

Abstracts

Darwin 200 and Opening Ceremony

Temperature as a major selection pressure on the evolution of *Drosophila* populations

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According to the neo-Darwinian theory, evolution results from genome variations and selection of certain genotypes better adapted to environmental constraints. The *Drosophila* group of species is a good model to try to characterize specific selection pressures and their effect on certain genes or groups of genes which are involved in speciation. Among sexual isolation mechanisms between species, pheromone production and discrimination are important. In a number of insects like *Drosophilidae*, contact pheromones like long chain cuticular hydrocarbons (CHC) bearing double bonds in various positions, are components of the cuticle and thus also play an important role in the interaction of the organism and the outside like the protection against desiccation. *Drosophila melanogaster* and *Drosophila simulans* are two sibling species which can be found in many ecosystems around the world. However both have appeared and differentiated in central Africa. These species then migrated from west to east and reciprocally, and from central Africa to north and/or south. During these migrations, gradual significant changes have been shown to take place in both their contact pheromones and their potential isolation with ancestor individuals. Moreover biochemical and molecular studies have shown that candidate genes coding for some of the biosynthetic enzymes were temperature sensitive, suggesting that temperature might be a major selection pressure. However, behavioral studies have shown that sexual selection also played a role. Other studies of *Drosophilidae*, especially endemic to the Hawaiian archipelago, involve multiple complex environmental changes.

Early Cretaceous biological evolution: perspectives from the Jehol Biota, China

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The Early Cretaceous represents a geological time from 145 to 100 million years before present. It records some of the most significant geological and paleoenvironmental events across the Earth such as sea floor spreading, volcanic eruptions, and the occurrence of the paleogeomagnetic Cretaceous Normal Superchron. This period is also characterized by an increase of atmospheric CO₂ and a greenhouse climate that reached a peak in the middle Cretaceous with a global annual mean temperature 3-10°C higher than today. Furthermore, paleontologists have discovered that during this geological interval occurred the first flowering plants, the greatest diversification of insects, the first radiation of birds and the first appearance of powerful bird flight, the earliest appearance of eutherian and metatherian mammals, and the appearance of feathers and arboreal adaptation in various theropod dinosaurs. The Early Cretaceous Jehol Biota from Northeast China, about 131-120 millions years old, is characteristic of exceptional preservation and a rare diversity and abundance of fossils, comprising the best evidence for understanding biological evolution at this critical geological time. In the Early Cretaceous, there was also a general body size increase among major vertebrate groups, i.e. birds, dinosaurs, pterosaurs and mammals, possibly as a response to global temperature increase. The Early Cretaceous also marks the emergence of the modern terrestrial ecosystem, providing the first example of pollinating insects, specialized seed-eating birds and pterosaurs, diverse herbivorous theropod dinosaurs and scansorial mammals, suggesting that floral evolution had played a key role in the evolution of these animals.

All climate change is local: how mechanistic models can provide an understanding of ecological responses to climate change

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While many factors are important to the fitness of intertidal invertebrates, an organism's body temperature is among the most important. In intertidal ecosystems, species range shifts of up to 50 km/decade are already occurring, largely in response to temperature shifts. Patterns of body temperature are often much more complicated than corresponding patterns of weather and climate. Importantly, organisms respond physiologically and behaviorally only to the very local characteristics of their immediate environment. Predicting where, when and with what magnitude the likelihood of changes in community structure are likely to occur requires that we understand how large-scale processes ('environmental signals') are downscaled to the level of the organism. Biophysical models (models that track heat inputs and outputs) provide a key tool for analyzing how environmental signals drive organism and ecosystem processes. Importantly, such approaches show that predictions cannot always be made through correlations with large-scale signals, such as those measured by buoy or satellite. While such measurements are a vital tool for measuring environmental parameters over large spatial and long temporal scales, they are not sufficient for predicting organism responses. While aspects of climate change such as temperature, salinity and ocean chemistry often cannot be modified on a local level, an explicit understanding of where these impacts occur can nevertheless inform management and policy. By understanding how factors such as body temperature interact with stressors such as nutrient load, fishing pressure and sedimentation, we can triage specific locations by modifying these latter stressors through effective management.

Biodiversity and climate: some key issues

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Most of the international concern about climate change focuses on mitigation and cutting the emission of greenhouse gases. Much less international attention is being given to the impacts on biodiversity of climate change, though humans are most likely to feel the impacts of climate change through the symptoms of changes to species and ecosystems. This paper will present an overview of recent findings of the ecosystem changes that are being brought about by climate change, and the direct and indirect impacts of these changes on species. Results from recent studies on birds, mammals, reptiles, plants and marine organisms will be presented. The changes in range and behavior of species is likely to have profound implications for human well-being, and may also have national security implications. Hence what may appear as merely a scientific concern rather quickly becomes a major security and political concern. It is essential that policy-makers are well informed of the implications of climate change on species and ecosystems, and this paper will suggest some of the key issues that should be covered.

Do current insect invasions fit Darwin's evolutionary theory?

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In his naturalization hypothesis, Darwin proposed that introduced plants are less likely to invade areas with congeneric native species because the introduced species would compete with the closely-related native ones, and furthermore would encounter native herbivores and pathogens that could use them as hosts. Darwin also considered as an alternative possibility the pre-adaptation hypothesis in which successful invaders are more likely to belong to the same genera of species native to the recipient community because they are more likely to contain traits that pre-adapt them to the new environment. Although these hypotheses are supposed to apply to plant species, there is no theoretical reason that they do not apply to animals. Some studies support the naturalization hypothesis,

e.g. with plants, whilst others reject it, e.g. with fishes. Using the database generated by the European project DAISIE, we tested the naturalization hypothesis on 1517 exotic species of terrestrial invertebrates established in Europe. It appeared very difficult to identify failed invaders in invertebrates but the successful invaders mostly belong to families and genera already present in Europe, contrasting with Darwin's predictions. However, Darwin's hypothesis was somehow verified when considering the recruitment of phytophagous species by exotic tree species with and without native congeners. These without native congeners recruited only a limited entomofauna, mostly including polyphagous species, whereas exotic trees with native congeners quickly recruited most of the insects colonizing the native congeneric conifers.

Reproduction in desert rodents: how are environmental signals conveyed to the reproductive system?

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Changes in photoperiod are a key environmental cue for seasonal function of the reproductive systems in rodents existing outside of the tropics. For instance In Israel, the social vole *Microtus socialis* which is at its limit of distribution breeds mainly under short photoperiod (SP) conditions. On the contrary to Mediterranean ecosystems, the desert is an unpredictable environment in regards to seasonality as water input into it through precipitation is not guaranteed every year and when it rains it can be in different months of the year. As reproduction is a highly energy demanding process rodents should reproduce in seasons in which water and food are not limiting factors. Therefore, a question to be asked in regards to reproduction for a desert adapted rodent is: what reliable environmental signal is used by such species for reproduction? We have been studying this topic in our laboratory for several years using the golden spiny mouse *Acomys russatus* as an animal model. Tested mice were acclimated to two photoperiod regimes short day (SD) and long day (LD). In order to challenge the osmoregulatory system we used gradually increasing salinity up to 7% NaCl, in the water source of the

tested mice kept on a high protein diet. Our results showed that increasing salinity has a negative effect on the reproductive system of females and males. As we assumed that with the increase in salinity the mice will secrete more vasopressin (VP) which has an anti-diuretic effect.

A fitness cost for thermal tolerance in a marine copepod: Implication on long-term biological effects of global warming

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Tigriopus japonicus Mori, 1932 (Copepoda: Harpacticidae) are inhabitants of shallow supralittoral tidepools, in which they are subjected naturally to fluctuating temperatures. *T. japonicus* are, therefore, able to cope with a certain degree of thermal stress. However, different geographical populations of *T. japonicus* may have different extents of thermal tolerance, probably due to differences in genetic make-up and environmental temperature. This study used a two-generation experiment to compare the thermal tolerance of a temperate South Korean population (SK) and a tropical Hong Kong population (HK) of *T. japonicus*. 81 mate-guarding pairs (F0; 27 pairs each) were raised in three temperatures: 15, 20 and 25°C for 30 days. The first brood of each female was cultured in the same medium and their offspring were crossed with others exposed to the same temperature. The results showed that F0 of SK copepods demonstrated a lower lethal temperature (LT) at 42.9°C than F0 of HK copepods (LT: 45.3°C). At 25°C, although F1 of SK copepods showed an improved thermal tolerance, their reproductive output was significantly less than that of the HK's F1 group. Also, expression of heat shock proteins (hsp20 and hsp70) in the SK group was generally higher than that in HK group after heat stress at 30°C for 90 minutes. Our results suggest that multigenerational development of thermal tolerance in SK copepod is possible,

but at the expense of population fitness. Such observations have profound implications on the long-term biological consequence of global warming on *T. japonicus* populations along the Western Pacific coasts.

