contributing to the conservation planning, habitat restoration and corridor construction of GSMs.

Our team has been cooperating with Japanese scientists for more than 20 years, spanning two generations. We will continue to participate in and actively promote international cooperation on global biodiversity conservation and persistently contribute to the construction of a better planet.

New Tools for Conservation: Automated Behaviour Analysis of Wild Chimpanzees using Deep Learning

Daniel Schofield*

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Video datasets are increasingly important for conservation and ethology, however large-scale analysis of animal behaviour is limited by time and resources. I outline a novel framework using artificial intelligence (AI) to perform individual identification and behaviour recognition of wild chimpanzees for analysis of social behaviour. I review the strengths and limitations of such an approach, and discuss the future directions and challenges for widespread adoption of the technology in conservation and research.

The Discovery and Reach of Culture in Primates' Lives

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In the present century the study of animal cultures has veritably exploded, revealing that culture – in the sense of an array of socially transmitted traditions shared by a community – is widespread across the animal kingdom: it is pervasive through some species' lives, and spans most domains of behaviour one can think of, from foraging skills to tool use, vocal repertoires and migratory pathways (Whiten 2021).

But these discoveries are based on foundations dating back seven decades, and primatology played a major role in these, beginning of course with the work of Imanishi, Itani, Kawamura, Kawai, and colleagues on the 'precultures' of Japanese macaques. Some quite different research on birdsong dialects and avian foraging innovations provided important foundations too, but initially failed to connect by any cross-referencing between this work and the primatological discoveries. The bigger picture of animal culture has become appreciated only much later.

Primatology continued to contribute breakthroughs that led and shaped this bigger picture of animal culture through the latter parts of the twentieth century and beyond, notably through field research with chimpanzees, again pushed forward by major Japanese figures but also by a more international array of inspiring primatologists.

In this talk I will first offer a personal overview of the progressive discoveries that shaped our understanding of the scope and significance of primate culture through the last 70 years or so. I then survey what we have come to recognize as the substantial 'reach' of culture in the lives of primates and other animals (Whiten 2017a, 2017b). This includes culture's reach across a diversity of species of primate, across a diversity of types of behaviour, across a diversity of cognitive processes, and extending through primates' lifetimes (Whiten and van de Waal 2018).

Whiten, A. (2017a). Social learning and culture in child and chimpanzee. *Ann. Rev. Psychol.* 68, 129-154.

Whiten, A. (2017b). How culture extends the scope of evolutionary biology in the great apes. *Proc. Natl. Acad. USA* 114, 7790-7797.

Whiten, A. (2021). The burgeoning reach of animal culture. *Science*, 372, eabe6514.

Whiten, A. & van de Waal, E. (2018). The pervasive role of social learning in primate lifetime development. *Behav. Ecol. Sociobiol.* 72, 80.

The Values of Biodiversity and SDGs

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There are several obstacles to spreading the importance of the biodiversity concept to the business world. One of them is the "apparent" biodiversity problem. Biodiversity issues are often reduced to the conservation of rare species or the extermination of alien species. What is the point of investing large amounts of taxpayers' money to bring back extinct animals like the crested ibis or the oriental stork? Or why do we have to spend a lot of manpower to exterminate black bass and lanceleaf tickseed, which have already become familiar to us? Another "apparent" biodiversity problem is the question of whether we should protect useless and even harmful species. Modern agriculture is based on the extreme reduction of biodiversity by eliminating weeds and pests in order to efficiently produce only certain "exotic" crops. In addition, modern medicine often talks about eliminating pathogens and the organisms that transmit them as well.

The value of biodiversity can be divided into three categories: 1) heritage value, 2) functional value, and 3) indicator value. What I would like to focus on here is the indicative value. The existence of ecosystems rich in biodiversity is the source of a safe and secure world for human society and a human society with high resilience against various disturbances such as climate change and large-scale disasters. The crested ibis and the oriental stork are not only "flag species" which attracts popularity, but also "umbrella species" which covers the functions of a whole ecosystem. Rural landscapes with crested ibis and oriental storks are the places that nurture rich biota and ensure that we can obtain safe and healthy agricultural products.

Recently, the concept of SDGs has been gaining attentions widely. Of course, each of the 17 goals is important for building a sustainable planet and human society, but the most important message of the SDGs is an attempt to the simultaneous achievement of all 17 goals. Otherwise, we will not be able to reach the ultimate goal of "a world where no one is left behind". The 17 goals can be categorized into 4 goals for the biosphere, 8 goals for human society, and 4 goals for the economy. So far, economic development has been the top priority among the policy makers and most of our society, and human society and the biosphere have been "left behind". The right way to use the SDGs is not to claim that your business is making a significant contribution to any one goal, but to check whether it is making a negative impact on other goals. Rather, the list of goals in SDGs is to check whether your business has a negative impact on any 17 goals. Otherwise, we will end up with a pretended environmentalism i.e. "SDG wash" that emphasizes the contribution to one goal and ignores or hides the negative impact on other goals.

Our society is on the verge of a major game change, a "new capitalism". We must realize that corporate entities that continue to "exploit" workers and also ecosystems without paying proper returns and fail to fulfill their responsibilities as a member of our planet will no longer be chosen for investment in the rapid economic recovery after COVID-19.

Climate and Biodiversity Crises

Prof. Masahiko Horie
Former Ambassador for Global Environmental Affairs
Special Advisor to the President of Meiji University

The Planet and Humans are at a crossroads. We are confronting with two globally imminent crises of climate and biodiversity.

The damages caused by climate change are quite serious and the State parties of UNFCCC are committed to achieve carbon neutral under Paris Agreement.

This climate crisis is accelerating the speed of biodiversity loss and worsening another crisis of biodiversity.

Based on the presentation by Prof. Yumoto who will focus on the value of biodiversity and SGDs, I will focus more on the international efforts facing global warming under the Paris Agreement.

We need to double our efforts to save this planet and all the peoples of the world must uphold and achieve the Sustainable Development Goals.

Are Neonates the Most "Infant-Like"? Early Development of Physical Features in Free-Ranging Japanese Macaques

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Human infants have distinctive physical features compared to adults. Infants with more prominent infant-like features are more attractive to adults, enhancing their caretaking motivation. Interestingly, human infants are most attractive at around the age when their mobility begins to develop. This might be because infants with immature mobility may need strong attention from their caretakers and protection from danger. In nonhuman primates, infant-like physical features are believed to attract the caretakers. It might also be adaptive for nonhuman primate infants that physical features become the most infant-like at around the age when they begin to explore their environment, but their mobility is immature. However, no studies have quantitatively assessed how infant-like the infants are and examined their developmental changes in nonhuman primates during the early postnatal period. This study aimed to quantify the infant-like physical features in Japanese macaques (*Macaca fuscata*) and examine their developmental process until 12 weeks of age. We studied the provisioned Japanese macaques at Arashiyama Monkey Park Iwatayama in Kyoto. By photographing the target individuals, we quantitatively measured their physical features of the frontal face, profile, whole body, and body color without any contact with the animals. First, we identified physical features typical of infants in Japanese macaques by measuring all individuals in the group and quantitatively evaluated how infant-like features the targets had in the frontal face, profile, whole body, and body color. Next, we examined the developmental changes of infant-like physical features by measuring infants within 12 weeks of age once a week. The results showed that the degree of the infant-like features of the profile, whole body, and back hair color decreased with growth. In contrast, the degree of the infant-like features of the frontal face and face color increased with growth. This result raises three possibilities: (1) infants who have immature mobility most attract caretakers using their infant-like faces, (2) various physical features indicate developmental stages of infants, (3) this result is merely a byproduct of anatomical development. Future research is needed to examine the relationship between infant-like physical features and caretaking behaviors that infants receive.

The Founder Sociality Hypothesis

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In this review, we propose that the social dynamics of founder populations in novel and newly available environments can have critical effects in shaping species' sociality and can produce long-lasting changes in social structure and behavior. For founder populations which expand into an underexploited niche separated from the parent population, the necessity of bond formation with strangers, lack of clear territories, and initial abundance of resources can lead to altered initial social dynamics to which subsequent generations adapt. We call this the founder sociality hypothesis. After specifying the theoretical reasoning and mechanism of effect, we focus on three particular cases where the social dynamics of founder populations may have a central role in explaining their modern behavioral ecology. In particular, we develop and review evidence for three predictions of the founder sociality hypothesis in territorial, mixed-sex group forming species: relatively stronger social bonds in the dispersing sex with relatively weaker bonds in the nondispersing sex, reduced territoriality, and increased social tolerance. We briefly touch on the implications for human evolution given our species' evolutionary history marked by frequent expansion and adaptation to novel environments. We conclude by proposing several experiments and models with testable predictions following from the founder sociality hypothesis.

