Conference report

The joint scientific meeting of Amphibian and Reptile Conservation (ARC) and the British Herpetological Society took place in the lecture hall of the Bournemouth Natural Science Society on 5 December 2010, and encompassed the following contributions.

Population genetics of sand lizards, *Lacerta agilis*, **in Dorset**

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The sand lizard *Lacerta agilis* is a widely distributed reptile with a range that stretches from northwest Europe across Asia into Mongolia. It reaches the western edge of its range in the United Kingdom, where its specific habitat requirements have left it restricted to three separate geographical areas, the most significant populations occurring within heathland habitats in the southwestern county of Dorset. Lizards from these three areas have been shown previously to be genetically distinct from each other.

The current range of sand lizards in Dorset is crossed by six rivers of differing sizes, which may constitute a historical barrier to dispersal. Sand lizards from seven sites separated by rivers were sampled and genotyped at 15 microsatellite loci. $F_{\rm st}$ estimations indicated significant levels of genetic differentiation between sampling sites. Genotype data were analysed using a Bayesian approach (the STRUCTURE program), which infers population clusters based on genetic information alone; this identified six sand lizard populations across the Dorset range. The relationship between genetic differentiation and geographical distance, and other landscape features, between populations is investigated.

Frog diversity in Amazonian Peru

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The diversity of frogs in Peru is extraordinary but very poorly understood in terms of both ecology and conservation. This research was undertaken in the Pacaya-Samiria National Reserve in Peru during May–June 2009. The research aims were to discover what species were present at this site, and to determine their habitat use.

Eighteen days were spent in the field during which 104 transects, each measuring 100 metres, were undertaken on the river and on land. A total of 326 individual frogs, comprising 30 species from six families (Bufonidae, Dendrobatidae, Hylidae, Leptodactylidae, Microhylidae and Strabomantidae) were recorded in this period. This was compared with a previous study undertaken in 2003. Slightly more species were found in 2009 than in 2003. When comparing the number of individuals representing each family, however, there were declines in the number of bufonids and dendrobatids, and increases in the number of hylids and leptodactylids. Eighteen species were represented by less than ten individuals, with only two species,

Leptodactylus diedrus and *Sphaenorhynchus dorisae*, found in high abundances of more than 30 individuals. My study of the habitats used by these species highlights the importance of floating meadows, which were utilized by 12 different species.

Further research may include 1) longer-term studies to observe real declines in species abundances rather than general fluctuations; 2) the impacts of climate change on long-term population trends; and 3) evaluating whether tropical amphibian assemblages can act as surrogates for environmental change.

Assessment of an established population of atypical grass snakes, *Natrix natrix*, in the Aire Valley, UK

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During the past 20 years, an atypical population of grass snakes, Natrix natrix, has been observed within and around the Esholt Sewage Treatment (EST) facility, West Yorkshire, UK. Based solely on phenotypic appearance (the presence of dorso-lateral striping and a relatively indistinct collar), the snakes were suspected to originate from southern or eastern Europe. Principally, this study sought to ascertain the origin of the snakes through genetic means. Mitochondrial DNA samples, collected using buccal swabs, were extracted, amplified and sequenced. Sequences were then inserted into the existing natricine phylogeny, thus enabling inferences to be made about the origin of the snakes. An estimate of the population size (Nc) was calculated using a capture/mark/recapture (CMR) programme. Population estimates were generated using a Peterson-Schnabel maximum likelihood census model. Based on the output of this model, an effective population size (Ne) was also produced. Comparative morphometric studies investigated the possibility of discriminating between races using multivariate statistics (principal component analysis). Phylogenetic trees indicated that the Esholt population had originated from eastern Romania and was genetically distinct from native snakes. The CMR programme calculated an Nc of approximately 46 snakes (95% C.I. 28-106), although the actual value was expected to be higher. This figure translated to an Ne of 25, assuming constant sex ratio and excluding individuals of less than 300 mm SVL. No native snakes were encountered during the surveys and historical records for the area are unclear. Morphometric studies indicated that snakes could be discriminated based on a combination of morphometric measurements and phenotypic descriptions. Despite the fact that individuals from non-native populations were significantly

larger than native snakes, there were no significant differences in gape size (F=1.11, df=2, P=0.334), a principal determinant of prey choice. There are no physical barriers enclosing the EST facility and striped grass snakes have been reported in the surrounding areas. Future monitoring is recommended in order to ascertain population trends and range expansion.