Amphibians in the Aegean Sea: the uncertain future of living in islands-in-islands

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The Aegean Sea encompasses over 4000 islands and harbors thousands of species, many of which are endemic. Among the diversity of habitats found in this region, none are more important, or more threatened than wetlands. Natural reasons, such as low amounts of rainfall, and

human-induced causes, such as water consumption, irrigation and touristic infrastructure, threaten with devastation these unique oases. For amphibians, strictly tied to standing water for their reproduction, Aegean wetlands function as a 'double island'. Having poor over-water dispersal abilities, amphibians were isolated in the insular wetlands by the formation of islands from rising sea levels. Since the Holocene they evolved in absolute isolation, following pathways of speciation. The Aegean Sea hosts ten species, three of which are endemic, while at the same time it is the western boundary for two species. Furthermore experts believe that some populations may be discrete species. Here we describe the condition of amphibian populations in Aegean Sea. Comparing climate parameters and wetland status over the last twenty years, we attempt to assess the survival possibilities for Aegean amphibians. Unfortunately our findings suggest that their future is uncertain. Many wetlands have been drained, while average temperature has risen and rain levels decreased considerably. Habitat fragmentation due to road building, non-sustainable water use and tourism are the main dangers facing remaining wetlands. To evaluate the status of the Aegean amphibians, specialized studies that monitor key species and survey populations are required.

Stream 1: Biological consequences of global change

Physiological responses to climate changes in small mammals

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Small mammals in temperate zones usually show seasonality in body mass, energy intake, and thermogenesis with changes in ambient temperature, photoperiod, and food availability and quality. Plateau pika and root voles are dominant species in the Qinghai-Tibet plateau and Mongolian gerbil and Brandt's vole are dominant species in Inner Mongolian grasslands. The Qinghai-Tibet Plateau and Inner Mongolia have different climate properties but both have a long cold period. Small mammals have difficulty migrating or undertaking long-distance dispersal when environmental conditions and the climate change greatly. We expected all small mammals have the ability to use environmental factors as a cue(s) for their physiological functional changes to adapt their micro- or macro-environmental variations. All four species mentioned above show a physiological sensitivity to cold weather. Plateau pika and Mongolian gerbils are relatively non-sensitive to photoperiod and root vole and Brand's vole are sensitive both to temperature and photoperiod. Food quality is also important for their survival and reproduction. Taken together, we can expect that global warming will change the geographic distribution and physiological function for these small mammals. Understanding the mechanisms of these small mammals to adapt to changes in environmental conditions will help us to predict and understand the effect of global climate changes on our survival.

Consequences of global change in the Sundarban mangrove ecosystem, Bangladesh

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The Sundarban is the world's largest contiguous mangrove and covers approximately 600 000 hectares in Bangladesh alone. Characteristics and interactions of major ecosystem components especially hydrology, topography, texture of the substratum have resulted in very high habitat heterogeneity and thereby house one of the richest natural gene pools for fauna and flora in the world. The forest consists of small forested islands and mudflats separated by a complex network of tidal waterways, and exemplifies the ecological processes of monsoon rain flooding, delta formation, tidal influence and plant colonization. Since the viability of the Sundarbans rests on the hydrology of the Ganges and its tributaries, which supply the fresh water influx, climate change is expected to have significant impact on this system. In addition to the altered hydrology, sea level rise will also have adverse impacts on the forest, directly through enhanced inundation and indirectly by enhancing saline intrusion in river systems. In this paper, efforts are made to address a shortcoming in our understanding of how climate change will affect this important area using local ecological knowledge and scientific ecological knowledge. Local knowledge revealed that many people believe plants and animals have declined and salinity has increased. Among animals, 90% of fish have declined, decreased, 4% of birds, 3% of mammals, 2% of amphibians and 1% of reptiles. Salinity is ranked as the most important (47%) factor out of five ecological components (plant, animal, water level, soil degradation and salinity).

Physiological and reproductive responses of amphibians to climate change

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Over the next century, average global temperatures are expected to rise several degrees Centigrade, and current precipitation patterns are predicted to change. Extreme weather events (record-breaking storms, temperatures, and droughts) are predicted to become more common than now.

The current distribution of amphibians is determined more by moisture patterns than temperature. Although many terrestrial amphibians have some tolerance to desiccation, extremely long or intense droughts are likely to cause many species extinctions. Temperature is a factor that determines biochemical, cellular and physiological rate processes. A 10 C increase in rate will cause most physiological rates, such as metabolism, heart rate, digestive rate, etc. to increase two- to three-fold. Most amphibians currently live in areas in which they are not exposed to temperatures that come close to their upper thermal tolerances and even if such temperatures exist in their habitats, they avoid them behaviorally by nocturnality and use of microclimates. As long as food supplies remain available, it is anticipated that changes in precipitation rather than temperature will pose the greatest challenges to amphibians. Some data exist that amphibians are responding to increasing spring temperatures by breeding earlier than previously. These studies are largely correlational, and much more study of cause and effect of rising temperatures and the consequences of earlier breeding are needed.

Sub-lethal impacts of physical stress on intertidal limpets and barnacles: how important are multiple stressors?

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The large-scale distribution of intertidal organisms is strongly determined by geographic climatic and oceanographic processes, and on a smaller scale by the steep environmental gradient dictated by the tide. Extremes of environmental conditions (physical stress) are traditionally invoked to explain the distribution limits of intertidal organisms and there is much speculation over the potential role of climate change affecting large-scale patterns of species distribution. The evidence to support this paradigm is compelling, but our understanding of actual effects and therefore our ability to accurately predict possible impacts are poor. Experimental tests of the importance of these physical stressors are often performed

in the laboratory, under un-realistic conditions and usually only investigating the isolated effects of one stress. Data from tropical areas are also noticeably limited, and projections for these areas are often based on temperate comparisons. The limpet genus *Cellana* and barnacle *Tetraclita* are common along the Western Pacific coast and are therefore good candidates to investigate the effects of physical stress on their distribution and survival. Using a variety of field-based and laboratory experiments, involving both heat stress and the effects of monsoon rains, both single and synergistic effects of environmental stressors can be demonstrated. These lead to direct effects on populations but also potential indirect effects on intertidal communities. More realistic experiments, based on good natural history and investigation of the effects of multiple stressors are needed, in order to achieve more precise predictions of the potential effects of climate change.

Predicting effects of climate change on large mammals of Armenia's mountains

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The patchy nature of mountainous habitats and the high demand of large mammals for space make these landscapes and species particularly vulnerable to climate change. Only two species, golden jackal (*Canis aureus* L., 1758) and Indian crested porcupine (*Hystrix indica* Kerr, 1792), benefit from warming in Armenia due to their omnivorous diet and tolerance to arid conditions and human activities. The Armenian mouflon (*Ovis orientalis gmelinii* Blyth, 1841), European roe deer (*Capreolus capreolus* L., 1758), grey wolf (*Canis lupus* L., 1758), bezoar goat (*Capra aegagrus* Erxleben, 1777) and leopard (*Panthera pardus* L., 1758) are tentative losers. Mouflons use the snow-free areas for wintering and would possibly benefit from warming-driven snowmelt rates, but their life is strongly linked with snow cover which offers soft substrate for basking and ensures moisture for lush alpine vegetation. Roe deer inhabit gorges in moist broadleaf forests and suffer from dryout and reduction of local dense vegetation. Wolf distribution is correlated with mountain pastures for domestic livestock and drought-triggered hardship of animal husbandry causes

wolves to dwindle or shift to predation on wild boars (*Sus scrofa* L., 1758). The heat-loving bezoar goat and leopard would likely benefit from accelerated snowmelt, but azonal precipitous cliffs where they live are small, scattered and contain few options to retreat from hunger during drought. The brown bear (*Ursus arctos* L., 1758), Eurasian lynx (*Lynx lynx* L., 1758) and wild boar inhabit arid sparse and moist broadleaf forests, so their response to climate change would depend on forest-specific pressures of warming.