Body Representations in Chimpanzees

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A representation of typical bodies could help with species recognition, as well as gesture and action understanding in animals. However, little is known about non-human animals' body representation. We studied chimpanzees, humans' closest living relatives, hoping to provide insights on their body representation and its evolution. In Study 1, we used eye-tracking tasks to examine chimpanzees' attention towards typical and atypical bodies. Compared to the looking times toward the normal arms and legs, chimpanzees had significantly longer looking times toward the human arms and legs on chimpanzee bodies. The looking times toward the wrongly-placed arms and legs on chimpanzee bodies were also longer than the normal parts, but the difference just failed to meet significance. These results suggest that chimpanzees might have a body representation that is sufficiently sensitive to detect these aspects of strangeness. In Study 2, we further tested chimpanzees' body part categorization using touchscreen tasks. The results of the training phase of this ongoing study will be reported. Chimpanzees are trained to choose the same body part on a chimpanzee body in a matching-to-sample task, until they reach the preset criteria. Among the wrong choices, they had the most choices in the arm-leg pair, suggesting that their discrimination of arms and legs may not be that clear, compared to other body part pairs. These results could help understand chimpanzees' knowledge about body structures and its evolutionary path.

Study on Adorable Two Dog Breeds' Personalities and Their Genetic Basis

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The personality of an individual is a factor to be considered in maintaining health. Understanding the personality of animals may be useful for the welfare of mute animals. With this in mind, this study analyzed the relationship between personality and genetics in a familiar animal species, the dog (*Canis lupus familiaris*).

In this study, we focused on two of the most familiar dog breeds, the Toy Poodle and the Dachshund. A questionnaire survey was first conducted among owners to assess the personality of individual dogs, and then the association between these characteristics and genes was examined. A total of 233 toy poodles and 161 dachshunds were used for personality assessment. Different personality factors were found between the Toy Poodles and Dachshunds. These may be due to selection during the breeding process. We suggest that breeds be considered in future studies of canine personality.

Genome-wide association study (GWAS) was conducted for each personality factor in the genome-wide approach, and GWAS revealed genetic variants that may be associated with personality in Dachshunds. It suggests 2 genes (*PAK5*, *KIRREL3*) possibly are associated to the personality. To understand dogs' personality from genetic data, more knowledge of those genes and other breeds are needed.

Difference in the Sensitivity to Bitter Compounds in Coffee based on TAS2R Gene Polymorphism

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Introduction. Coffee is one of the most consumed beverages worldwide despite its bitterness. Its consumption has prospective effects on various aspects. I want to know about the influence of polymorphism in human to their perception of bitter taste of coffee. There are some genes whose SNPs can be related to drinking behavior of coffee such like TAS2R43, CYP1A1, and CYP1A2. I mainly focus on TAS2R43, one of the bitter receptors called TAS2Rs. Besides, it is reported that other substances in coffee have stronger influence on bitterness of coffee than caffeine does, and their receptors are TAS2R43 and -46. Both TAS2Rs have SNPs in their coding sequences, with major 2 haplotypes called "ancestral type" and "derived type." It has been unclear the effect of these SNPs to human bitter sense of coffee, and the data of human bitter sense has been gathered by questionnaires based on participants' image in previous study. In this study, I examined the relationship between TAS2R43 and -46 functional difference based on polymorphism and their phenotype as human taste sense.

Methods. The haplotype effect to each TAS2R function was investigated by calcium assay using HEK cells. TAS2Rs genotypes of the participants were identified by PCR and DNA sequence using DNA samples extracted from their oral swab samples. In order to gather the data of the threshold of people's feeling bitterness, taste test with tasting caffeine and coffee solution was conducted.

Results. In both of TAS2R43 and -46, ancestral types responded higher to the bitter substances than derived type. There was a tendency that TAS2R43-ancestral carriers can percept lower caffeine concentration than TAS2R43-derived carriers. The combination of the SNPs in TAS2R43 and -46 showed a tendency that the carriers percept caffeine bitterness at lower concentration than others.

Discussion. W35 in the ancestral haplotype of TAS2R43 is responsible to keep higher response to various bitter substances. Because amino acid residue is in the intracellular region, the change of its reaction can be caused by the change of the localization or the interaction with G protein. L228 in the ancestral type of TAS2R46 is responsible to keep higher response of the receptor to various bitter substances. Because the amino acid residue is in the transmembrane region, the change of its reaction can be caused by the change of the binding site, the localization, or the interaction with G protein. There was no tendency about bitter perception of coffee. This suggests that bitter perception of coffee can be affected by other bitter substances response to other TAS2Rs. Interestingly, the frequency of the SNPs in the bitter taste receptors are different among ethnic groups and it is similar to the SNP in CYP1A2, a caffeine catabolic enzyme. This suggests that these genes can be coevolved associated to caffeine intake in human expansion.

Conclusion. SNPs in TAS2R43 and -46 can affect the response to caffeine. The bitter receptors and CYP1A2 may be coevolved in human expansion associated to caffeine intake by changing the bitter taste sensitivity and metabolic responses, respectively.

Metabarcoding of Fecal DNA Indicates Frequent Plant Consumption in Wild Snow Leopard (*Panthera uncia*)

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Felids (colloquially called cats) are the obligate carnivores, but they sometimes eat plants both in the wild and in captivity. However, little is known about their interaction with plants. A big cat, snow leopard (*Panthera uncia*) inhabits high mountains in central Asia. Interestingly, their scats contain plant materials in the wild more frequently than other *Panthera* species. Although no previous study focused on dietary plant species, grasses and *Myricaria* bush were mentioned in the prey animal survey (Fox and Chundawat., 2016). Identification of plant species leads to figure out the interaction between carnivores and plants in alpine ecosystems and understand the mechanism and adaptation of plant eating in carnivores.

In this study, we aimed to identify prey and plant species in wild snow leopard scats using a DNA metabarcoding method. DNA-identified scats of wild snow leopards in Kyrgyz Republic were investigated (37 samples for detection of vertebrates, 34 samples for plants). After whole scat DNA was extracted, five DNA barcode regions (*12SV5* for prey vertebrates and *ITS2*, *rbcL*, *trnL*, *ITS1* for dietary plants) were amplified by PCR and then massively sequenced by a Illumina MiSeq. Probabilistic co-occurrence analysis of vertebrates and plants was conducted to check the effect of gut contents of prey vertebrates.

We found prey species was dominated by Siberian ibex (*Capra sibirica*) and Argali (*Ovis ammon*) as previously reported (Jumabay-Uulu et al., 2013). The family *Poaceae*, *Tamaricaceae* (genus *Myricaria*), *Asteraceae* were dominant plant taxa in snow leopard scats. Significant co-occurrence was detected between Argali and some *Poaceae* species. However, plants including *Myricaria* species were often detected from samples which did not contain vertebrate DNA. This indicates that detected plants were not always derived from gut contents of prey vertebrates and snow leopard voluntary eat plants.

Focusing on dietary plant species of carnivores will be a new perspective to understand animal-plant interaction and may provide new information to evaluate the importance of plant species in the ecosystem.

Seasonal Variation of Gastrointestinal Helminth Infection in Japanese macaques of the Jigokudani Snow Monkey Park

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The Japanese macaque is the most northerly-living non-human primate, distributed from warm-temperate to cool-temperate forests across Japan. Like all primates, Japanese macaques host several kinds of gastrointestinal parasites. Parasite infection is generally driven by temporal changes related to seasonal variations in environmental conditions and host characteristics. The impact of seasonality on gastrointestinal (GI) helminth infection in Japanese macaques has been investigated in multiple habitats. Here, we quantified seasonal variation in GI helminth load in Japanese macaques residing at one of the higher altitude and latitude of their distributions: The Jigokudani Snow Monkey Park in Nagano. We collected 253 fecal samples opportunistically from sixteen adult female macaques during three study periods: winter 2019 – 2020, summer 2020, and winter 2020 – 2021. Our results showed similar patterns of infection with past studies conducted in different habitats where *Trichuris trichiura* infection was noticeably higher in winter than in summer, while *Oesophagostomum aculeatum* and *Strongyloides fuelleborni* infections were higher during summer. To our surprise, *Streptopharagus pigmentatus* was more prevalent and abundant in winter. The nematode requires coprophagous beetles, which are unlikely to be abundant during cold seasons, as intermediate hosts to complete their life cycle. Further studies on the life history of *S. pigmentatus* and the seasonality of the intermediate hosts are necessary to understand the unexpected increase. Moreover, complete seasonal investigations in the study site and other populations could provide additional information on the parasite infection patterns in Japanese macaques.

Keywords: *Macaca fuscata*, parasite transmission, seasonality, monkey parks, wildlife tourism

Demographic History of the Endangered Malayan Tapir: A Whole-Genome Resequencing Approach

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The Malayan tapir (*Tapirus indicus* Desmarest) is an endangered species in Southeast Asia. Captive breeding programs are important conservation maneuvers. To better understand the demographic history of the Malayan tapir in order to inform better conservation efforts for the species' conservation, this study use a whole-genome approach to investigate its effective population size before present. We used 38 microsatellite markers and the mitochondrial DNA control region to select six samples among 31 Malayan tapirs from the Japanese zoos. We re-sequenced the whole genomes of the six tapirs, putatively from Peninsular Malaysia, Thailand, and Sumatra to infer its demographic history based on pairwise sequentially Markovian coalescent (PSMC). We also created pseudo-diploid X chromosomes for each population pair for PSMC analysis to investigate divergence time between populations. We found two peaks in the projections of effective population size, which might signify population expansion or migration events during Early and Late Pleistocene periods. We observed an overall decrease in its effective population sizes over time, that went down to < 10,000 since Middle Pleistocene and < 5000-6000 before entering Holocene. PSMC on pseudo-diploid X chromosomes showed that the tapir populations might have started diverging around 5-9 kya, corresponding to the rise of sea water level post last glacial maxima that leads to the submergence of the landmass of Sundaland, including the land-bridges between the Malay Peninsula and the Sumatran Island. The projections of the effective population size were similar to that of the Sumatran rhinoceros, showing that both species might have a similar evolutionary history in Southeast Asia. We recommend future investigation into mutational load and functional genes associating with environmental adaptations in these diverging populations to inform future captive breeding programs to minimize the effects of either or both inbreeding and outbreeding depressions.

