

XTH YOUNG SYSTEMATISTS' FORUM

2nd December 2008, Flett Theatre, Natural History Museum, London, UK

Programme

09.00	Registration	
09.30	Welcome	Juliet Brodie
09.35	Opening comments	Tim Barraclough (Imperial College)
09.50	Andrea Crowther	Taxonomic uncertainty in the sea anemone genus <i>Lebrunia</i>
10.10	Ellen Cocquyt	Phylogenetic relationships of the green algae refined: new insights from a combined analysis of nuclear and plastid genes
10.30	Magdalena Zarowiecki	Investigating evidence that the mosquito <i>An. vagus</i> is a cryptic species complex
10.50	Sognigbé N'Danikou	Phenetic analysis of Wild populations of <i>Momordica charantia</i> L. (Cucurbitaceae) in West Africa and inference of the definition of the new subspecies <i>M. c. macroloba</i> Achigan-Dako & Blattner
11.10	Coffee and posters	
11.40	Therese Holton	Phylogenomic reconstruction of the Metazoa
12.00	Lucy Bittner	Unravelling diversity of crustose (non-geniculate) coralline algae of the South Pacific
12.20	John Wilson	Comparison of DNA barcode readers for the identifications of butterflies
12.40	Alba Vergés	The genus <i>Kallymenia</i> (Kallymeniaceae, Rhodophyta) from the north-eastern Atlantic
13.00	Lunch and posters	
14.00	Claire Standley	On the trail of a snail: elucidating the species complexes of East African <i>Biomphalaria</i>
14.20	Mine Kocyigit	A phylogenetic analysis of <i>Allium</i> sect. <i>Codonoprasum</i> (Alliaceae) using TRNL-F plastid and its nuclear DNA sequences
14.40	Rachel Walker	The evolution and development of specialised floral traits in the South African 'beetle daisy' <i>Gorteria diffusa</i> Thunb.
15.00	Romina Novo Henriques	The influence of the Benguela cold current system in the genetic sub-structure of four fish species
15.20	Tea and Posters	
15.50	Hanno Schaefer	Systematics of <i>Cucumis</i> (Cucurbitaceae) and the origin of melon and cucumber
16.10	Katie Davis	A species-level supertree of birds and the future of supertree construction
16.30	Noemí Sánchez	Critical taxonomic features of the genus <i>Gloiocladia</i> (Faucheaceae, Rhodophyta)
16.50	Claudia Ciotir	DNA barcoding outcomes from testing four plastid regions in <i>Cyclamen</i> L. (Myrsinaceae)
17.10	Closing comments / presentation of prizes	Peter Olson / Eileen Cox (NHM Botany)
17.25	Reception and Posters	
-19.00		

Organized by **Juliet Brodie** (Dept Botany, NHM) and **Peter Olson** (Dept Zoology, NHM) with support from:



ABSTRACTS

ORAL PRESENTATIONS

Taxonomic uncertainty in the sea anemone genus *Lebrunia*

Andrea L. Crowther

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In current classification, the sea anemone genus *Lebrunia* (family Aliciidae) has two valid species: *L. danae* (Duchassaing and Michelotti, 1860) and *L. coralligens* (Wilson, 1890). Most specimens of *L. danae* are larger than specimens of *L. coralligens*, so it has been hypothesized that the name *L. coralligens* has been applied to juvenile specimens of *L. danae*. In order to evaluate if there are one or two species, I studied multiple specimens of *Lebrunia* ranging in size. I tested whether there was any relationship between pedal disc diameter and number of branching orders of the pseudotentacles. In the 48 individuals investigated, there was a clear absence of intermediate forms with pseudotentacles that branched four times. This supports the current classification of separate species: specimens of *L. danae* possessed pseudotentacles that branched at least five times, while those of *L. coralligens* possessed pseudotentacles that branched three times at most.