Potential impacts of climate change on Yangtze River cetaceans

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Aquatic animals in temperate regions, particularly riverine cetaceans, are susceptible to climate change because of the limitation of their distribution and the vulnerability of their habitat. Yangtze finless porpoise is the only freshwater subspecies of the *Neophocaena phocaenoides* and lives in the middle and lower reaches of the Yangtze River and associated lakes. We collected some evidence of the impacts of climate change on this animal. Death of finless porpoises directly caused by extreme weather was discovered: five animals including a pregnant female in Tian-E-Zhou Reserve, which is the only *ex situ* conservation practice for cetaceans in the world, were killed by ice cover in early 2008 due to an extreme snowstorm that dominated southern China. This was the first time the reserve had frozen since it was founded in 1972. This incident reminds us that extreme weather will be a challenge for the conservation of the Yangtze finless porpoise in this reserve. Here, we also report on the male-skewed population structure of the Yangtze finless porpoise. In summary, we propose that possible impacts of climate change on the reproduction of this river cetacean by referring to anatomic and physiological characteristics of this species.

Climate change and invasive species: double jeopardy

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Climate change is already having a measurable impact on nature and all evidence suggests that things will get worse – temperatures will increase, sea levels will rise, ocean chemistry will change – even if we act tomorrow to mitigate any future increases in greenhouse gas emissions. At the same time, invasive species remain an important threat to biodiversity globally. Interactions between these two threats are resulting in increased impacts on the environment and people's livelihoods. Taking action against one of these threats without considering the additional influence of the other can lead to unexpected, and potentially harmful, results. Managing the combined impacts is vital yet presents greater challenges to field conservationists as well as policy makers. Tools are available to support planning and implementation of mitigation and adaptation action and awareness of the links between the two should underpin all management planning.

Effect of fur removal on thermal conductance and energy budget in lactating Swiss mice

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The limits to sustained energy intake are important because they determine the ceiling within which all competing physiological processes are limited. It has recently been posited that sustained energy intake is limited by capacity to dissipate heat. However some previous data for Swiss mice (*Mus musculus*) are inconsistent with this hypothesis. To examine the role of heat dissipation limits on sustained energy intake, the body temperature, thermal conductance and lactation performance were measured in dorsally shaved Swiss mice. Shaving reduces external insulation and the heat dissipation limits hypothesis predicts such animals should be capable of eating more food and raising heavier litters. Shaved mice had a significantly higher thermal conductance and a faster reduction in body temperature

following noradrenalin injection. At peak lactation, shaved mice spent more time in feeding behavior, and increased food intake above that observed in non-shaved controls, indicating that limitation on sustained energy intake might be imposed by the capacity to dissipate heat. However, shaved females did not spend more time in suckling their pups, and did not raise heavier litters, which was inconsistent with the expectation of heat dissipation limitation hypothesis. The strong correlations between resting, feeding and suckling behavior at peak lactation suggested that there might be a trade-off in the time distribution between the behavioral patterns. These data suggest that limits on performance may be set at different levels in different strains or species. Further work on a range of additional species or strains will be necessary.

Biochemical adaptations of intertidal limpets to thermal stress: organism's distribution and climate change

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Limpets are common inhabitants of rocky intertidal communities, occupying different intertidal zones and microhabitats. In the intertidal zone, high temperatures during low tide can play an important role in determining species' zonation patterns. To elucidate the physiological adaptations of limpets to thermal stress, heat shock responses and functional and structural properties of cytoplasmic malate dehydrogenases (cMDHs) were studied among groups of closely related limpet species with different latitudinal and vertical ranges. Results suggest different strategies of Hsp70 expression in limpets occupying different heights on both temperate (species in the genus *Lottia*, California, USA) and tropical rocky shores (species in the genus *Cellana*, Hong Kong). High intertidal animals (*L. scabra* in Monterey; *C. grata* in Hong Kong) had higher constitutive levels of Hsp70 than low- and mid-

intertidal species. The higher levels of constitutive Hsp70 are seen as a possible "preparative defense" strategy to tolerate the extreme and unpredictable environments experienced in the high-intertidal zone. Differences in apparent Michaelis–Menten constants of the cofactor NADH (K_m NADH) and thermal stability between orthologs of *L. digitalis* and *L. austrodigitalis* result from a single amino acid substitution. At position 291, the glycine residue in cMDH of *L. digitalis* is replaced by serine in *L. austrodigitalis*, a change that favours additional hydrogen bonding and reduced conformational entropy. This difference between closely related congeners demonstrates the role of minor alterations in protein sequence in temperature adaptation and suggests that such variation is important in governing shifts in biogeographic range in response to climate change.

Himalayan brown bear conservation and threats in India

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Kugti wildlife sanctuaries, Chamba district, Himachal Pradesh, the Himalayan brown bear (*Ursus arctos isabellinus*) occurs in low densities and is potentially facing a precarious future. Due to habitat degradation, livestock grazing pressure (sheep and goat), collection of medicinal plants, brown bear population is highly disturbed and threatened. Food habits, habitat use pattern and nomadic shepherds-brown bear conflicts have been studied in Kugti wildlife sanctuary during 2002-2004. Dietary composition revealed that the frequency of occurrence of plant matter was higher than the animal matter in all the seasons. Based on 57 direct feeding observations, brown bears were found feeding on 29 species of plants including agricultural crops. There were a total of 1539 livestock casualties by brown bears in different pastures during 2002-2004. Maximum number of livestock depredation occurred during mid-night. Crop depredation by brown bear was found to be quite visible in crop fields. For conservation and management of brown bear, there is need for planning strategies for conflict mitigation, regulation or imposing restriction on livestock grazing, collection of medicinal plants in the highly

preferred habitat use areas.

Environmental proteomics: Proteomic plasticity in larval and juvenile marine invertebrates to climate change and ocean acidification

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Climate change consequences such as rising sea surface temperature and augmented ocean acidification are expected to have deleterious effects on marine calcifiers within this century. Our understanding of organism's adaptive response at molecular level, such as proteome plasticity, to these emerging stressors remains poor, though it is an important milestone in predicting the sustainability of marine ecosystems in the future. Recent advances in the proteomic technology can provide a direct measurement of functional gene expression in terms of presence and relative abundance of proteins. Proteomics rely on two-dimensional

gel electrophoresis, where the proteomes are separated first according to charge (i.e., isoelectric point, pI) and then molecular size (M_w). Following staining, selected protein spots will be trypsin-digested and their identity can be determined using matrix-assisted laser desorption ionization-time-of-flight (MALDI-TOF/TOF) mass spectrometry. In this project, a dominant benthic marine invertebrate in the global intertidal zone, the barnacle *Balanus amphitrite* Darwin (Cirripedia; Thoracica), is used as the animal model. The aim of this study is to investigate the effect of three climate change-driven environmental stresses (either alone or in combination), namely (i) ocean acidification, (ii) reduced surface seawater salinity brought about by precipitation extremes, and (iii) increased surface seawater temperature, on the proteome plasticity of *B. amphitrite* at the sensitive larval and rapidly developing juvenile stages. Through this research, the adaptability of the critical early life-stages of the barnacle in the adverse environmental conditions as well as the mode-of-action of these stressors would be better understood.

Stream 2: Marine organisms and amphibians

Temperature-dependent toxicities of chlorothalonil and copper pyrithione to the marine copepod *Tigriopus japonicus*

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The anticipated anthropogenically driven climate change may result in increased incidents of temperature extremes that may have profound implications on the toxicity of chemical contaminants and hence their ecological risks to marine organisms. We hypothesized that chemical toxicity generally increases with increasing temperature over the thermal tolerance range (TTR) of a species, and further exacerbates at temperatures lower or higher than the TTR. We tested this hypothesis by investigating the acute toxicity of two popular antifouling biocides, chlorothalonil and copper pyrithione (CuPT), to the marine copepod *Tigriopus japonicus* Mori, 1932 (Copepoda: Harpacticidae) over a wide range of temperatures. Our results verified that toxicities of the two biocides, in terms of 96h-LC50, were highly temperature-dependent. The LC50 values of *T. japonicus* exposed to CuPT decreased with increasing temperature between 15–31°C (i.e. assumed TTR), whereas the LC50 values at 4 and 35°C were significantly lower than those at 10 and 25°C, respectively. The toxicity of chlorothalonil to the copepod followed the same pattern of CuPT between 25 and 35°C but the LC50 values between 4 and 25°C were similar. Such dissimilar temperature-dependent toxicity profiles between the two biocides may be partially attributable to the differences in temperature-mediated modifications of their physicochemical properties, bioavailability, toxicokinetics, and thus resultant toxicity to the test organism.