Age but Not MHC Similarity Predict Reproductive Success of Captive Japanese Golden Eagles

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Captive breeding programs are increasingly being implemented as conservation strategies of endangered populations. The success of such programs depends on individuals' reproductive success and maintenance of genetic diversity in ex-situ populations. The Japanese golden eagle (*Aquila chrysaetos japonica*) is a large apex predator listed as endangered by the Japanese Ministry of the Environment, and has been subject to captive breeding efforts since the 1990s. Though the captive population has grown over the decades (~50 individuals in zoos around Japan), a modelling study has shown that it will become inbred and go extinct within the next 200 years without the regular addition of wild individuals to the gene pool. This is largely because breeding success remains mainly limited to three pairs, but the reason for this is unknown. One possibility is the genetic compatibility of pairs–– captive golden eagles are artificially paired based on studbook relatedness, i.e., they do not freely choose their mates themselves. Generally, it is thought that individuals will choose a genetically dissimilar mate to avoid deleterious effects of inbreeding. In particular, genes of the major histocompatibility complex (MHC), which influence immunocompetence in many vertebrate species, are said to play an important role in mate choice. In this study, we tested whether MHC similarity, MHC diversity, studbook relatedness, and age of captive pairs of Japanese golden eagles influence reproductive success. Using amplicon sequencing, 20 individuals (10 pairs) were analyzed at the highly polymorphic MHC class II *DRB* exon 2, which encodes for the peptide binding region (PBR) of the antigen presenting MHC class II protein. For each breeding pair, the number of *DRB* alleles of males and females, proportion of shared alleles, the predicted mean amino acid distances at the PBR and non-PBR were calculated. The number of eggs laid, proportion of fertilized, non-fertilized, and hatched eggs, and number of successful adoptions were extracted from the past 25 years' reproduction data provided by zoos across Japan. A generalized linear mixed model approach was used to analyze the relationship between the MHC gene variables and reproductive data. Individual, pair, and zoo IDs were used as random effects, and each model was tested for overdispersion. False-discovery tests were used control for multiple testing. Although the results did not show any associations between MHC variables and reproductive success, there was a weak negative association between age and proportion of eggs hatching ($p < 0.057$). Declining fertility with age may have contributed to this decreased hatching success – golden eagles live long in captivity (up to 47 years), and they therefore may be paired for longer than they would usually reproduce in the wild. In the future, we plan to compare the captive data with baseline age information and genetic similarities of wild golden eagle pairs to further discuss these results. In doing so, we hope to apply this information towards both in-situ and ex-situ conservation of the Japanese golden eagle.

Evolutionary Patterns of the Facial Part of the Cranium in Strepsirrhines: Combining Geometric Morphometrics and Phylogenetic Comparative Methods

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Strepsirrhines are one of the two major groups of living primates, including the lemurs of Madagascar (Lemuriformes) and the lorises and galagos of the African and Asian continents (Lorisiformes). Compared to other primates, i.e., monkeys, apes and humans, they show primitive craniofacial features similar to common mammals, but they also show derived (specialized) features that are adapted to the environment they live in, making them diverse. In particular, since Madagascar had been separated from the African mainland for more than 100 million years and has tremendous regional diversity flora and fauna, it is thought that the ancestors of lemurs that arrived in Madagascar achieved enormous adaptive radiation. By contrast, traditionally, another radiation represented by lorisiformes (galagos and lorises) in African and Asian mainland is considered to be limited (e.g., less variation in body size, fewer species, and more ecological similarities). In this study, we focused on the facial cranium, which is thought to reflect any ecological habits, to quantitatively compare the differences in adaptive radiation between Madagascar lemuriformes and Asian and African lorisiformes in order to test the traditional view that Madagascar lemurs are more morphologically diverse as a result of their more enormous adaptive radiation.

We used geometric morphometrics to quantify the morphology of the cranium. By obtaining the coordinates of specific parts of the cranium, we quantified the morphology of the cranium as a set of coordinate data. Principal Component Analysis (PCA) was then used to capture key aspects of cranial diversity and to show main interspecific differences in a two-dimensional plot. By comparing the morphological variance of the lemuriformes (Madagascar lemurs) with that of the lorisiformes (galagos and lorises), we statistically determined which group had more morphological diversity. Finally, by performing a phylogenetic ridge regression combining phylogenetic information and morphological data (in this case, coordinate data), we estimated the evolutionary rate and morphology of the ancestral species to discuss when and how adaptive radiation occurred and whether there were differences between lemuriformes and lorisiformes in evolutionary patterns.

Contrary to expectations, morphological diversity of the facial cranium was higher in lorisiformes. Moreover, no rapid increase in evolutionary rate occurred in the last common ancestor of lemuriformes, and the hypothesis that their last common ancestor rapidly filled various niches by diversifying morphologically after reaching Madagascar is not supported, at least for the facial cranium. The last common ancestor of the Asian and African lorisiformes also did not show a rapid increase in evolutionary rate of facial morphology. These findings cause a stir in the traditional view of the evolutionary diversification of the strepsirrhines and indicate that a reassessment of their macroevolutionary pattern is needed.

Effect of Light Bias on Male Mating Signal and Female Mate Choice in a Sexually Dimorphic Amazonian Fish, the Sailfin Tetra (*Crenuchus spilurus*)

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The sensory drive hypothesis predicts that male's visual mating signals that match prevalent ambient lighting characteristics are more conspicuous and effective in triggering female's sensitivity. However, strong bias in lighting transmission can also decrease the variation in the transmission signals, affecting signal reliability of signals used in female mate choice. Amazon water types have striking differences in prevalent lighting: black waters are strongly red-biased while clear waters show no apparent colour bias. The sailfin tetra *Crenuchus spilurus* is a sexually dimorphic Amazonian fish species whose males have hyperallometric dorsal and anal fins conspicuously ornamented with red and yellow markings. The species is composed by two main lineages, which inhabit black waters and clear waters, respectively. Here, we evaluated the evolution of male ornaments coloration and female mate choice under clear and black water colours. Additionally, some populations of the Clear water lineage, CJ1 population, undergone secondary contact with black water environments, providing an opportunity to investigate convergent evolution in male ornaments coloration and female mate choice to black water environments. Comparison of the red coloration index of the male ornaments among lineages under not-biased and red-biased lighting conditions indicates that red bias increases the perceived intensity of red coloration but decreased the among-individual variation in colour spectra. In the mate choice experiments, females of all these lineages preferred males with larger-sized ornaments. Females of the Clear water lineage were more likely to accept males under red-biased (black water) lighting (which increases the apparent red coloration), suggesting the importance of the red coloration in their mate choice. On the other hand, male acceptance by females of the Black water lineage and CJ1 population did not change by light condition, suggesting that the signals other than the red coloration (e.g. size of ornaments) were more important in their mate choice. Our results suggest that females of Black water lineage and CJ1 population shifted the mating signal as a result of convergent evolution in black water lighting condition, though male ornament coloration of CJ1 populations was still similar to those of Clear water lineage. The lower among-individual variation in male's colourful mating signal posed by the black-water lighting condition might reduce its reliability as signals for mate assessment and caused the sift of mate-choice signals.

Effect of a Novel Skill in Social Rank of Chimpanzees (*Pan troglodytes*) and Bonobos (*Pan paniscus*)

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In human society, although dominance and prestige are both considered viable strategies for attaining social rank, prestige is yet considered unique to humans. By sharing novel knowledge that are desirable by the group members in exchange of their deference, humans earn social rank through prestige. In contrast, nonhuman animals are often considered to rely on dominance-oriented strategies as a means of gaining social rank that individuals exert their power to and coerce their subordinates to become high-ranked. However, recent studies have documented behaviors that resemble human prestige, discovering an increase in social centrality of performers after sharing their novel skills with group members. These results raise the possibility of the efficacy of prestige in nonhuman animals, but whether prestige can also aid in acquirement of social rank remains in mystery. Here we propose a plan to investigate chimpanzees and bonobos if changes in both their dominance rank and social centrality occur after sharing a novel skill. Specifically, we will first choose a few individuals to seed a skill of operating a novel apparatus that produces a food reward once solved. These individuals will share this skill with other naïve conspecifics allowing them to observe and learn the skill. Then, we will compare the change in the demonstrators' dominance rank and social centrality subsequently. Chimpanzees and bonobos are known to exhibit strikingly different social organizations. Chimpanzees mainly utilize aggressions to ascend through the steep hierarchies and conquer the top rank to monopolize limited resources. On the other hand, bonobos regulate through female alliances while showing high tolerance and prosociality, such as food sharing, towards lower-ranking or outgroup conspecifics. Therefore, we predict an increase in centrality in both species, but more prominent change in bonobos as their society is characterized by features of prestige and thus would be well recognized. However, no change is expected in dominance rank in both species. From this comparative approach, we hope to reveal how prestige has emerged and decoupled from dominance in humans.