Phylogenetic relationships of the green algae refined: new insights from a combined analysis of nuclear and plastid genes

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Green algae exhibit a remarkable cytological diversity ranging from unicellular microscopic algae with a single nucleus, over multicellular filaments and foliose blades, to coenocytic and siphonous life forms that are essentially composed of giant cells containing thousands of nuclei. Classification of this diverse and evolutionary ancient group is tempting and requires the analysis of additional nuclear markers since classic ribosomal RNA and plastid genes failed to resolve relationships among the major groups. Suitable nuclear markers were designed by searching the literature, using genomic sequences of *Chlamydomonas reinhardtii* and *Ostreococcus tauri* and making an EST library for *Cladophora coelothrix*. Seven nuclear genes were successfully amplified and analysed in a Bayesian and maximum likelihood framework together with traditional markers. Some features of the amplified genes provide additional support for certain relationships: usage of a non-canonical code in some groups and gain-loss dynamics of two non-overlapping elongation factor genes (EF-1 α and EFL).

The mosquito *Anopheles subpictus* species complex- How many species are there in South East Asia?

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The purpose of this research is to study the evolutionary history of the mosquito species complex *An. subpictus/An. indefinitus*. *An. subpictus* s.l. is a major vector of malaria in South East Asia, and like the closely related *An. gambiae*, it is a species complex. The species complex consists of two informal, but well studied species from India, and two described, but less well known, species from South East Asia. We have used DNA sequences for cytochrome c oxidase 1 (CO1) and internal transcribed spacer 2 (ITS2) from >200 specimens from 15 populations throughout the species range, and the results confirm that *An. subpictus* is a species complex consisting of at least 6 species, some of which are sympatric throughout a large part of their range. Furthermore, *An. subpictus* shares CO1 haplotypes with another major vector of malaria, which could lead to major mistakes if these species were to be identified using standard molecular barcodes.

Phenetic analysis of Wild populations of *Momordica charantia* L. (*Cucurbitaceae*) in West Africa and inference of the definition of the new subspecies *macroloba* Achigan-Dako & Blattner.

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Momordica charantia is a paleotropical species used as medicinal plant or vegetable in West Africa. This paper examines the morphological variation within some wild populations of *M. charantia* in West African phytogeographical regions. The results point out that ecological conditions affect leaf size in that species, as well as the size of male and female bracts and peduncle lengths. However, characters linked to the fruit size are uniform across regions except for pericarp thickness and the tubercle height. The location of the bract on the peduncle is independent of environmental conditions. Multivariate analyses clarify also the long-standing ambiguity for the identification of *M. charantia* observed in Benin in clear discriminating two taxonomical groups on the leaf lobe characters, the common subspecies *charantia* and the new subspecies *macroloba* Achigan-Dako & Blattner endemic to the Dahomey gap and the Sudano-Guinean phytoregion of Benin and Togo.

Phylogenomic reconstruction of the Metazoa

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Despite extensive study, the phylogeny of the Metazoa remains largely unresolved and intensely debated. Central to this dispute is the phylogenetic position of nematodes. Traditionally it was thought that animals advanced through successive increments of

complexity, implying a basal placement of nematodes: the Coelomata hypothesis. However, this view of bilaterian evolution was contested by a study of 18S rRNA genes, which radically proposed grouping nematodes and arthropods, in a clade of moulting animals known as Ecdysozoa. This 'new animal phylogeny' fails to attain unanimous support from phylogenetic analyses. Moreover a phylogenomic scale study has yet to advocate the Ecdysozoa topology. Failure of phylogenomic analyses to recover Ecdysozoa has been attributed to systematic error, which is often compounded further by sparse taxon sampling. In light of this, we have conducted a genome wide analysis of the animal phylogeny, investigating to what extent outgroup choice and taxon sampling influence the resultant topology.

Unravelling diversity of crustose (non geniculate) coralline algae of the South Pacific Ocean

Lucy Bittner

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In molecular assisted taxonomy, gene trees are used to infer species boundaries by setting a genetic threshold to delimit species. Applying this rule to two large datasets of DNA sequences (one mitochondrial and one plastid marker), we assessed the biodiversity of a species-rich red algal order, the Corallinales, with a sampling effort mostly focussed on the South Pacific area (Vanuatu, Fidji, New Caledonia). In these tropical regions, crustose species of Corallinales are profuse and constitute conspicuous, dominant populations. Therefore, they are a key-component of coral-reef ecosystem, the most species-rich marine-ecosystem on the planet. In the present study, we tested further our taxonomic hypothesis inferred from genotypic clustering by confronting our results with ecological data (e.g. habitat, bathymetry). Finally, we discussed the contribution of our large dataset to the understanding of the evolutionary story of this red algal lineage and to the inventory of marine biodiversity of the South Pacific Ocean.