Human impacts on Eastern Pacific green turtles in Costa Rica

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The Pacific coast of Costa Rica hosts important nesting sites for the endangered green turtle (*Chelonia mydas*). We studied the nesting of this species, the areas it occupied and its behavior during an inter-nesting period. We observed turtles on the nesting beach and attached ten satellite transmitters to turtles nesting on Playa Nombre de Jesús and Playa Zapotillal. The satellite transmitters were equipped with a data recorder and allowed us to analyze diving behavior. Turtles spent the 14 day inter-nesting interval in the vicinity of the nesting beach (from the Papagayo Gulf to the Tamarindo Bay). Turtles occupied depths between five and 15 m and executed dives that lasted between two and 25 min. Diving profiles obtained for four turtles were characterized by “U” shaped dives and suggests that the predominant behavior during this interval was resting. Through direct observation we encountered fishing boats using artisanal longlines and gillnets operating in the area and turtles caught on longlines. This population is also threatened by poaching of eggs and uncontrolled tourism activity on nesting beaches. Thus, human activity threatens this turtle both on land and in the water. Conservation of this species will require not only beach patrols to protect nesting turtles and their eggs but water patrols to control unregulated fishing by local people.

Raising atmospheric carbon dioxide, ocean acidification and larval metamorphosis: developmental, chemical and molecular approaches

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The transformation of pelagic larvae to benthic adults (“metamorphosis”) in a variety of marine organisms is energetically demanding and involves rapid physiological and molecular changes. Understanding how this process is affected at developmental, physiological, and molecular levels as atmospheric carbon dioxide increases has far-reaching implications for present and future aquaculture, fisheries, and coastal management. The past and predicted rises in pCO₂, and subsequent decreases in oceanic pH (ocean acidification), call for quantitative studies on the effects of increased pCO₂ on larval metamorphosis. The critical question of how elevated pCO₂ might affect marine larvae remains virtually unaddressed. To fill this gap, we simultaneously measured developmental (metamorphic success), physiological health (energy reserves), and molecular (proteome – all proteins expressed by a genome) responses of the barnacle, *Balanus amphitrite* Darwin (Cirripedia; Thoracica) larvae by exposing them to various levels of pCO₂. Our results showed that there was a strong and significant decrease/inhibition in larval metamorphosis at elevated pCO₂, as well as a significant trend in decreasing energy reserves. This is the first study to examine the changes in protein expression pattern (proteome plasticity) due to ocean acidification. We demonstrated that proteomics technology can be an effective tool when identifying changes in larval protein expression signatures as a response to ocean acidification. For instance, we have discovered remarkable proteome plasticity as the barnacle larvae respond to increased pCO₂. Our results suggested that the predicted future seawater CO₂ conditions would potentially adversely affect the metamorphosis of barnacle larvae, possibly through reducing their energy reserves.

Conservation genetics of the American crocodile along the Pacific coast of Costa Rica

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Understanding the genetics of populations is crucial to the long term management and survival of endangered and threatened species. The American crocodile (*Crocodylus acutus*) is listed as endangered throughout much of its range. Despite this, little is published about this species. We carried out this study to describe the genetic structure of *C. acutus* populations living along the Pacific coast of Costa Rica. We collected blood and tissue samples from 164 *C. acutus* individuals from five different areas. Ten microsatellite loci characterized the genetic structure and gene flow rates within and between these populations. All Costa Rican populations were in Hardy-Weinberg equilibrium for at least three loci and moderate differentiation was found between all population pairs (FST = 0.132). The populations were significantly different and a small amount of gene flow (Nm = 0.6 migrants per generation) occurred between populations. Observed heterozygosity for all loci was 0.644 indicating that the populations are genetically diverse. No significant correlation between genetic subdivision and geographic distance was found. These data show that crocodile populations should be managed separately in Costa Rica due to the moderate degree of differentiation. These findings can now be used to create more effective management plans and help to minimize human impacts for *C. acutus* in Costa Rica.

The acorn barnacle as a biological habitat on thermally stressed intertidal rocky shores

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Positive interactions, whereby one species ameliorates environmental stresses, such as heat and desiccation for another species are predicted to become more important as these stresses become more severe, particularly under predicted climate change scenarios. On Hong Kong shores, low tides fall in the afternoon during the summer monsoon resulting in stressful periods when animals can be emersed for up to 8 hours and rock temperatures can exceed 50°C.

As a result, many mobile intertidal species migrate down the shore and hide in refuges when inactive, but despite these strategies, mass mortalities of these species can occur. Throughout the year, the acorn barnacle *Tetraclita japonica* Pilsbry, 1916 maintains ~ 40% cover on the mid shore, and many mobile organisms are found hiding amongst and within empty tests during emersion periods. Laboratory experiments tested the hypothesis that barnacles create a biological habitat, reducing environmental stress for mobile species via a shading effect. When heated, mobile species on open rock surfaces suffered from increased heart rates and osmotic stress, as mantle water and haemolymph osmolality increased. In contrast, animals taking refuge in the shade of barnacle tests had similar heart rates, mantle water and haemolymph osmolalities as unstressed individuals. During the tropical summer therefore, associating with barnacles is an important interaction for mobile species to reduce thermal stress. Given the wide distribution of the genus *Tetraclita* in the Western Pacific, its role as a biological refuge is likely to be more important where environmental stresses are predicted to become more severe.

Trophic niche differences along a predation gradient in two species of neotropical stream fish from Trinidad

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A species' isotopic niche represents the area occupied by individuals in the δ -space – obtained by plotting $\delta^{15}\text{N}$ against $\delta^{13}\text{C}$. As part of a broader NSF-FIBR, we assessed

the isotopic niche of two fish species, *Rivulus hartii* (Hart's killifish) and *Poecilia reticulata* (guppy), from nine Trinidadian streams characterized by different predation and competition regimes. We also performed gut analyses and conducted behavioral observations on the two species. In each stream, fishes were sampled and observed from three different communities: Rivulus+Poecilia+other fish predators, Rivulus+Poecilia, and Rivulus only. We hypothesize that the niche breadth of the two fish species is directly affected by different predation and competition regimes. Preliminary results suggest that *Rivulus* and *Poecilia* show a larger niche overlap when they co-occur with predators. When only two species are present, their niche overlap is smaller, due to a greater resource partitioning. The diets of both *Rivulus* and *Poecilia* changes from sites with high and low predation level. We found no evidence of ontogenetic niche shift in *Poecilia*. *Rivulus* showed ontogenetic niche shift when released from predation pressure and competition. Our findings confirm the utility of multiple approaches to understand community dynamics in tropical streams.

Mitochondrial DNA diversity of *Coilia mystus* (Clupeiformes: Engraulidae) in three Chinese estuaries

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Sequences of mitochondrial cytb and 16SrRNA gene segments were obtained in order to clarify the genetic diversity and population structure in three Chinese estuary populations of *Coilia mystus*: 21 individuals from ChangJiang River estuary, 22 from MinJiang River estuary, and 22 from ZhuJiang River estuary. We obtained 607 bp of consensus cytb sequence. Thirty four distinct haplotypes were detected among the 65 cytb sequences. The indexes of nucleotide diversity (π) in these three populations were ChangJiang 0.533%, MinJiang 1.135%, and ZhuJiang 0.268%. Genetic distances within the populations were between 0.3 and 1.2%, and 0.8 to 10.8% among populations. The largest genetic distance was 10.8% between the ChangJiang and ZhuJiang populations, and the smallest was 0.8% between

MinJiang and Zhujiang populations. Analysis of molecular variance (AMOVA) analysis revealed that variation among populations accounts for 90.25% of total variation, suggesting that this is the main source of total variance. We obtained 470 base pairs of consensus sequence of 16SrRNA. We detected 19 distinct haplotypes among the 65 sequences. The indexes of nucleotide diversity (π) in these three populations were ChangJiang 0.108%, MinJiang 0.843%, and Zhujiang 0.097%. Genetic distances were between 0.1 and 0.9% within populations and 0.5 to 1.9% between populations. The largest genetic distance was the 1.9% between the ChangJiang and MinJiang populations, and the smallest was 0.5% between the MinJiang and the Zhujiang populations. AMOVA analysis disclosed that variation among populations accounts for 74.61% of total variation, suggesting that this is the main source of total variation.