The epidemiology of *Batrachochytrium dendrobatidis* **in the UK**

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Batrachochytrium dendrobatidis (Bd) is the causal agent of amphibian chytridiomycosis, a potentially lethal fungal disease that has been described as "the worst infectious disease ever recorded among vertebrates, in terms of the number of species impacted, and its propensity to drive them to extinction" (ACAP, 2005). The organism has achieved global distribution and has been implicated in the decline of more than 200 amphibian species. This project concerns the epidemiology of Bd in the UK. Where is it? What species are becoming infected? Can we identify risk factors that might be facilitating its spread?

The UK's first national Bd survey, which took place in 2008, has already allowed us to start unpicking these questions – a testament to the incredible dedication of volunteer surveyors up and down the country. We now know that Bd-infected sites can be found widely distributed across the UK. Moreover, it seems likely that all of our native amphibian species can become infected. Plans are now underway for the second UK Bd survey, due to take place in 2011.

Five-a-day for frogs: the influence of carotenoids on fitness related traits in anurans

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Carotenoids are yellow, orange and red pigments that are manufactured by plants, fungi and bacteria. Carotenoids are involved in vertebrate reproduction, antioxidant systems, immunity and coloration. Vertebrates are entirely reliant on dietary sources for these pigments. The role of carotenoids has been studied extensively in mammals, birds and fish, but their importance to amphibian health and reproduction has not been studied.

Amphibian captive breeding programmes are becoming increasingly important due to global population declines, but health and reproductive success are often poor in captive populations. Captive frogs often show reduced coloration, though it is unclear whether this is a result of limited carotenoid availability, and to what extent this would affect reproductive success and health. Our research aims to address these questions by examining delivery of carotenoids via different invertebrate species; the role of carotenoid-based coloration in the breeding behaviour of wild frogs; the effects of carotenoid supplementation on colour, health and reproductive success in captive frogs; and interactions between carotenoid availability and exposure to ultraviolet light. We have shown that carotenoid availability to captive frogs is greatly dependent on the diet fed to their invertebrate prey; however the carotenoid carrying capacity of commonly used invertebrates differs between species. Preliminary results show that carotenoid-based coloration may play a role in mating in wild nocturnal tree frogs, and that this coloration is greatly reduced in captive-bred individuals. Finally, we have found that administering carotenoids to captive frogs can significantly improve skin coloration, as well as some evidence that carotenoids may influence survival and growth during larval stages.

Pool frog re-introduction: an update

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The native status of the northern clade pool frog was recognized only as the last known population in England declined to extinction. Since then a reintroduction programme has been implemented. Between 2005 and 2008, 80 adult frogs, 88 juveniles and approximately 2,000 tadpoles have been translocated from Sweden to a site in Norfolk. The reintroduction has followed IUCN guidelines and includes monitoring pool frog numbers and their health. Amphibian species resident prior to the reintroduction have also been monitored, as have grass snakes, a potential predator of pool frogs.

There are initial indications of the success of the reintroduction. Frogs seem to grow well at the reintroduction site and are in good health, and tadpoles grow and metamorphose successfully. Peak counts of adult frogs have shown an increasing, although not consistent, trend, rising to counts of 31 adults. The peak count dropped in 2010, probably due to poor metamorph production in 2007 and 2008. High counts of metamorphs in 2009 and juvenile frogs in 2010 give prospects of greater recruitment into the breeding population in 2011. Further monitoring will be required before the long-term success of the reintroduction can be confidently determined.