Comparison of DNA barcode readers for the identifications of butterflies

John James Wilson

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A major problem for DNA-based species identification is determining whether the differences between a DNA sequence from an unknown specimen and the reference database sequences are intraspecific variation or whether they signal interspecific relationships. The solution is an accurate DNA barcode reader. I compared the ability of six different DNA barcode readers to accurately identify unknown sequences to species present in a reference database or alternatively to determine when the species of the unknown was not present. The results suggest that currently no algorithm is particularly better than any other. The choice of reader presents a compromise between obtaining names for most of your unknown organisms and a higher risk that some names will be incorrect, or the unsatisfactory situation of obtaining fewer names but a lower risk that these few names will be false.

The genus *Kallymenia* (Kallymeniaceae, Rhodophyta) from the north-eastern Atlantic

Alba Vergés

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The genus *Kallymenia* J. Agardh currently includes a single species in the Atlantic European shores: *K. reniformis* (Turner) J. Agardh. After a revision of numerous specimens identified as *Kallymenia reniformis*, *Kallymenia* sp. or *Meredithia microphylla* (J. Agardh) J. Agardh in several herbaria (BCF, GALW, HGI, L, MA and SANT), we found distinctive vegetative and reproductive characteristics that led us to believe that a new *Kallymenia* is present in this zone. The new species is characterized by a short, pink to red stipitate and a deeply lacerated frond, with a dentate margin. The five cortical cell layers diminish in size outwards, with the inner cortical cells measuring up to 110 μm in diameter. The medulla is lax and composed of some scattered filaments and highly refractive stellate medullary cells with arms up to 1000 μm in length. The species is non procarpic, with monocarpogonial cell systems composed of a supporting cell, a three-celled carpogonial branch and up to eight subsidiary cells. After presumed fertilization, a fusion cell is formed from where numerous connecting filaments emerge and fuse with the supporting cell of an auxiliary cell system, which comprises the auxiliary cell and up to six subsidiary cells. The gonimoblasts develop on the connecting filament and carpospores are liberated through a pore. Tetrasporangia are scattered in the outer cortex and cruciately divided. Rejecting the hypothesis of an introduced species and comparing this species with the rest of monocarpogonial *Kallymenia* we observed that it possess exclusive anatomical and reproductive characters that confirms the new taxon. At present, we are working on molecular data to fully confirm its identity.

On the trail of a snail: Elucidating the species complexes of East African *Biomphalaria*

Claire Standley

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Snails of the genus *Biomphalaria* are small, freshwater pulmonate gastropods which are found mainly in tropical Africa and South America. They are of biomedical importance as the intermediate host of *Schistosoma mansoni*, a trematode that causes intestinal schistosomiasis in humans. Some species of *Biomphalaria* appear to transmit schistosomiasis more readily than others while there are also some appearing to be completely refractory. This complex pattern of parasite transmission has generated much interest in methods for discerning between species and populations - previous morphological techniques have proven unreliable or inadequate, and current research is focused on molecular markers which bring new insight into taxonomic structure in *Biomphalaria*. This information is particularly relevant in highly endemic areas, where national control programmes require up-to-date data on the distribution of highly susceptible snail populations and at-risk human communities. For example, the shoreline environment of Lake Victoria is not only rife with schistosomiasis, but its huge size and relatively young age provide an interesting natural laboratory for studying the evolution and dispersal of organisms within it. Here, the preliminary results of analysis of the 16S region of individuals from 81 sites along the Lake Victoria shoreline of Uganda and Tanzania are presented, demonstrating once more the morphological ambiguity of species, and populations thereof, of *Biomphalaria* in the region, as well as their relatively recent evolutionary history in the region.