Keywords: Social rank, prestige, dominance, centrality in social network, *Pan troglodytes*, *Pan paniscus*

Do Horses Evaluate Humans Based on Skillfulness?

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The ability to recognize and evaluate others is considered to be an important ability in selecting cooperative partners. There have been some studies on this ability in both humans and nonhuman animals, however, little empirical research has been conducted to examine what human characteristics or skills are recognized by horses. Here, we investigated whether horses are able to evaluate the skill of humans in a situation where food is likely to be available. We showed horses a pair of two experimenters, one skilled and the other unskilled, trying to open the lid of a container either with or without food in it. Looking behaviors toward each experimenter were recorded and used as a proxy measurement of the attention of the horses. Horses watched the skilled experimenter longer than the unskilled experimenter in the food condition. They also looked at the skilled experimenter for longer in the food condition compared to the no food condition. These results suggest that horses can recognize skillfulness of humans.

Comparative Study on the Social Behavior of Sambar Deer (*Rusa unicolor*) in Three Selected Captive Facilities in Peninsular Malaysia

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Sambar deer was up-listed from Least Concern to Vulnerable by the IUCN Red list in 2015. The local government has initiated Ex-situ conservation efforts to boost sambar deer numbers in captivity and reintroduce them into the wild. The reproductive success of sambar deer and their welfare management practices in captivity are important components for effective captive breeding programs. However, there has been a lack of study on sambar deer in recent years, especially about their behavior in captivity. This study aimed to identify environmental factors that may influence the behavior of the captive sambar. Three captive sites were selected and observed for an average of 40 days at each site (minimum 37 days to maximum 43 days, 6 hours/day). A Generalized Linear Model was used to determine the correlation between social behavior and extrinsic parameters. 'Captive sites' showed the strongest correlation in behavioral variability environmental settings, such as the size of the enclosure, could force the deer to spend more time in a herd, which increases the frequency in grooming, which was recorded to be highest in Zoo Negara compared to other captive sites. Time of day also significantly influenced certain behavior skewed towards morning slots. It could be due to an adaptive behavior to the feeding time in the captive sites being often in the morning, which caused the deer to rest towards the afternoon. A suggestion would be to create a more erratic feeding schedule to ensure that the deer adapt to behavior variations. An extensive study needs to be done on sambar deer to pinpoint the specifics and better understand these possible influential factors in their behavior.

Keywords: Captivity, ex-situ conservation, generalized linear model, sambar deer, social behavior

Evaluating Stressors in Captive Tsushima Leopard Cats (*Prionailurus bengalensis euptilurus*) Based on Stress Hormone Concentration and Behavioral Changes

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The Tsushima leopard cat is an endangered wild cat species that lives only in Tsushima, Japan. Because of its rarity, it is the only mammal in Japan for which a technique of reinforcement has being developed. However, the success rate of reinforcement reported in mammals as a whole is very low, and one of the possible causes is stress. However, not all stress has a negative impact on animals and maintaining a certain degree of stress is also thought to be important for reinforcement. Therefore, I will evaluate stress in individuals from both physiological and behavioral aspects in order to clarify what events are stressors in the reinforcement process.

Specifically, I have been monitoring the dynamics of stress hormone levels and observing the behavior of individuals. Stress hormones have been monitored continuously by collecting feces from individuals in the reinforcement process and measuring the cortisol concentration in the feces using the enzyme-immunoassay method. I have also been collecting feces from individuals transfer between zoos and measuring stress hormones before and after transfer facilities to investigate the degree of stress caused by transfer and environmental changes for individuals. The results of the survey showed that some individuals had high cortisol levels for a few weeks after the transfer of the facility, and then their baselines decreased. Behavioral observations have been conducted using video data. Since some types of facilities don't allow us to observe the behavior of individuals in some places, I evaluate the activity level of individuals based on the accelerometers attached to them. Behavioral observations showed that the captive environment was more similar to the wild, that is a large case, the more time differences in the daily activity of individuals were observed, indicating nocturnal behavioral characteristics. In future studies, I would like to analyze the combination of the changes in behavioral observation and hormone concentrations to clarify stressors.

Keywords: reinforcement, leopard cat, cortisol level, accelerometer

Application of Methylation-Sensitive High-Resolution Melting (MS-HRM) to Estimate Epigenetic Age in Captive Asian Elephants (*Elephas maximus*)

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Asian elephants (*Elephas maximus*) are classified as endangered by the International Union for Conservation of Nature (IUCN) due to anthropogenic effects. This has led to a decline in population with only approximately 50,000 individuals left. Knowing an animal's age is an important ecological tool in wildlife conservation. This is such as providing information on demographic trends and population viability in the wild and allowing appropriate care in captivity. However, current methods to estimate age are not always accurate and difficult to observe in the wild for many species. This is also applicable for Asian elephants where age is estimated through observing their physical features, mostly from culled individuals and not living. Visual estimation in long-lived species like the elephants is also prone to gross overestimation and/or underestimation. As they mature slowly, the visual determination of 'maturity is subjective and difficult. Thus, a reliable method to estimate age is required.

This study used a candidate gene approach, to find age-related markers related to epigenetic changes in Asian elephants that will allow us to estimate age accurately, ultimately to apply for wild individuals. We have extracted DNA from blood samples *(n* = 38) of known-aged captive individuals (*n* = 24, 21 females and 3 males), with age ranging from 0.25 – 65 years old. We referred to candidate marker genes using information on age-associated DNA methylation from previously successfully studied species, to design primers for Asian elephants. To do this, different programs such as NCBI via BLASTN was used to compare the CpG sites of Asian elephant genome and candidate marker genes. As a results, we have re-designed *RALYL* (RALY RNA binding protein-like) gene as a primer. Through finding the optimum RT-PCR conditions, amplification success was seen, suggesting that it could be a potential age-related marker for Asian elephants. Using methylation-sensitive high-resolution melting (MS-HRM), methylation rates were measured. *RALYL* in Asian elephants were significantly correlated with age ($cor = 0.71$, $p < 0.001$). When creating the age-estimation model for Asian elephants between predicted age and chronological age the mean absolute deviation (MAD) was 13.23 years. To increase accuracy for the age-estimation model, we are designing more potential age-related markers and increase the number of samples (blood and faecal samples). Successfully identified epigenetic clocks could be used for future uses in conservation efforts where accurate estimates of age are needed to predict demographic trends.

Activity Budget of Semi-Captive Drill Monkeys at Drill Ranch, Cross River State, Nigeria

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Drill monkeys (*Mandrillus leucophaeus*) are known to be of utmost conservation priority. Hunting and habitat loss are believed to be the major causes of the species decline in the wild.

It is for this reason that we report a two hundred and forty hours observation of the behavior of semi captive drills at Drill Ranch, Cross River State, Nigeria in line with its reintroduction plan. Data was collected from December, 1st 2020 to January, 2nd 2021. Scan and focal sampling was carried out in seven mixed drill groups. Continuous sampling started from 8:30 am and ended at 4:30 pm, where the following behavioral categories were recorded: Foraging, movement, resting, socialization, grooming, play, aggression and vocalization. Data analysis comprised of descriptive and inferential statistics. The time budget was recorded as follows; 10.0% resting, 46.0% foraging, 24.0% movement, 4.0% grooming, 10.0% play, 4.0% aggression and 2.0% vocalization. There exist a significant difference between behaviors and age/sex classes, ($P<0.05$). Adult males spent more time resting than any age-sex class ($X2=478.4$, $df=6$, $P<0.05$). Female adults spent most of their time foraging than any age-sex class ($X2=234.5$, $df=6$, $P<0.05$). Female adults also dominated in grooming than any age/sex class ($X2=234.5$, $df=6$, $P<0.05$). Adults males exhibited more aggressive behavior than any other age-sex class ($X2=428.4$, $df=6$, $P<0.05$), There was a significant difference for resting between adult males and juveniles ($X2=173.4$ $df=6$ $P<0.05$), there was also significant difference for resting between female adults and juveniles ($X2=96.3$ $df=6$ $P<0.05$). Also, there was significant difference for resting between female and male adults ($X2=162.5$ $df=6$ $P<0.05$). The study revealed a more aggressive interaction between the adult males, less aggressive interaction between adult females, sub-adult males and sub-adult females, and the juveniles. Results also revealed that the fights and scream vocalizations were the dominant calls heard during the study period; these were often heard during feeding and play times. The adult male 2PG call was the least vocalization heard during the study period. All of these inferred that, the semi-captive drill population at drill ranch has not loss its natural behaviors.

Key words: Drill monkey, semi captive, behavior, foraging, vocalization and socialization.