Proteomic profiling of metallothionein isoforms in the green-lipped mussel exposed to cadmium and hydrogen peroxide

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Metallothioneins (MTs) are a group of low molecular weight, soluble, cysteine-rich, heat-stable, and non-enzymatic proteins with multiple physiological functions. In general, they are induced by and can bind to various kinds of trace metals, and thus have important functions in homeostasis of essential metals and detoxification of toxic metals or essential metals in excess. Recent studies suggest that MTs can also scavenge oxidative free radicals and thus play an essential antioxidant role for protecting cells and an entire organism from oxidative stress. This study was designed to use proteomic and genomic approaches to characterize different MT isoforms in hepatopancreas of the common marine biomonitor, the green-lipped mussel *Perna viridis* L., 1758 (Bivalvia: Mytilidae) after exposure to the control (seawater only), cadmium (0.5 ppm; Cd) and hydrogen

peroxide (2.0 ppm; H₂O₂), respectively for 14 days. After removal of non-MT proteins through heat-treatment, MTs enriched tissue lysates were investigated using two dimensional gel electrophoresis followed by immuno-blotting analysis. The results revealed that MTs were up-regulated in both Cd and H₂O₂ groups, and distinctive MT-like isoforms differentially expressed corresponding to the two different chemical treatments. Furthermore, ten MT isogenes were revealed in the tissues of *P. viridis* and could be further divided in two clusters with clear differences in amino acid sequences, molecular weights and isoelectric points. Both proteomic and genomic results confirmed that MTs in *P. viridis* exist as multiple isoforms and the expression of a particular MT isoform is likely associated with a specific type of chemical exposure (i.e. exposure specific).

Antibacterial and antifungal activity of some marine sponges from the Bay of Bengal

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Microbial promoted infectious diseases are a major public health issue worldwide and antibiotic resistant microorganisms are considered very problematic to human society. Marine organisms are a rich source of structurally novel and biologically active metabolites and their biological activity is being studied and developed as new pharmaceuticals. In this regard, we aimed to determine the antimicrobial activity of extracts from the marine sponges *Leucosolinia elenor* (Urban, 1905) and *Agelas conifera* (Schmidt, 1870) collected from the coast of the Bay of Bengal. Antimicrobial activity was determined against the following microorganisms such as *E. coli* (Migula, 1895), *Pseudomonas aeruginosa* (Schröter, 1872), *Klebsiella pneumonia* (Schroeter, 1886), *Staphylococcus aureus* (Rosenbach, 1884) and *Candida albicans* (Berkhout, 1923) using the paper disk assay method. Extracts of *L. elenor* were shown to have higher antimicrobial activity against all

microbes when compared to other extracts. However, the extracts of all three species (dry sample) were found to have higher antimicrobial activity against *E. Coli*, *P. aeruginosa* and *C. albicans*. It is important to note that the extracts of all the three marine sponge species (both dry and wet samples) were found to have significant antifungal activity against *C. albicans*. The present study concludes that the extracts of *L. eleanor* exhibited higher antimicrobial activity compared to other species, and further research is planned to identify the chemical compounds involved in antimicrobial activity.

Microbial community structure in sand on two olive ridley nesting beaches, Costa Rica

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Olive ridley (*Lepidochelys olivacea*) turtles nest in high density events which are referred to as 'arribadas' (Spanish for arrival) at only a few nesting beaches in the world. Arribada nesting behavior results in the destruction of a large number of clutches by subsequent nesting turtles that inadvertently dig out previously laid clutches. The increased organic content (broken eggs) in the sand then causes high microbial build up in nests and it has been speculated that egg harvest may increase hatching success by reducing the number of decomposing eggs in the sand. This speculation is the basis for many management programs for olive ridley arribada nesting beaches around the world. In this study, microbial community structure, diversity and abundance was assessed via terminal restriction fragment (TRF) analysis on two sea turtle nesting beaches in Central America, Playa La Flor in Nicaragua and Playa Nancite in Costa Rica. Phylotype richness and diversity of bacteria was higher in high nest density at Playa Nancite and in higher zones of the beach at Playa La Flor. Phylotype abundance did not change in different zones of the beach or in different densities at both Playa Nancite and Playa La Flor. To better conserve olive ridley sea turtles and manage egg harvest on arribada nesting beaches further studies should focus on identifying bacterial species that are pathogenic to turtle eggs and their effects on hatching success.

Comparative proteomic responses in intertidal limpets to summer low tides on tropical rocky shores

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The tropical intertidal zone is an extremely harsh physical environment, dictated by the tidal cycle and daily exposure to the sun's heat. Intertidal organisms adopt different behavioral and physiological strategies to survive these stressful conditions. Physiological responses can involve the collective actions of protein expression and modification, which can be evaluated using expression proteomic and phosphoproteomic approaches. On-shore protein expression profiles and phosphorylation patterns were measured in the foot muscle of two intertidal limpets, *Cellana grata* and *C. toreuma*, over a summer low tide on a rocky shore in Hong Kong. In a preliminary study, animals were collected whilst "Awash", "Emersed" or "Re-awash" phases of the tidal cycle when temperatures during emersion exceeded 38°C and showed differential patterns of protein expression and level of phosphorylation. Using two-dimensional electrophoresis, ~200 protein spots were resolved from the limpet foot muscle proteome, of which expression of 28 spots were up-regulated and 27 spots were down-regulated from the "Awash" to "Emersed" phase. From the "Emersed" to "Re-awash" phase, 14 and 39 spots were up- and down- regulated, respectively. The number of phosphoprotein spots was also greatly increased in the "Emersed" than in "Awashed" and "Re-awash" phases, the average percentage of proteins phosphoslyated being 25%, 48% and 32% in "Awash", "Emersed" and "Re-awash" phases, respectively. The observation suggested that substantial actions in both protein expression and modification are undertaken over the tidal transition. Further identification of the differentially expressed and phosphoslyated protein spots are being conducted using MALDI-TOF/TOF tandem mass spectrometry.

Where and when to fish sea urchins? Spatial and

temporal variation in sea urchin population structure, gonad yield and food source utilization within and outside Marine Protected Areas

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Sea urchins are considered to be functionally important in determining biological structure of rocky substrates. The relationship between high grazing pressure (i.e. urchin abundance) and dominance of encrusting coralline algae (i.e. coralline barrens) is well-known all over the world. In Hong Kong, amongst three common sea urchin species, only the sea urchin, *Anthocidaris crassispina* (A. Agassiz, 1863; Echinoidea: Echinometridae), is heavily exploited by fishermen, especially from Mainland China. Such selective

exploitation of a particular urchin species is likely to change sea urchin and sessile species composition on rocky substrates and results in variation of food source utilization and gonad yield of sea urchins. Stock assessment (density and size) of sea urchins was conducted through bimonthly underwater surveys in 20 rocky habitats for one year (2008 - 2009) in Hong Kong waters. Sessile species compositions (i.e. potential food sources of urchins) on rock substrates were recorded in both summer and winter. Stable isotopic ratios and fatty acid profiles and gonad biomass of sea urchins were determined to investigate spatial variation in food source utilization, sea urchin size and maximum gonad yield. Sea urchin populations within and outside the Marine Protected Areas (MPAs) will be compared. Based on the overall results, the function of local MPAs in conserving sea urchin stock is discussed. This work is supported by the Research Grants Council (CERG Project: HKU 778207M) and the Environment and Conservation Fund (ECF Project: 04/2007).

Stream 3: Species conservation and biodiversity

Wildlife conservation priorities and threats in India

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India's *in situ* Wildlife Network encompasses 96 national parks, 510 wildlife sanctuaries, 30 tiger reserves and 25 elephant reserves. Schedule 1 of the IWPA lists 80 mammalian, 48 avian 23 reptilian and three amphibian species. Central Government-aided conservation projects exist for the tiger (*Panthera tigris tigris*), Asiatic lion (*P. leo persica*), snow leopard (*Uncia uncia*), red panda (*Alurus fulgens*), rhinoceros (*Rhinoceros unicornis*), Asian elephant (*Elaphas maximus*), lion-tailed macaque (*Mcaca silenus*), tragopan pheasant (*Tragopan melancephalus*), vulture (*Gyps spp.*) and gharial (*Gavialis gangeticus*). Key projects on these species and the consequences for wildlife management are discussed. Finally, I propose that a data bank is needed at a national level to collect information on intrinsic and extrinsic factors affecting the conservation of key species in India.

Mus musculus sensu lato complex: adaptations and evolution of invasive species

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The purpose of presentation consists of analysis of adaptations of commensal species of *Mus musculus* s.l. complex and role of invasions in their speciation and evolution. Our investigation of behavior in large enclosures demonstrated that commensal (but not wildliving) species exhibited high level and specific strategy of exploratory activity, different patterns of neophobic behavior to new objects and high plasticity of spatial-ethological structure of demes. Additionally ethological and ecological characters of commensal species are: adaptation to human activity and high level of stress reactivity, high level of mobility and

quick change style of life (migrant-resident); high level of interspecific aggression and dominance in human dwellings. Our conclusion is that the phenomenon of commensalism is result of a specific combination of physiological, behavioral and ecological characters, and not some unique single character. Analysis of hybrid populations of house mice in Russia demonstrated the particular significance of hybridisation in the evolution of commensal taxa. This enhanced role in commensals is linked to their unique ability to expand their geographic range through human exploitation and even survive as commensals in areas that are beyond their physiological tolerance.