A phylogenetic analysis of *Allium* sect. *Codonoprasum* (Alliaceae) using *TRNL-F* plastid and its nuclear DNA sequences

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Allium L. is the largest genus of petaloid monocotyledons, comprising about 750 species naturally distributed only in the Northern Hemisphere, mainly in seasonally dry regions. The genus is characterized by bulbs enclosed in membranous tunics, free or almost free tepals, and often a gynobasic style. The genus *Allium* is represented in Turkey by 174 species divided into 14 sections. Section *Codonoprasum*, the largest section in Turkey, consists of 44 taxa of which 18 are endemics to the country. In this study, we investigate the phylogenetic relationships within this section using 23 taxa (including nine endemics) and DNA sequence data from the plastid *trnL-F* region and the nuclear internal transcribed spacers (ITS). We examine the resulting phylogenetic tree in light of morphological characters. Results of this study will be important to understand the evolution of this particular group and help prioritize conservation effort of these attractive plants in one of Earth's biodiversity hotspot.

Title: The evolution and development of specialised floral traits in the South African 'beetle daisy' *Gorteria diffusa* Thunb.

Rachel Walker¹, Dr. Beverley Glover¹ and Dr Paula Rudall²

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The relationship between floral traits such as pigmentation and the behaviour of pollinators is an important factor in the co-evolution of plants and their pollinators. Genetic studies of model plants such as *Arabidopsis Heynh.* and *Antirrhinum L.* have provided an understanding of the developmental evolution of floral organs. A model species to study more specialised floral traits is *Gorteria diffusa* Thunb. (Asteraceae) - the South African 'beetle daisy'. The bee-fly *Megapalpus nitidus* pollinates *G. diffusa* and is attracted to the 3-dimensional insect-mimicking spotted florets, which are composed of three cell types and a concentrated deposition of anthocyanin. These structures represent a floral adaptation that itself could be a major factor in the extensive radiation and diversification of the angiosperms. The genes encoding the regulators of the anthocyanin pathway in the beetle spot will be identified and by analysing the phylogenetic relationships of the morphotypes of *G. diffusa*, an overall understanding of the evolutionary history of this species will be achieved.

The influence of the Benguela cold current system in the genetic sub-structure of four fish species

Romina Novo Henriques, I Barnes, and P Shaw

Royal Holloway

The semi-perennial upwelling cell in the centre of the Benguela Cold Current System off SW Africa has been suggested to represent a barrier to the movement of pelagic eggs and larvae and thus may promote genetic divergence by limiting gene flow. To test this hypothesis, patterns of genetic variation across the system were studied for three fish species (leerfish, blacktail and geelbeck) and one fish species complex (kob). The kob complex is composed of three species, with their distributions confined to either side of the upwelling cell. However, leerfish, blacktail and geelbeck do not exhibit such an evident

taxonomic break, being considered single entities across the Benguela Current region. Preliminary results based on mitochondrial DNA data suggest that this 6.5 million year-old physical oceanographic boundary is reflected in the genetic structure of all taxa. The observed genetic divergence between northern and southern populations for the three cosmopolitan species is similar to that observed in the species complex, thus implying the need for a review of systematic status.

Systematics of *Cucumis* (Cucurbitaceae) and the origin of melon and cucumber

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Melon, *Cucumis melo*, and cucumber, *C. sativus*, are among the world's most important crops. Traditionally, *Cucumis* has been thought to be African, with only two Asian species. Ongoing breeding efforts are all based on this circumscription. Using chloroplast genes, spacers, and introns plus nuclear ITS sequences for a dense species sample, we tested the monophyly and position of the genus and discovered that *Cucumis* must include five smaller genera to become monophyletic. Unexpectedly, cucumber is nested in an Australasian clade, with melon sister to this clade rather than being close to African species. The closest relative of the genus is *Muellerargia*, with one Australasian and one Madagascan species. Molecular clock dating and DIVA analyses suggest an Eocene origin of *Cucumis* in Southeast Asia; *C. sativus* apparently diverged 3 (6-1) million years ago.