Generalization of Video-Referent Correspondence in Chimpanzees

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Humans extract information about a massive list of currently absent entities from language. The ability to generalize relational rules of correspondence between reference and its referent, is featured as a driving force of the expanding referentiality. Although referential correspondences exist in nonhuman animals, the extent to which they can be readily generalized to new exemplars is still not clear. We will use a transfer test to address this issue in chimpanzees, who have previously demonstrated successful utilization of video stimuli as a reference. They will see novel choice pairs in video for the first time, in a familiar test context. If chimpanzees can pass the test from the very beginning, we can safely conclude that they can grasp and apply relational rules of the abstract structure between video and its referent. Otherwise, their generalization of such referential correspondence may be inflexibly tied to concrete object properties of particular exemplars. In this presentation, we will report the preliminary results.

Social Associations of Mothers and Offsprings in Free-Roaming Horse Groups

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Mothers of group-living animals have influences on their offspring's various social traits ranging from dominance rank to social associate choice, as the first social partner and social role model for their offspring. Female horses in a polygynous harem group are known to form and maintain long-term social bonds with other female members; the bonded horses show affiliative interactions and stay close to each other. Previous studies have reported that foals tend to show affiliative behaviors toward the human who contacted the mother horses friendly, and that foals preferentially associate with another foal whose mother is the most preferred partner of their mothers. The maternal effect on the way foals build social relationships, however, has been little studied in feral horse groups living in a natural social environment with less human interference. In free-roaming Misaki horse harems with foals and yearlings, the affiliative/agonistic social behaviors and proximity between horses were recorded to examine whether and how the mother's social life affects the offspring's sociability and the dynamics of social relationships. The data was collected during the summer-early autumn and late autumn of 2021, using a video camera and a drone. In this presentation, the association patterns of mother-offspring pairs and their social tendencies will be presented.

How Can We Get Information from Old Fecal Samples of Iriomote Cats?: Methods for Species Identification, Sex Determination, and Hormone Measurements

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Although studies on the behavior of the Iriomote cat (*Prionailurus bengalensis iriomotensis*) have been conducted, such as studies using radio-trackings, camera traps, and diet analysis, there have not yet been reproductive physiological studies. Therefore, the purpose of this study is to clarify their reproductive trait physiologically and to contribute to their conservation. For this purpose, fecal steroid hormones concentrations have been measured to understand their estrus, pregnancy, and stress state, which is still unknown.

Firstly, we focused on the feces of the Iriomote cats collected in the conservational program by Okinawa National Forest District Office for the past three years. To confirm whether hormones were preserved in old feces, we compared fecal cortisol concentrations, which is one kind of stress hormone, between feces kept in ethanol and dried out. The results showed that more cortisol was preserved in the ethanol-preserved feces than in the dried feces. By measuring cortisol from feces, chronic stress can be revealed. By comparing the differences in stress status seasonally and by environment, we can reveal the suitable environment for their habitat in terms of physiology. Regarding the comparison between environments, we plan to classify the environments around the collection route into some categories and produce differences in cortisol levels between environments. As the next step, we'll measure fecal other steroid hormone concentrations, and compare their physiological state between seasonal differences. The hormones to be analyzed are estrogen and progesterone, the sex steroid hormone, in addition to cortisol. Sex steroid hormones are measured only in females, so we need to conduct species identification and sex determination on the fecal sample. Fecal DNA was extracted using a lysis buffer. The results for sex differences were unclear although species differences were clear. Further improvement of the measurement conditions is required.

Keywords: *Prionailurus bengalensis iriomotensis*, feces, sex determination, species identification, hormone analysis

Contagious Urination among Captive Chimpanzees: Investigation of Social Centrality of Initiator and Follower

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Contagious behavior has gained increased scientific attention as it is a fundamental component of maintaining group cohesion. Our study focused on the possibility of urination synchronization as a contagious behavior of chimpanzees and aimed to clarify initiator and follower dynamics when urinations were synchronized within 60 sec. Direct observation of four groups of five captive chimpanzees each (N=20) at Kyoto University Kumamoto Sanctuary was conducted for 421 cumulative hours. We recorded urinations, grooming interactions and inter-individual proximity, as well as frequencies of aggression, pant-hoots, and dominance displays. Generalized linear models were then applied to investigate the effects of social factors on the probability of initiating/following synchronized urinations. Network eigenvector centrality and weighted-degree centrality were calculated using simple ratio indices of grooming, play, and proximity. Individuals with higher eigenvector centrality of grooming and proximity as well as weighted-degree centrality of proximity were more likely to be followers, although no significant effects of social factors were found for initiation. Our results provided novel insights into the contagious behavior of chimpanzees which may also be developed among different species of primates including humans.

Are Semi-Arboreal Felids at Greater Risk from Habitat Destruction than Terrestrial Felids? The Ecology of the Marbled Cat in Southeast Asia

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Southeast Asia supports the greatest diversity of felids globally, but this felid diversity is likely to be threatened by the severe forest loss and degradation occurring in the region. The response of different felid species to disturbances is expected to differ depending on their ecology. For example, the largely terrestrial leopard cat (*Prionailurus bengalensis*) has been shown to thrive in disturbed habitats such as oil palm plantations where it hunts rodents on the ground. However, our understanding of how the sympatric and similarly sized marbled cat (*Pardofelis marmorata*) responds to disturbed habitats remains poor. We hypothesized that because marbled cats are more arboreal than leopard cats, and rely on tree connectivity for hunting, they are unable to adapt to relatively open oil palm plantations and other degraded landscapes. To investigate the marbled cat's habitat associations in Southeast Asia, we collated a large dataset of marbled cat occurrence records from both new and previously published camera-trapping studies. We found that marbled cats were positively associated with large, intact forests, and high forest cover and, in contrast to leopard cats, negatively associated with oil palm plantations. These findings suggest that in the face of ongoing conversion of tropical forests to oil palm plantations the marbled cat's IUCN Red List conservation status should be upgraded from Near Threatened to Vulnerable. We posit that semi-arboreal felids could be more greatly threatened by habitat degradation than their terrestrial relatives and that our findings may be generalizable to other felid species.

Do Pit Vipers Assess Their Venom? Defensive Tactics of *Deinagkistrodon acutus* Shift with Changed Venom Reserve

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To maximize survival probability, animals must assess predation risks and adopt flexible defensive strategies based on specific conditions. Pit vipers utilize venom for predation and self-defense, and venom status significantly influences its effectiveness. Thus, pit vipers may evaluate their venom reserve and adopt corresponding defensive tactics. Twenty-three sharp-snouted pit vipers (*Deinagkistrodon acutus*) were grouped by different venom status and were subjected to eight behavior trials. Subjects' defensive behaviors were recorded and analyzed. Results showed that the normal venom group displayed stable responses across the trials. The low venom group showed fewer strikes and more fleeing behaviors at the end of experiments. After given prolonged intervals for replenishing the venom, significant increases of strike behaviors were observed in the replenishing venom group. These results demonstrated the capability of adopting flexible defensive tactics based on varied venom reserve and provided new evidence for venom-status-recognition in venomous snakes.

Keywords: Crotalinae, Defensive strategy, *Deinagkistrodon acutus*, Venom metering

The Social Networks and Hierarchy of a Captive Group of Orphaned Infant Chimpanzees

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Wild chimpanzees live in complex fission-fusion networks with strict dominance hierarchies that can be disrupted under captive conditions. Social network analysis (SNA) and dominance hierarchy analysis can help provide important information about a captive chimpanzee group's social structure for decision making about future group compositions and to ensure the group's welfare. Additionally, information about infant chimpanzee social behavior under captive conditions has received little attention to date. This study analysed the social networks for affiliative and agonistic behavior as well as for proximity of a group of orphaned infant western chimpanzees (*Pan troglodytes verus*) at Tacugama Chimpanzee Sanctuary in Sierra Leone. It investigated the question if age or sex of an infant influences its position in the social networks or dominance hierarchy of the group. Data collection was conducted for 9-weeks at Tacugama Chimpanzee Sanctuary using focal-animal sampling and instantaneous scan sampling. Social networks were constructed, community detection was carried out and using centrality network metrics (degree centrality, betweenness centrality, closeness centrality and clustering coefficient) an individual's position in the network was determined. Additionally, randomized Elo-rating was used to analyse the group's dominance hierarchy. It was determined that certain individuals were crucial for connecting subgroups in the affiliative network of the orphaned infant chimpanzee group. Furthermore, age seemed to play a role in the affiliative network but not in the other networks, likely because of the dependence of individuals to each other. The affiliative and proximity network did not give the same results, probably because the individuals could not exercise their spatial preferences. Sex did not seem to influence an infant's position in the networks which contradicts previous studies. Lastly, the dominance hierarchy was not influenced by age or sex, but older individuals seemed to be higher in the hierarchy, probably because of increased body size. Tacugama Chimpanzee Sanctuary should ensure that the individuals connecting the subgroups of the group should remain in the group and should provide more resting places for the infants. Additionally, further studies about infant chimpanzee social networks and dominance hierarchy are needed.