Life history patterns and fitness in Hainan Eld's deer

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By using field data on life history traits such as age specific fecundity, age specific mortality, age at first reproduction, brood size and adult expectancy, we analyzed life history patterns and population fitness of an *in situ* population of Hainan Eld's deer (*Cervus eldi hainanus* Thomas, 1918), an endemic and endangered species inhabiting Bangxi Natural Reserve of Hainan Island. The population fitness index was found to be 0.012, which was at the second lowest level compared with other mammal species. Our results show that this precious species is still highly endangered and needs further careful protection.

Conspecific competition enhances intensity of wild apricot seed-hoarding in Korean field mice and Chinese white-bellied rats

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The presence of conspecific competitors may modify food hoarding strategies of rodents as their presence can impact upon the risk of food pilferage, forage competition and sexual social dominance. It is not well clarified how hoarding intensity (scatter- vs larder- hoarding) of rodents changes in the presence of their conspecific competitors. In this study, we investigated behavioral responses of Korean field mice (*Apodemus peninsulae*) and Chinese white-bellied rats (*Niviventer confucianus*) in the presence of same- or opposite- sex conspecific competitors in semi-enclosures in a warm temperature forest in China. The study aims to assess how the risk of pilferage, forage competition and sexual dominance might change seed-hoarding intensity in these species. We found that presence of both same-sex and opposite-sex conspecific competitors increased seed removal and total seed-hoarding, supporting both the pilferage compensation hypothesis and forage competition hypothesis. We found the Korean field mice shifted their hoarding strategy from a scatter hoarding to a majority of larder hoarding. We did not find a significance difference in seed-hoarding intensity between same-sex competitor and opposite-sex competitor groups, suggesting sexual dominance might not be important in modifying hoarding behaviors. We also found large-sized Chinese white-bellied rats had only larder hoarding behavior, while the smaller sized Korean field mice did both scatter- and larder-hoarding. Body size related dominance may help to explain the scatter- or larder- hoarding strategy in these two species, but other factors (e.g. reproduction, forage competition) may also attribute to the evolution of hoarding behaviors in small rodents.

Comparative analysis of sexual behavior in the closely related species *Mus musculus* and *Mus spicilegus*

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Mus musculus (L., 1758) and *M. spicilegus* (Petényi, 1882) (mound-building mouse) are sympatric species belonging to the *Mus musculus s.lato* species group. *M. musculus* is polygynous and *M. spicilegus* is presumably monogamous. The purpose of this study was to compare sexual and other patterns of behavior of *M. musculus* and *M. spicilegus* in relation to differences in social organization, mating systems and ecology. Under laboratory conditions we investigated the behavioral patterns (sexual, agnostic and amicable behaviors) of these species over forty 90 min dyadic hetero- and homospecific encounters. I found a difference in the frequency and duration of some copulatory, agnostic and amicable behaviours and I propose that these differences can be connected to species-specific mating system and social organization in these closely related species.

Comparison of fatty acids in wild and farmed freshwater prawns

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We examined one group of *Macrobrachium rosenbergii* (De Man, 1879) from the wild and another one from two commercial farms. Fatty acid profiles of the midgut gland (MG), ovary and eggs were studied to find the amount present during sexual maturation and spawning. In both groups mono-saturated, polyunsaturated and highly unsaturated fatty acids (14:0, 16:0, 18:0, 18:1n-9, 18:2n-6, 18:3 n-3, 20:4 n-6, 20:5 n-3 and 22:6n-3) were present in large quantities. Among these saturated fatty acids dominate over mono-unsaturated (MUFA), polyunsaturated (PUFA) and highly unsaturated fatty acids (HUFA). Though all the four groups of fatty acids are found in both groups, with the exception of n-3 HUFA other categories are higher in wild brooders. Furthermore, there is a significant difference

between total weight, total length and clutch weight of farm and wild brooders, but the gonadosomatic (GSI) and midgut gland somatic index (MSI) did not vary. These findings clearly indicate that wild brooders possess greater amounts of all essential fatty acids than farmed brooders.

Integrative research on *Miscanthus* plants: ecology to molecular evolution.

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Miscanthus (Anderss) is widely distributed in Asia and across the Pacific and is comprised of 20 species. The plant is a C4 perennial grass with high productivity of biomass. In the 19th and early 20th centuries in Taiwan, *Miscanthus* was a very important crop used for forage grass, clothing, and shelter. The relatively high germination, and high yield of biomass made the plant available for people of Taiwan. While the taxonomic knowledge of *Miscanthus* is well developed, its ecology has been a topic only since 1972. Chou and his associates paid great attention to elucidate the mechanism of dominance of *Miscanthus* vegetation and found that allelopathy plays an important role. In addition, the population biology of *Miscanthus* taxa by using polyacrylamide gel electrophoreses to examine patterns of peroxidase and esterase among populations (over 100) of *Miscanthus* in Taiwan have been completed. This research group has also elucidated the phylogenetic relationship among species and varieties in Taiwan and molecular phylogeny was attempted to clarify the population heterogeneity of *Miscanthus sinensis* complex. This knowledge is now available for engineering hybridization between *Miscanthus* species and its related species, such as *Saccharum* (sugar cane) spp. which is a high energy resource plant. European scientists used Asian *Miscanthus* species and bred a new hybrid called *Miscanthus x giganteus*, which is now being used as bio-fuel in Europe and may be widely used around the world.

Mother-young relations and prey capture in the Indian false vampire bat

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The Indian false vampire bat (*Megaderma lyra* Geoffroy, 1810) lives in caves, unused buildings and temples. It feeds on frogs, mice and geckos and in order to detect and capture prey on ground uses a combination of passive listening to prey-generated sound, echolocation and possibly vision. To capture prey at water surface, for example frogs, the bat uses echolocation. *M. lyra* gives birth to a single young from March to May. In a study under captive conditions, four out of eight young (Group 1) at 60-63 days of age began to capture dead frogs that were pulled with a long thread on the sandy floor of the flight room. However, the mothers continued to suckle until their young became 85 days old. The mothers of the remaining four young (Group 2) stopped suckling when their young attained the age of 60 days. Nevertheless, these mothers transferred either entire or partly consumed frogs (bodies with no head, half bodies, paired hind limbs and single hind limbs) to their young solicitors. Such food transfers occurred based on the body lengths of frogs. Mothers transferred small frogs entirely, but as the body length of frogs increased, mothers transferred smaller body parts to their young. Occasionally, audible vocalizations of mother and young were associated with food transfers. When these young bats became 74 days old, their mothers stopped food transfers. It appears that lactating females of *M. lyra* take care of their young by supplementing milk with solid food, similar to other megadermatid bats.

Molecular evolution of GPCR in pheromone signalling in mammals

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In the realm of pheromone research, few advances have outstripped the role that G protein-coupled receptors (GPCRs) have come to play in the development of signaling agents. In this review the authors attempt to enhance our understanding of how GPCRs are evolved to organize signal transduction and intracellular activities. Pre-Darwinian, Darwinian and neo-Darwinian concepts are discussed with special reference to the evolution of GPCRs structure and functions. G protein-coupled receptors (GPCRs) constitute a large and diverse family of proteins whose primary function is to convert extra-cellular stimuli into intra-cellular signals. They belong to a large and most diverse protein family. Based on their homology with rhodopsin, they are identified as a member of seven trans-membrane protein sub-family. These proteins have an extra-cellular N-terminus and an intracellular C-terminus and also named as seven trans-membrane receptors (or) the heptahelical receptors. Interestingly GPCRs transduce extra-cellular stimuli to give intra-cellular signals through interaction of their intracellular domains with heterotrimeric G-protein. Here, we mainly focus on the molecular evolution of GPCRs. The presence of GPCRs in the genomes of bacteria, yeast, plants, nematodes and other invertebrate groups argues in favor of a relatively early evolutionary origin of this group of molecules. Evolutionary analysis has revealed the diversity of GPCRs based on both multiplicity of stimuli to which they respond as well as by the variety of intra cellular signaling pathways, they activate.