A species-level supertree of birds and the future of supertree construction

Katie E. Davis and Rod D. M. Page

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Supertrees are a useful method of constructing large-scale phylogenies by assembling numerous smaller phylogenies that have some, but not necessarily all, taxa in common. Supertrees have now been produced for a diverse range of taxa including dinosaurs, mammals and crocodiles. Birds are an obvious candidate for supertree construction as they are the most abundant land vertebrate on the planet and no comprehensive phylogeny of both extinct and extant species currently exists. Here, a species-level supertree has been constructed containing 5274 taxa from 757 source trees. The tree shows generally accepted relationships between the main groups, with only a few novel clades, most of which can be explained by a lack of information regarding those taxa. The tree was constructed using a strict protocol, which ensures robust, accurate and efficient data collection and processing. The resulting supertree provides an insight into the challenges and pitfalls of constructing ever-larger supertrees.

Critical taxonomic features of the genus *Gloiocladia* (Faucheaceae, Rhodophyta)

Noemí Sánchez

University of Girona, Faculty of Sciences

Within the family Faucheaceae (Rhodymeniales, Rhodophyta) *Gloiocladia* J. Agardh is the genus more expanded, at around 30 species. Currently, in the Eastern European shores, there are three *Gloiocladia* species: *Gloiocladia furcata* (C. Agardh) J. Agardh, the generitype, *G. repens* (C. Agardh) Sánchez et Rodríguez-Prieto and *G. microspora* (Bornet ex J.J. Rodríguez) Sánchez et Rodríguez-Prieto, both ancient species of *Fauchea* Bory de Saint-Vincent et Montagne in Montagne. In this work, we want to remark morphological similarities between the three species, both at vegetative and reproductive level, trying to summarize the results obtained during detailed studies conducted in the Mediterranean and Eastern Atlantic Ocean (Sánchez & Rodríguez-Prieto, 2005; Rodríguez-Prieto *et al.*, 2007 and non published data). They are characterized by a much branched and somewhat dichotomous thallus, an outer cortex composed by filaments arising from a subcortex that forms a network parallel to the thallus surface and medullar cells which are elongated in longitudinal section. The three species are procarpic with 3-celled carpogonial branch and 2-celled auxiliary branch growing on a cortical supporting cell. Nutritive cells and the *tela arachnoidea* are present since early postfertilization stages and fusion cell is formed through coalescence of the auxiliary branch, the fused cells of the carpogonial branch and the surrounding cells; supporting cell can participate in the fusion process. Constituents of the fusion cell are indiscernible when the structure is mature and the gonimoblast arises of the primary gonimoblast cell. Prominent and ostiolated cystocarps are situated on the branch margin or around the thallus. Tetrasporangia develop in nemathecia, they are subapical, cruciately, decussately or irregularly divided and they connect basally with the nemathecial filament. Although at that time the knowledge about this genus has improved there are still many poor known species that require further studies.

DNA barcoding outcomes from testing four plastid regions in *Cyclamen* L. (Myrsinaceae)

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The goal of this study is to determine the efficiency of four potential barcoding regions *matK*, *trnH-psbA*, *atpH-atpF* and *psbK-psbI* for 22 species of *Cyclamen*. *Cyclamen* is an interesting genus to test for barcodes. Although is a relatively old genus it includes a much younger species complex where phylogenetic relationships were difficult to reconstruct. One of the main points of interest is to see whether DNA barcodes reveal a correlation between genetic distances, sequence identification and species delimitation. Previous studies have identified *matK* and *trnH-psbA* as the potentially useful barcoding regions. Our analysis found the lowest interspecific distances for *matK* and *atpH-atpF*, and the highest for *psbK-psbI* and *trnH-psbA*. *psbK-psbI*, *trnH-psbA* permit identification of more than 85% of the sequences. Combining regions together increases the barcoding 'gap' in *Cyclamen* permitting more efficient identification. Failure of identification occurred for *C. repandum*, *C. peloponnesiacum*, *C. balearicum* and *C. creticum* species. This group contains relatively young lineages and previous phylogenetic reconstruction has proven to be uncertain and difficult in resolving their relationships. DNA barcodes show considerable promise for *Cyclamen* and could help clarify the taxonomic boundaries at the infra and interspecific level.