Studying the Effect of a Raccoon Dog Invasion of Yakushima on the Ecology of Disease, from the Ground Up (to the Sky): a Pilot Study

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Infectious disease is a global threat to human, animal, and ecosystem health. Conflicting ideas about whether biodiversity is positively or negatively associated with disease risk (and vice versa) have urged several authors to postulate that specific *changes* in ecological communities might matter more in shaping the biodiversity-disease relationship. This is crucial in view of current trends in biodiversity loss, but also in contexts like biological invasions, where the introduction of a species to an ecosystem adds another layer of complexity over simple increases in biodiversity. Raccoon dogs (*Nyctereutes procyonoides*) - also known as tanuki (タヌキ or 狸) - are iconic in Japan but are also among the most successful invasive carnivores in Europe. Raccoon dogs also invaded some islands in Japan, including Yakushima – a UNESCO World Heritage site. However, their effects as potential predators, competitors, and disease carriers in Japan are unknown. In this poster, I present several methods our team implemented during a recent pilot study to ultimately: (1) map the distribution of raccoon dogs and other mammal communities in Yakushima; (2) establish the social structure of raccoon dogs in the Western coastal forest (Seiburindo) and consequent opportunities for intraspecific disease transmission; and, (3) identify what pathogens or parasites raccoon dogs can carry in the system. During this pilot study, we successfully performed latrine surveys and located 24 latrines in the Kawahara and Hanyama areas of the Seiburindo, and were able to collect ~50 fresh fecal samples. We were able to collect adult nematodes passed during one defecation event. We also performed multiple test flights with a drone capable of thermal-imaging to evaluate its ability to rapidly locate and possibly identify endotherms in the area. Ongoing laboratory analyses are being conducted to identify different pathogens or parasites that are capable of infecting other species on Yakushima, including humans, such as the tapeworm *Echinococcus* spp., the skin mite *Sarcoptes scabiei*, and the bacteria *Campylobacter* spp. and *Leptospira* spp., among others.

Asymmetry of Early Social Interactions between Infants and Non-Mother Individuals in Yakushima Japanese Macaque (*Macaca fuscata yakui*)

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Infants in the early developmental phase show interest in the social environment surrounding them; Adults and juveniles watch for an opportunity to handle the infants while staying near their mothers. These statements look natural and are revealed by previous studies, but they are not enough to explain the early social interactions of Yakushima Japanese macaque (*Macaca fuscata yakui*). This study shows that infants tend to be not handled by macaques who they approach or (try to) contact and, rather, to be avoided or left by them within two months after their birth. This asymmetry is clearly found in interactions with relatives in juveniles. Adult relatives (e.g. sibling, aunt, grandmother) tend to handle their related infants less than non-related females do, even though they are gazing at the infants approaching them. (Related/unrelated) Juveniles tend to avoid or flee from the infants. 4-year-old sisters only rapidly become handlers of their youngest siblings. The asymmetry of early social interactions of macaques will be meaningful when we consider how different they take a role in infant growth and how fast they overcome unfamiliarity towards newborn infants.

Tracking Visual Attention of Crows Using a Motion Capture System

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Recent technological advances made it possible to record postural changes of moving animals in behavior/cognition research. Although several technologies are available for this purpose, one underexplored possibility is the use of an infrared motion-capture system, which excels at tracking subtle and rapid 3D movements like saccadic head movements of birds. For most of the bird species, head movement is a good proxy for the direction of attention. In this study, we aimed to establish a method to track the direction of visual attention in freely-moving large-billed crows (*Corvus macrorhynchos*) using a motion-capture system. To estimate the direction of visual attention, we examined how crows looked at the object which were presented in various locations using a motion capture system. We also examined the visual configuration of crows using an ophthalmoscopic reflex technique. The results showed that crows oriented specific head angles to the objects, but not same as the angle of optic axis (i.e., line passing through the center of the cornea and lens). The maximum eye-movement amplitude was over 30 degrees, which suggested that eye-movement were an important factor on the direction of attention. These results suggested that by considering the position of the object in relation to the crow and their eye movement, we will be able to estimate the direction of visual attention in crows. We thus show that a motion capture system has a good potential to examine the direction of visual attention in freely-moving birds.

Comparative Transcriptome Analysis in Stranded Cetaceans Provide Novel Insights into the Evolution of Lipid Utilization

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Extant cetaceans (whales, dolphins and porpoises) acquired specialized high-frequency hearing used in the fully aquatic environment, and conversely, quite degenerated their olfactory and gustatory capabilities. 'Melon' in the forehead, 'intramandibular fat body' in the lower jaw bone, and 'extramandibular fat body' on the external surface of the lower jaw bone are developed as novel morphological traits in toothed whales, called acoustic fat bodies. These acoustic fat bodies are used for echolocation that contributes to hunting, navigation, and communication in the aquatic environments. Even though how novel traits arise in organisms has long been a major question in biology, few studies on the acoustic fat bodies at the molecular level has been reported.

To investigate the molecular basis of the evolution of acoustic fat bodies, we comprehensively examined mRNA expression of the multiple tissues of cetaceans. Fresh acoustic fat bodies as well as skin, muscles, and connective tissues were opportunistically collected from stranded cetaceans in Hokkaido. Finally, total RNA (transcriptome) was extracted from three toothed whales (pacific white-sided dolphin, harbour porpoise, and Stejneger's beaked whale) and a baleen whale (minke whale). Massively parallel sequencing of mRNA (RNA-seq) was performed using Illumina NovaSeq6000 and the expression level of each gene was investigated. Our results suggest that the acoustic fat bodies are originated from mixed tissue of muscle and subcutaneous fat (blubber). Interestingly, we found significant expression of olfactory receptor genes in melon. These findings provide novel insights into the evolution of lipid utilization and acoustic ecology in cetaceans.

Transcriptome Changes of Maternal Immune System Related to Embryonic Diapause in Brown Bears

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Embryonic diapause defines the dormancy of the embryo before implantation. It occurs in over 130 mammals. It allows them to control the timing of their mating and parturition. Ursidae is one of the few families that all species occur embryonic diapause. Among them, brown bears (*Ursus arctos*) basically mate in late spring to early summer. Their embryo implants in early winter and gestation occur approximately 60 days after implantation. Embryonic diapause allows them to give birth in a safe den during hibernation. It also let only mothers who can get weight enough in autumn give birth.

In captivity and wildlife monitoring, it is difficult to judge their pregnancy. Brown bears spent over half of the gestation period as embryonic diapause. Furthermore, non-pregnant bears are often shown pseudopregnancy and their hormonal dynamics are similar to pregnant bears. Hence, hormone levels do not always reflect their pregnancy.

In mammals, the maternal immune system changes dramatically to accept the embryo. Therefore, the gene expression related to the maternal immune system may change with the phase of gestation. The purpose of this study was 1) to identify embryonic diapause regulation mechanisms of the immune system at the molecular level, and 2) to establish gene-markers for a pregnancy test that can collect easily and assess early stage. In this study, a total of 28 blood samples were collected from mated or not mated captive female brown bears at the Noboribetsu Bear Park, Hokkaido, Japan in 2021. Among them, RNA sequencing were performed in 12 samples.

To identify the correlation of each sample, principal component analysis was performed. As a result, there was no correlation between estrus and embryonic diapause period in mated-parturition bears samples, while not mated bears sample correlated between estrus and embryonic diapause period. Therefore, pregnant bears' blood transcriptome seems to change in embryonic diapause.

To identify differentially expressed immune-related genes, the expression of each transcriptome was compared between estrus and embryonic diapause. As a result, a total of 408 transcriptomes were identified as differentially expressed. Among them, 47 were upregulated and 361 were downregulated in embryonic diapause. That analysis also revealed that some immune-related genes were downregulated in embryonic diapause. Therefore, the maternal body is likely to recognize the existence of the embryo, and the immune system was suppressed to avoid rejecting the embryo.

To estimate the regulation of embryonic diapause related to the immune system, it is necessary to perform enrichment analysis of identified DEGs. In addition, to establish gene-markers for an early pregnancy test, determination of the identified downregulated immune-related genes expression quantities is required.

Possibility of Gustation Being Used during Breastfeeding by Cetacean Infants

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Breast milk is the only available foraging resource in the early stages of mammalian development. The ability of infants to recognize breast milk is considered essential for survival. In this study, we hypothesized that cetaceans, which have lost their sense of taste and smell during evolution, use their few remaining chemosensory senses for breast milk recognition, which is essential for mammals. We focused on taste of fat, which is the most abundant nutrient in cetacean breast milk.

Three studies were conducted to test this hypothesis in two species of the genus *Tursiops* (*T. aduncus*, Indo-Pacific bottlenose dolphin and *T. truncates*, Atlantic bottlenose dolphin). We performed comprehensive gene expression analysis (RNA-seq) in a tongue of *T. aduncus* infant that accidentally died on August 1, 2020, on Mikura Island. Seventeen regions were collected from three parts (marginal papillae, V-shaped groove, and control) of the tongue of infants. Based on the RNA-seq, expression levels of fatty acid receptor genes were examined. Expression of fatty acid receptor genes was found in all three compartments, with the highest expression in the limbic papillae, a structure that is particularly prominent in infancy.

This study is the first report to examine the availability of fat taste in cetaceans. The expression of fatty acid receptor genes in the tongue of *T. aduncus* infants supports the possibility that infants of the genus *Tursiops* can discriminate fatty acids. Fatty acid taste is expected to be useful for breast milk recognition and foraging learning in infancy. In future, we aim to verify this hypothesis by conducting preference tests for breast milk components such as fatty acids in reared individuals and by analyzing the function of receptors at the cellular level.