Re-establishment of harpy eagle populations in Central America through captive breeding, re-introduction and community education

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Habitat loss and hunting pressure are the two largest threats to top predators across all continents. The harpy eagle (*Harpia Harpyja*) is the world's largest raptor and has suffered severe population declines across South and Central America. Much of the preferred habitat of harpy eagles has been converted to agriculture. Many rural communities rely on these converted lands for sustenance

and are fearful that harpy eagles prey on livestock, pets and young children. These fears lead local people to hunt harpy eagles and further exacerbate the population declines that have occurred due to habitat loss. The Peregrine Fund has developed a multi-faceted approach to stemming further decline of harpy eagle populations. Through educational programming, captive breeding and a re-introduction program the number of harpy eagles being hunted has decreased and extirpated populations of harpy eagles are being re-established. The continued loss of habitat remains a major threat to the recovery of large predators across Central and South America.

Mounting behavior and the estrous cycle in buffalo

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The role and importance of chemical signals in reproductive behavior have been well studied in several species of mammals. One of the striking examples is that the male identifies estrus through pheromonal signals and proceeds for mating, such as in buffaloes (*Bubalus bubalis* L., 1758). The most likely sources of such signals are urine and cervico mucus secretion. Mounting is another important behavior in a sexual context. Like flehmen, mounting is exhibited by more males than females. We allowed bulls in a female herd for 15 min. Bull mounting behavior was observed in response towards the six consecutive estrous cycles and recorded data after the bull mounted the back of female. The mounting activity exhibited by bulls was highly significant ($P < 0.01$) during estrus than during other phases. The average mounting activity of six bulls was 2.72 ± 0.17 included a maximum 3.0 ± 0.89 and a minimum 2.67 ± 0.82 in response to estrus phase. By contrast, in response to other stages the male showed a significant reduction (0.5 ± 0.55) in mounting behavior. When the bull was allowed to investigate the female, he first inhaled the pheromonal substances and exhibited flehmen behavior. We conclude

that bull mounting behavior under natural conditions in response to the estrous cycle of females is consistent with

the view that the presence of specific volatile compounds present in urine help the bull to detect estrus accurately.

Stream 4: Infectious diseases

H5N1 in wild birds in Siberia from 2005 to 2008

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The H5N1 influenza virus among wild and domestic birds was registered for the first time in Russia in 2005. All of the isolated influenza viruses were identified as the Qinghai-like HPAI H5N1 type and were brought by wild birds from China. The circulation of HPAI H5N1 was also found in Russia in again in 2006. The most interesting isolate was H5N1, isolated from clinically healthy common gull (*Larus canus*) that had IVPI equal to 1.7. The hemagglutinin cleavage site of this isolate was similar to those of HPAI. Studies carried out in 2007 showed that along with local outbreaks of HPAI H5N1 viruses and expansion of variety of virus subtypes observed. In 2008 the local outbreak caused by HPAI H5N1 was found in the Primorye region in Russia. In 2007 to 2008 we isolated two LPAI H5N2 viruses in wild ducks. The phylogenetic analysis of these isolates showed that they were genetically similar to the viruses circulated in China in 2006 and 2007. The isolation of these viruses indicates that the outbreak of 2005 was not caused by the sporadic accidental transmission introduced from China, but rather because of existing systematic virus transmission between these regions.

Population status, habitat use and health monitoring surveys of Tibetan antelope (*Pantholops hodgsoni*) in Ladakh, India

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Tibetan antelope (*Pantholops hodgsoni*) or Chiru, are endemic to Tibet, Xinjiang and Qinghai, China. Small populations of Chiru also wander into the extreme

northwestern parts of Ladakh, India and spend the summer months (from late May to early August) in remote upper Chang Chenmo valley in Changthang region and Daulat Beg Area (DBO) of Ladakh. Indirect and direct monitoring of the status, distribution, abundance, habitat use and health of Chiru was carried out over two months of Summer in 2004 and 2005. Data was also collected on daily activity patterns of Chiru and other mammals in relation to resource availability (food, shelter and disturbances) along trails, streams and contours. In 2004, a total of sixteen surveys were carried out across four survey blocks and 230 hours and 63 males were sighted (53 adults, nine young). In 2005, we located 60 individuals in the same area. Mean group size was 6.30 ± 4.82 (range 1 to 12). We sighted fewer animals in sandy plateau environments compared to riverine habitat but the presence of dung was consistent among these environments. We observed a population of 200 to 250 female Chiru near Daulat Beg and therefore estimate the Indian population to be between 250 and 310 animals.

Hidden threat of leptospirosis – an emerging zoonotic pathogen

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Leptospira spp., the causative agents of leptospirosis, adhere to components of the extracellular matrix, a pivotal step in colonization of host tissues during infection. Previously, we and others have shown that *Leptospira* Immunoglobulin-like proteins (Lig) of *Leptospira* spp. bind to Fn, laminin, collagen and fibrinogen. In this study, we report that *Leptospira* can be immobilized by human tropoelastin (HTE) or elastin from different tissues including lung, skin, and blood vessels, and that Lig proteins can bind to HTE or elastin. Moreover, both elastin and HTE bind to the same LigB immunoglobulin-like domains including LigBCon4, LigBCen7'-8, LigBCen9, and LigBCen12 as demonstrated by

ELISA and competition ELISA assays. The LigB immunoglobulin-like domain binds to the 17th to 27th exons of HTE (17-27HTE) as determined by ELISA (LigCon4, $K_D = 0.50\mu\text{M}$, LigBCen7'-8, $K_D = 0.82\mu\text{M}$; LigBCen9, $K_D = 1.54\mu\text{M}$; LigBCen12, $K_D = 0.73\mu\text{M}$). The interaction of LigCon4 and 17-27HTE was further confirmed by steady state fluorescence spectroscopy ($K_D = 0.49\mu\text{M}$) and ITC ($K_D = 0.54\mu\text{M}$). Furthermore, the binding was enthalpy-driven and affected by environmental pH, indicating it is a charge-charge interaction. The binding affinity of LigCon4D341N to 17-27HTE was 4.6-fold less than that of wild type LigCon4. In summary, we show that Lig proteins of *Leptospira* spp. interact with elastin and HTE and conclude this interaction may contribute to *Leptospira* adhesion to host tissues during infection.

Monitoring of infectious diseases among wild birds in Siberia

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When antiepidemic and antiepidemic measures are being taken, pathogen monitoring is a key task. As the World Health Organization announced that special attention should be given to the potentially pandemic H5N1 influenza virus, we began studying this pathogen in Russia and Commonwealth of Independent States. Since the spread of this virus is connected with wild birds, we have given special priority to collecting and studying material from them. Four among six main migration flyways go through Russia and the Commonwealth and we have been collecting samples in the territories of possible introduction of influenza viruses and in the territories of feasible viruses spreading by birds during Summer. From 2005 to 2008 the number of expeditions conducted by us increased considerably which led to an increase in collected and studied samples. In 2005 we collected 5658 samples, in 2006 4956, in 2007 9097 and in 2008 9333. In total we collected 29044 samples from wild birds and 9547 from poultry. As a result of testing, we isolated 177 influenza viruses: 107 were H5N1 influenza viruses and 70 viruses had other subtypes. Seventy-one viruses were isolated in 2005, 37 in 2006 and 2007, and 32 in 2008. To date, materials and highly qualified staff are available for the successful monitoring of avian influenza.

Surveillance for diseases in wildlife that pose a threat to agriculture or human health

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Some wildlife diseases are unique to only a single or a few species. Some diseases found in wildlife (e.g., bovine tuberculosis and highly pathogenic avian influenzas) are zoonotic with potential to infect humans. Others (e.g., chronic wasting disease) pose threats to domestic animal and livestock. Because disease risks are dynamic, the USDA/APHIS/Wildlife Services, National Wildlife Disease Program (NWDP) continually evaluates and alters its focus to address the most current challenges to wildlife, agriculture, and humans. This paper provides an overview of current perception of wildlife disease issues in the United States. Focus highlights aspects of highly pathogenic avian influenza, plague, rabies, chronic wasting disease, tularemia, west Nile virus, bovine tuberculosis and feral swine diseases. Diseases are briefly described along with potential risks and current activities implemented by USDA/APHIS/Wildlife Services. High path avian influenza remains an immediate focus of wildlife disease surveillance. Expanding range and population of feral swine render this species a continuing concern for animal health officials and wildlife managers. USDA/APHIS/ Wildlife Services continues effort to stop the spread of rabies in parts of the USA. Where human health risks exist, the NWDP conducts plague and tularemia surveillance. The NWDP also follows emerging diseases impacting wildlife, such as chronic wasting disease and bovine tuberculosis. Conducting disease surveillance increasing the likelihood emerging concerns are identified enabling livestock producers or human health officials to enhance bio security practices and wildlife managers can implement practices to reduce threats.