Habitat restoration and management carried out for the reintroduction has greatly increased the ecological value of the receptor site, demonstrating biodiversity gain driven by a species-focused project. The pool frog reintroduction is a joint project involving Amphibian and Reptile Conservation, Natural England, Anglian Water, the Forestry Commission, Norfolk Wildlife Trust and the Universities of Greenwich, Kent and Sussex. Health screening of amphibians has been carried out by the Institute of Zoology.

Evolutionary processes in a recent colonizer: the case of the UK wall lizard (*Podarcis muralis*)

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Invasive species attract much attention, not only because of their conservation concern but also because the novel conditions that introduced organisms face can result in extremely rapid evolution. I study how the UK wall lizard (*Podarcis muralis*), which consists of two subspecies that hailed from France and Italy between 30 and 150 years ago, has evolved to cope with the UK environment. My research questions range from how the cooler UK climate affects mating systems and egg incubation in these lizards, to the interactions between the two subspecies in their artificial "hybrid zones". Whilst this is a brand new study system, preliminary results suggest that this species can provide an invaluable resource for studying contemporary evolutionary processes, and may provide an important resource for how invasive species can adapt (or not) to their new environments.

Does *Batrachochytrium dendrobatidis* **have an effect on natterjack toads in the UK?**

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The aim of this project is to establish the scale of the threat posed by *Batrachochytrium dendrobatidis* (Bd) to the UK's natterjack toad (*Bufo calamita*) populations. Bd is a fungal pathogen that infects many amphibian species and can result in the disease chytridiomycosis. Little is known about the effect of Bd on natterjack toads. The scale of the threat is being assessed by a combination of fieldwork and laboratory studies. The principal fieldwork component is intensive monitoring of natterjack populations in north Cumbria, where Bd infection has been recorded at several natterjack sites since 2005.

Two seasons of fieldwork have now been completed, including mark-recapture studies of adult natterjacks, with repeat Bd sampling of individuals from April to September. Data collected over three breeding seasons will be used to generate comparative estimates of survival for infected versus uninfected adult toads, using appropriate models. Juvenile stages have also been sampled to assess disease prevalence and seasonal changes in infection status. Semi-captive and laboratory experiments are being used to try and answer some of the questions raised by the patterns of infection observed in the wild.

Reconstructing an African viper radiation: phylogeny and historical biogeography of the genus *Bitis*

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The advent of DNA technology has provided the exciting opportunity to reconstruct the evolutionary history of related species using empirical methods. Despite this potential, however, studies of widespread African reptile lineages remain scarce. Our work focuses on a well known group of venomous snakes, the genus Bitis, which includes iconic species such as the puff adder and gaboon viper. With seventeen currently recognized species distributed throughout sub-Saharan Africa in addition to Arabia and Morocco, Bitis is Africa's most geographically widespread and species-rich genus of viperid snakes - but this only hints at the diversity present. Bitis also contains both the largest and smallest representatives of the sub-family Viperinae, and the variety of habitats occupied by Bitis species ranges from the arid sands of the Namib Desert to the tropical forests of west and central Africa.

Our analyses reveal Bitis to be an ancient genus, originating from a single common ancestor that existed around 28 million years ago. Extant species are divided among five major genetic clades that originated during a warm and climatically stable period from the late Oligocene to early Miocene. Nevertheless, a period of global cooling starting in the mid-Miocene is associated with parallel radiations of forest- and open-habitat living Bitis clades that generated most of the taxonomic diversity found today. Within the southern African dwarf Bitis, we have recovered unexpected relationships suggesting the presence of cryptic species and highlighting the need for future taxonomic revisions. We also reveal the existence of multiple divergent genetic lineages within the widespread and medically significant puff adder, Bitis arietans, which diverged shortly after a major expansion of grassland habitats in Africa.