Comprehensive Analysis of the Gut Microbiome in Gum-Eating Mammals

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The symbiont intestinal microorganisms are closely linked to the host health. Conventional methods is difficult to isolate and culture the intestinal bacteria. Recent improvements in genetic analysis techniques have made it possible to comprehensively investigate the composition and function of the intestinal microorganisms. In this study, we focus on the degradation of dietary fiber, a major role of intestinal bacteria in non-human primates. *Nycticebus coucang* (Sunda slow loris) is a non-human primate in tropic Asia and an obligate eater on fiber-rich gum. We compared the gut microbiome of wild- and captive-born Sunda slow lorises of the Sapporo Maruyama Zoo ($N = 7$). These lorises feed on gum Arabic every day. For 2 weeks, we estimated the amount of diet eaten and collected their fresh feces. The variable regions (V1-V2) of bacterial 16S rRNA genes in fecal DNA were sequenced by Illumina MiSeq and analyzed by QIIME 2.

The lorises were classified into three group: wild-born (W), F1 captive-born (F1), and F2 captive born (F2). Lorises of the W group completely eat provided Arabic gum every day and the F1 group eat most, but F2 group often leave it unfinished. Gut microbiome analysis shows that species composition of the family Prevotellaceae was significantly different among the groups. This indicates that the bacteria that respond to gum Arabic may decrease during the generation changes in captivity. In terms of animal welfare, we have to consider the gut microbiome and diet preference when we feed captive-born animals on fiber rich-diet.

Training of Matching to Sample Task for Killer Whales (*Orcinus orca*)

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Cognitive abilities are products of evolution and are shaped by both phylogenetic and environmental factors. In order to examine how environmental factors have affected on the evolution of cognitive abilities, it is essential to conduct comparative studies on mammals living in underwater environment, which is far different from terrestrial environment. Cetaceans (dolphins and whales) have been highly adapted to the underwater environment and have developed their cognitive abilities according to the characteristics of the environment. While many researches have been conducted on auditory abilities of cetaceans, it is not well understood how they use their visual abilities.

In my previous research, we succeeded to develop a new experimental method using a computer and monitors to examine visual abilities of killer whales (*Orcinus orca*). By using this method, it was suggested that contrast of luminance played an important role in their visual object recognition of cetaceans with low visual acuity and no color vision. In order to conduct further examination of their visual abilities, we decided to train them to participate in match-to-sample tasks. At first, we expected them to understand the rules of matching-to-sample task by using geometric figures displayed on three computer-controlled monitors. However, we couldn't make them to understand the rules of this task which is far different from the previous two-choice task.

Recently, we have started to use the real objects as the stimuli through the underwater window. We have selected three objects, boot, fin and net from the objects familiar in their daily life. As a flow of one trial, the subjects approached the underwater window are shown a sample stimulus and touched it with their rostrum. After that, the experimenter hides the sample stimulus once behind a box, and puts out the correct and incorrect stimuli from behind the box at the same time, and if they can choose the same correct stimulus as the sample stimulus, it is rewarded with the sound of the whistle and several pieces of fish. Currently, their correct rate in this training is over 90% for all pairs of these 3 objects, so we are planning to introduce a new object as a fourth stimulus to reinforce their understanding of the rules.

Comparing the Social Relationships of Feral Horses during Breeding and Non-breeding Season Using Drones and GPS Tags

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In many group-living animals, the social relationships and group cohesiveness show seasonal changes due to different resource availability, predation pressure, and reproductive status. Feral domestic horses (*Equus caballus*) form multilevel society, where harems and bachelor group (all-male units) gather to make a higher-level social group, which is called as “herd”. The membership of harems is known to be stable over 3 - 4 years, while a herd shows unstable, fission-fusion structure and harems become less collective during non-breeding seasons (around August - March). The details of such seasonal changes in inter-group level relationships are largely unknown in feral horses and other species showing multilevel society as well. In this study, we collected their positional data combining drones and GPS tags during June – October, 2021, and compared the social network between breeding and non-breeding seasons.

Exploratory Vegetation Survey by UAV in Yakushima Island

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We used a UAV to photograph vegetation in the World Heritage area of western coastal in Yakushima Island to explore what information could be read from the areal photos. This area has not been significantly disturbed by humans since about the 1960s. However, until about 1950, there was some logging and afforestation, with some patches of single species plantations. Since the logging and plantations were mainly done in private forest and their records are not available.

In August and December 2021, we used a quadcopter to take photographs and attempted to visually identify afforestation. Cedar (*Cryptomeria japonica*) and camphor (*Cinnamomum camphora*) trees are known to have been planted. The cedar plantations were found in small scattered patches. The camphor plantation was found to have large patches in the central and southern parts.

As for natural vegetation, we were able to distinguish dead pine (*Pinus thunbergii*) to some extent. Pine wilt disease by pinewood nematodes is occurring on Yakushima Island, and it is becoming a problem as pine dieback spreads from human settlements to World Heritage areas. From the photos taken this time, pine dieback was observed along the road in the entire heritage area, suggesting that it is spreading to the central part of the World Heritage area.

Longitudinal Working Memory Study in Chimpanzees

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Previous studies on working memory in chimpanzees (*Pan troglodytes*) have revealed that child chimpanzees have a greater ability than adult chimpanzees and adult humans (*Homo sapiens*). They found similarities and differences between chimpanzees' and humans' memorizing capacities and strategies. Furthermore, their results and other human studies suggested that the ability of working memory develops during infanthood and childhood, and it declines in the period of aging. There are, however, no longitudinal studies of working memory in a single chimpanzee. Even in human studies, most studies were based on the comparison of the young and the aged. This study aimed to chart the change in chimpanzees' working memory across their lifespan. Chimpanzees in Kyoto University Primate Research Institute (KUPRI) have participated in memory studies for more than a decade. For this study, I used computer-task data from three mother-offspring pairs which was stored at KUPRI to analyze them. These computer tasks required chimpanzees to memorize the order and position of Arabic numerals scattered on a touchscreen monitor. The data was collected from 2007 to 2021 and covered the life phase of chimpanzees from child/sub-adult to adult and from adult to elderly. As a result, their performance dropped especially in 'difficult' tasks and conditions. Moreover, the results suggests that chimpanzees' working memory ability might decline in age from their twenties to thirties, but there was no data of this period. Therefore, further data collection is needed to cover a longer period and to examine an effect of difficulties in tasks.

Population Genetic Structure of the Malayan Tapirs in Peninsular Malaysia Revealed by Nine Cross-Species Microsatellite Markers: Genetic Diversity Compared to the Japanese Ex-situ Population

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The Malayan tapir (*Tapirus indicus*) is an endangered species endemic to Southeast Asia. Deforestation and habitat fragmentation, leading to increasing cases of roadkill, are among the top threats the animal faces. Studies that use maternally-inherited, mitochondrial DNA genetic markers to describe the population genetics of the Malayan tapir across their habitat exist, but thus far none have used the biparentally inherited, nuclear genetic markers. In this study, the genetic diversity, population genetic structure, and sex-biased dispersal patterns of the tapir population in Peninsular Malaysia were investigated using nine cross-species microsatellite markers. For this, 67 tapir samples (39 wild, 11 captive-born, and 17 of unknown origin) were collected from the Wildlife Genetic Resources Bank, National Zoo of Malaysia, and Sungai Dusun Wildlife Conservation Centre. We also genotyped 31 Malayan tapirs from the Japanese zoos to contrast their genetic diversity with what present in Peninsular Malaysia. Low genetic diversity of the wild tapir population in Peninsular Malaysia was found, and two to four genetic clusters were inferred. Overall, the population genetic structure of wild tapirs was unclear due to disparate distribution of the genetic clusters and uneven sampling. As the number of genetic clusters increased to four, however, population genetic structuring arose in the southern forest complexes of the Central Forest Spine as genetic clines/clusters, which might be attributed to admixture between neighbouring populations, habitat fragmentation, and a complex landscape around the boundaries of a few forest complexes. Sex-biased dispersal was not detected in the current dataset. Results from this study will have conservation implications related to the implementation of the Central Forest Spine Master Plan in Peninsular Malaysia to connect major forest complexes. Genetic diversity of the Japanese *ex-situ* population was higher than both captive and wild populations in Malaysia despite the presence of many parent-offspring pair. This was due to the presence of tapirs from Malaysia, Thailand, and Sumatra origins. Principal coordinate analysis indicated genetic differentiation between these populations. Future genetic assessment of the wild populations in Thailand and Sumatra will provide more insights to the levels of genetic diversity of tapirs in Southeast Asia.

Rest Site Selection Analysis in Japanese Macaques

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It is generally known that animals prefer rest sites for thermoregulation. Rest site selection has also been widely confirmed in primates, for example, warming up by sun-bathing on cold days and cooling off in the shade on hot days. Thus, rest site selection at the individual level may affect the cohesiveness of group-living animals. In primate socioecology, cohesiveness determines types of feeding competition and affects sociality. As far as I know, however, it is unclear whether cohesiveness at rest determines rest site competition. Therefore, I examined whether the rest site selection influences sociality in this presentation.