One health: merging wildlife, livestock, and human surveillance programs for emerging infectious diseases

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The past two decades have seen an increasing emergence and re-emergence of pathogenic infectious diseases, such as BSE, FMD, Q-Fever, bluetongue, Rift Valley Fever, Nipah virus, WNV, SARS, highly pathogenic H5N1 avian influenza, plague, and numerous others. The majority (approximately 60%) of these are caused by zoonotic pathogens transmitted along a continuum between wildlife, domestic animals, and human populations. Over 72% of these zoonotic diseases have emerged over the past two decades (52% from 1990 to 2000) and were caused by pathogens with a wildlife origin. These statistics epitomize the increasing and significant threat zoonotic diseases pose to global health, and the importance of understanding and managing such diseases in wildlife populations, as well as in humans and domestic animals. Although this One Health concept is not new, recent health emergencies are revitalizing the concept for application today. A One Health approach to managing infectious diseases aims to promote and implement meaningful collaboration and communication between veterinary medicine, human medicine, and multiple allied disciplines working locally, nationally, and globally to attain optimal health for people, animals, and our environment. Such cross-species disease surveillance, prevention, control, and mitigation efforts that recognize wildlife as a casualty of disease as well as a potential reservoir of disease should be emphasized.

Maternal food restriction and its impact on the development of F1 and F2 offspring

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Trans-generational effect and the adaptive mechanisms of maternal food restriction (FR) on reproductive capabilities, endocrine function, and brain and behavior development of offspring are basically unknown, especially in wild species. This study aimed to investigate the effects of maternal food restriction on the reproduction, survival, early development, endocrine function, immune function and odor attractiveness of adult females and F1 and F2 offspring in the greater long-tailed hamster (*Tscherskia triton*). Maternal females were 70% food restricted during their term of pregnancy. Results showed that maternal FR affected negatively the body growth of F1 offspring, and the physical and neurodevelopment of both F1 and F2 offspring. The effect of maternal FR on F2 offspring was smaller than that on F1 offspring. Maternal FR reduced the sizes of reproductive organs and hormone concentrations of both male and female F1 offspring. FR maternal females also produced significantly more male than female F1 offspring. FR increased adrenal gland mass and decreased spleen and thymus mass in adult females, but in F1 males, FR only decreased adrenal gland mass. FR affected the serum cortisol and antibody levels in adult females and offspring, but the responses to FR among adult females and F1 and F2 offspring were often opposite to those of the previous parental generations and showed gender-dependent variations. Our results indicated that maternal FR had complicate and profound consequences on endocrine function, immune function, and odor attractiveness across generations in the greater long-tailed hamster.

Outbreak of Visceral Leishmaniasis in Nepal from 2005 to 2008

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Visceral Leishmaniasis (VL) is a potentially fatal vector-borne zoonotic disease caused by a protozoan parasite, *Leishmania donovani*. Although VL is regarded as a significant health problem in Nepal by the Ministry of Health, there is no active case detection program in the country. It was observed that the first confirmed case of VL was recorded in 1980. By 2003, the disease has spread to twelve districts of central and eastern regions of Nepal, and

nearly six million people residing in these districts are at risk of acquiring this disease. A survey team from the National Zoonosis and Food Hygiene Research Center carried out epidemiological surveys to collect data from eastern Terai regional, sub-regional, zonal and district hospitals. During 2005 the total number of cases was 1230 across six districts of eastern Terai of Nepal which is a very high risk area in Nepal. Out of which cured cases were 1184 (96.3%), total death 46 (CFR 3.7%). During the year 2006 there were total VL cases 371 of which total cases cured were 334 (90%), out

of total death cases were 22 (CFR 5.9%). During the year 2007 total cases of VL were 315 of which cured cases were 279 (88.57%) and mortality cases were 36 (11.42%). The case fatality rate was 11.42% per 100 people. Areas of major concern within the health system are health policy, environment health, administration and management of health services, community health, individual and family health and direct health services.

Posters on display

Terrestrial mammals inhabiting the Jiuduansha Wetland Nature Reserve

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Jiuduansha Wetland Nature Reserve is the only estuary wetland in original condition found in the Yangtze River estuary. Previous assessments of the area have mainly focused on vegetation, birds and macrozoobenthos only. A detailed survey of mammals in the area was undertaken to confirm the distribution of terrestrial mammals, and yield information to assist conservation and management of the reserve. We conducted surveys from September 2008 to April 2009 using traps and clamps set along transects in three different areas of the reserve. Animal tracks and habitat characteristics were recorded at the same time. We found evidence of brown rats (*Rattus norvegicus*) and some hair and feces of some other small mammals, and bats (Vespertilionoidea). The emergence of the brown rat will likely influence biodiversity and natural succession in Jiuduansha. Now that this species has been detected inside the reserve further research is needed to clarify the distribution characteristics, diet and relationship with fauna and flora.

Conservation status of monkeys on Bioko Island, Equatorial Guinea

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For its small size, Bioko Island has an unexpectedly high level of biodiversity. This is especially true in the case of primates, with eleven species, some of which represent subspecies found only on Bioko. All the primates are nominally protected through the demarcation of two protected areas, Pico Basile National Park and the Gran

Caldera - Southern Highlands Scientific Reserve (GCSH). However, these primates continue to face serious threats from illegal shotgun hunting for sale and consumption as bush meat. The Bioko Biodiversity Protection Program has monitored the primate population within the GCSH through both annual research expeditions and monthly forest patrols. Along with natural terrain features, such as a nearly impassable coastline and a sheer crater rim along the Caldera, these populations have been offered at least a minimal level of protection from exploitation. Our data from these areas show that monkey populations, when offered some level of protection, can persist in light of intense hunting pressure. Concurrent bush meat research has shown a trend towards increasing prices, which over time, will render the deterrents to hunting in the GCSH insignificant.

Epidemiological surveillance of snakes and snakebite cases in Nepal

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There are two regions of the world where snakebites cause the most concern to public health: Southeast Asia and Africa. The incidence of snakebite varies from 300 to 500 bites per 100 000 people in forested regions to 50 to 100 bites per 100 000 in the Sahara. The exact incidence of snake bites in Nepal is unknown. I collected primary and secondary and information on snakebite cases recorded and reported by different media and hospitals from 2004 to 2007. This is the first time that an extensive study has been carried out on snakes in general and venomous snakebites in Nepal. The total number of snakebite cases was 3866; of this 117 cases resulted in mortality (mortality 3.03) and 1947 cases were cured (morbidity 50.36). The regions most affected were the Western (N = 1058) and Eastern (N = 1018) Development Regions. In conclusion, for the control of snakebite cases in humans one has to limit the blood flow from the bite site to the rest of the body while getting the patient to hospital; the victim should try to relax; remember what kind of snake it was; do not try to kill the snake; do not take the alive

snake to the hospital, and wash the bite site with clean water to avoid infection.

Expression of DNA damage response genes in a marine copepod exposed to UVB

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The marine harpacticoid copepod *Tigriopus japonicus* which mainly inhabits the rock pool of intertidal zone is frequently exposed to extreme ultra violet (UV) radiation. In particular, the UVB may induce detrimental effects such as lowering the survival, reproduction and development rates of the copepod. To evaluate the effect of UVB on *T. japonicus*, we conducted an acute toxicity test with an extensive UVB dose range (0-20 or 0-36 kJ/m²) in the condition of two UVB

intensities (50 and 100 μW/cm²) and also examined the expression pattern of DNA repair related genes. To investigate the gene expression pattern induced by UVB, we conducted real time RT-PCR with the copepod which was irradiated to UVB (19 kJ/m² at 50 μW/cm² intensity). When the copepod was exposed to UVB with the intensity of 50 and 100 μW/cm², levels of median lethal dose (LD₅₀) were 19 and 21 kJ/m², respectively. Expression patterns of DNA repair related genes were shown with significant modulations according to time course. Also we checked activated forms of p38 kinase and Jun N-terminal kinase (JUNK) to see what kind of signal transduction pathway would be involved in DNA damage by UVB. Upon DNA damage by UVB, those kinases were up-regulated with up-regulated expression patterns of relevant genes. Therefore, *T. japonicus* can be considered as a promising marine species for ecotoxicity testing and risk assessment of UVB in coastal marine environments. Recent advancement of research on environmental genomics of *T. japonicus* will also be highlighted in my talk.