Molecular Evolution of Sweet Taste Receptor Genes in Elephants

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Taste perception is an important role for diet choice. Mammals having different dietary characteristics, even if they are closely related each other, would change molecular function of their taste receptors, resulting in the evolutionary adaptation to taste compound of their different diet. This has implications for efficient foraging and preference. Elephants are the largest terrestrial herbivores among the world that live in (sub)tropic Asia and Africa. They are currently classified into three species, African savanna elephant (*Loxodonta africana*), African forest elephant (*Loxodonta cyclotis*), and Asian elephant (*Elephas maximus*). African forest elephant and Asian elephant are far apart in distribution, but they have similar ecology. They live in tropical forests and like to eat fruit. On the other hand, the savannah elephant, lives in the savannah and eats less fruit than the two forest-dwelling species, possibly due to its availability. We focused on their sense of sweet taste because of this fruit-eating aspect. In this study, we examined whether differences in diet among elephant species actually affect sweet taste receptor genes through sequence analysis of *TAS1R2* and *TAS1R3*.

Genomic DNA of the three elephant species was extracted from blood samples provided by zoos, and performed PCR and Sanger sequencing for all exons of *TAS1R2* and *TAS1R3* with the designed primers. We constructed *TAS1R2* and *TAS1R3* gene phylogenies using the maximum-likelihood method and calculated dN/dS for three species of elephants in addition to mammoth, hyrax, manatee, dugong annotated from their whole genome assemblies.

The phylogenetic trees of both *TAS1R2* and *TAS1R3* were similar to those of the species trees. There was no significant difference in the likelihood of the elephant lineage, suggesting that the sweet taste receptor gene is conserved in the three species, suggesting that pure selection occurred overall. The dN/dS values were lower than 1 for both *TAS1R2* and *TAS1R3* in all branches of elephant phylogeny, suggesting that there was overall pure selection.

The conclusion is that diet does not affect sweet taste receptor genes in elephants, or that sweet taste receptor genes are highly conserved among three species of elephants. Since the functional changes of sweet taste receptors in elephants could not be obtained on a gene sequence basis, it is necessary to conduct experiments at the higher biological levels to analyze the phenotype of sweet taste receptors, such as behavioral analysis of preference to different types of sugar and cellular analysis of the sweet taste receptors.

Genetics, Behavior, and Evolution of the Visual Adaptation in Egg-Laying Mammals

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Animals have evolved their vision by adapting to their habitat. Mammals use two types of cone visual pigments, which differed in the wavelength range of the light they received for light reception in bright light color vision. Placental and marsupial mammals retained two types, SWS1 and LWS. On the other hand, monotremes (egg-laying mammals) retained SWS2 and LWS.

The common ancestor of extant monotremes (platypus and echidna) is thought to be semiaquatic. It may be the result of adaptation of vision to the light environment in water. There is only one species of semi-aquatic monotremes in the family Ornithorhynchidae (platypus), and only four terrestrial monotremes in the family Tachyglossidae (echidnas). The purpose of this study was to compare the function of the visual system between the platypus and echidna, and to examine how the visual system of monotremes has evolved adaptively.

In this study, we synthesized all nucleotides of SWS2 and LWS of both platypus and echidna, incorporated them into expression vectors, artificially expressed them in cultured cells, and measured the maximum absorption wavelength (λ_{max}). The λ_{max} of the cone visual pigments of the platypus and echidna was 444 nm and 448 nm, respectively, in SWS2, and 565 nm and 569 nm in LWS. This suggests that both SWS2 and LWS have evolved conservatively despite the large differences in the habitats of extant monotremes.

One of the major factors in visual adaptation is the activity pattern, but that of monotremes has not been quantitatively known very much. In order to quantitatively investigate their activity patterns, we analyzed the day/night behavior of two echidnas at Nagoya Higashiyama Zoo for one month. The results showed that echidnas' activity occurred 46% of the time during the light condition and 54% of the time during the dark condition. In conclusion, exposure to a variety of light through diurnal and nocturnal activity may be important in the evolution of vision in monotremes.

The Role of Intentional Communication in a Cooperative Activity in Horses (*Equus caballus*)

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Cooperative behavior requires sometimes certain degree of coordination between participants to achieve a common goal. In humans, this coordination is ensured through the establishment, maintenance and dissolution of joint commitments. The responsibility to engage and finish a previously-arranged commitment entails the display of signals by the participants to mutually agree and engage in the cooperative task. Although joint commitment has been more investigated in primates, a broader comparative approach is needed to understand the evolution of cooperative behavior. Horses are an interesting model for this, given their domestication process and their sociable skills. Allogrooming is defined by the cooperative action of two horses grooming each other on the neck and withers simultaneously. Besides playing a hygienic role and reducing heart rate, allogrooming serves to building social bonds between group members. The symmetrical pattern of allogrooming leads that if one individual wants to perform allogrooming, it is posed with the problem of how to signal the selected individual to engage in the behavior. If joint commitment is needed to engage in allogrooming, we will expect to find some signals before (and, presumably after) allogrooming behavior. Head bobs, head shakes and self-scratches have been preliminary observed in feral horses before allogrooming, although they have not been described in previous literature yet. The resemblance in form and function of the observed behaviors to allogrooming could suggest that these signals may have developed in horses to request grooming. The focus of the study is to prove the existence of referential signals during allogrooming in horses and to test whether these signals are affected by the partners' social dimensions, similarly to the human "face management".

All-You-Can-Eat: A Preliminary Study of Invasive Raccoon Dog (*Nyctereutes procyonoides*) Predation of Endemic Species and Dietary Competition on Yakushima Island, Japan

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Tanuki, or raccoon dogs, are ecological generalists and opportunistic hunters native to Eastern Europe and Asia, including mainland Japan. They have also been identified as one of the most successful alien carnivores in Central and Western Europe, and areas affected by a raccoon dog invasion reflect changes in the structure of food webs. In invaded habitats, racoon dogs have been found to compete with native small carnivores for resources, and alter populations of native small vertebrates, invertebrates, and edible plant species through predation. This preliminary study aims to address invasive raccoon dog dietary effects on Yakushima Island, a UNESCO world heritage site, through assessing: 1) the alteration of the structure of the unique biodiversity found on Yakushima through the consumption of rare and endemic species found on the island, and 2) areas of direct competition for resources through ingestion of the same prey items by tanuki and other native Yakushima carnivore species. During a pilot study in January-February 2022, tanuki latrine surveys were undertaken the Hanyama and Kawahara areas of the Seiburindo forest on western Yakushima. From the 24 latrines identified during the pilot survey, ~50 samples of fresh feces were collected for analysis. Ongoing analysis of the samples aims to detect presence of small mammal and invertebrate DNA to investigate potential dietary overlap between racoon dogs and Yakushima's endemic subspecies of Japanese weasel. Samples are also in the process of being analyzed for presence of endemic species DNA, including endemic species of crabs, insects, plants, amphibians, reptiles, birds and fish. As tanuki diets vary seasonally across their range, this study aims for a long term multi-seasonal investigation of tanuki impact on Yakushima flora and fauna through predation.

Estimating Epigenetic Age in Okinawa rail (*Hypotaenidia okinawae*) using Methylation-Sensitive High-Resolution Melting (MS-HRM)

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The Okinawa rail (*Hypotaenidia okinawae*) is a flightless bird endemic to the northern part of Okinawajima island. It is an elusive bird with a declining population due to habitat loss, non-native species and traffic accidents. Acquiring information on the age of individuals is very important because it will help us understand the dynamics and viability of populations, as well as which age groups are vulnerable towards certain threats such as traffic accidents. However, the Okinawa rail is long-lived up to 15 years in the captive and is difficult to judge its age by its appearance. In Japan, age estimation of wild birds is commonly done by attaching foot rings. However, this method requires recapture of individuals, and it does not necessarily provide accurate age data if the initial age at marking is not known. Therefore, we attempted to apply molecular methods to estimate age, focusing on DNA methylation. DNA methylation is an example of epigenetic change that occurs over an individual's lifetime, and thus has been reported to be an accurate method to estimate age in wild animals. Most previous studies have focused on mammals, so this study is one of the few, novel attempts to apply towards birds. Here, I used candidate genes to find age-estimation markers that are potentially associated with epigenetic changes in the Okinawa rail. I designed primers by referring to 7 genomic regions, whose methylation levels were reported to be correlated with age of shearwaters (*Calonectoris leucomelas*) in a previous study. Using NCBI BLAST, the sequences of the 7 regions were confirmed in the Okinawa rail genome and three primer sets were successfully designed for amplifying the *DHH* (desert hedgehog Drosophila homolog) gene, one of the aforementioned 7 regions. The reverse-transcriptase PCR (RT-PCR) conditions were optimized and Methylation-Sensitive High Resolution Melting (MS-HRM) was used to estimate the methylation rates in 99 DNA samples extracted from blood (n = 93) and muscle (n = 6) of captive Okinawa rails of known age (0-15). The results showed no significant correlations between age and DNA methylation rates in any of the tested regions of *DHH*. We suspect the targeted gene may function differently and/or may not experience the same epigenetic changes over time as in the shearwater because the two species are phylogenetically distant. Therefore, we are now preparing to survey the methylation rates across the entire Okinawa rail genome using Next Generation Sequencing (NGS) to search for combinations of age-related gene regions unique to the Okinawa rail. We also hope to expand this study to include non-invasively obtained samples such as molted feathers and feces, which will be useful for estimating age in wild individuals in the field.