POSTER ABSTRACTS

The molecular hurdle in the phylum Nematoda

Holly Bik

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Molecular data has shed much light on the deep phylogenetic relationships amongst nematodes, necessitating a complete restructuring of previous taxonomic frameworks. However, our overall knowledge of genetic diversity remains limited because of the difficulty in obtaining reliable sequence data from individual nematodes. After repeated unsuccessful attempts to collect data from deep-sea specimens, a series of methodological time series experiments were conducted to determine the root cause of molecular problems. Results clearly indicated the rapid degradation of DNA in worms prepared using traditional taxonomic protocols; this new data will be used to develop protocols specific for integrative morphological/molecular approaches. These experiments form part of a PhD project which aims to resolve evolutionary relationships within the Order Enoplida, a basal nematode clade.

Mitochondrial genome sequence and gene order of *Sipunculus nudus* give additional support for an inclusion of Sipuncula into Annelida

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Sipuncula are unsegmented marine worms, traditionally placed in their own phylum. Their position within the metazoan tree is in current debate – recent molecular and morphological studies suggest a close relationship to annelids. Since analyses of mitochondrial genomes have proven to provide valuable markers for pursuing phylogenetic questions, we studied the mitochondrial genome of the sipunculid *Sipunculus nudus* (GenBank: FJ422961), using both, amino acid sequence information and gene arrangements, as characters for systematic analyses. The mitochondrial gene order found in *Sipunculus nudus* resembles that of annelids, but shows also several derivations so far specific for sipunculids. According to the most parsimonious explanation, this gene order can be derived from the hypothetical annelid ground pattern, thus supporting a close relationship of Annelida and Sipuncula. Our sequence based phylogenetic analysis (ML) of 11 mitochondrial protein-coding genes results in significant bootstrap support for Annelida sensu lato, combining Annelida with Sipuncula, Echiura, Pogonophora and Myzostomida. Hence, sipunculids should be regarded as derived annelids, even though they show no external sign of segmentation. Supported by DFG Ba1520/10-1,2; DFG Li 998/3-1; DFG Bl 787/2-1 (all from priority programme 1174 "Deep Metazoan Phylogeny")

An integrative approach to resolving taxonomic controversy and reconstructing the evolutionary history of *Rana sphenocephala*

Catherine E. Newman and Leslie J. Rissler

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Phylogeographic studies have shown that species with broad geographic ranges are often composed of multiple genetic lineages. *Rana sphenocephala* is widely distributed in the United States and has a complicated taxonomic history. Two subspecies are currently recognized: *R. s. sphenocephala*, found only in Florida, and *R. s. utricularia*. We aim to reconstruct the evolutionary history of *R. sphenocephala* and resolve controversy over the taxonomic status of *R. s. sphenocephala* by employing an integrative approach that includes genetic and morphological analyses and ecological niche modeling. Preliminary results show little genetic differentiation between *R. s. sphenocephala* and *R. s. utricularia*. Instead, phylogenetic analyses of the ND1 mitochondrial gene reveal an eastern lineage and a western lineage with a 4.36% sequence divergence, and niche modeling results suggest lineage divergence may have been driven by postglacial expansion from refugia. Further analyses are currently being conducted to determine the appropriate taxonomic status of these two lineages.

Molecular phylogeny of *Ariocarpus* Scheidweiler (Cactaceae) and its distribution along the Chihuahuan Desert.

Victor M Rodríguez-G., Chris Yesson and Julie Hawkins

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The genus *Ariocarpus* is one of the most charismatic cacti in the world. The group contains six or seven species according to different authors. All of the species are endemic to the Chihuahuan Desert and some species can be widely spread in the region while others are restricted to few localities. We sequenced the chloroplast region *trnK/matK* for all the species of *Ariocarpus* and we present this phylogeny in relation to the specific distribution along the Chihuahuan Desert. The parsimony analysis supports *Ariocarpus* as a monophyletic genus. The spatial distribution analysis shows that the majority of the species are allopatric. Geographically isolated species conform usually independent clades, suggesting an initial congruence between physical distribution and molecular phylogeny.

Using mitogenomics to elucidate a higher level spider phylogeny

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Mitochondrial DNA is firmly embedded in contemporary molecular systematics, but more accurate phylogenetic reconstruction is often impeded by low sequence volume and number of loci. Advances in sequencing technology (Massively Parallel Sequencing – MPS) may resolve this issue, offering the potential of amplifying, sequencing and analysing multiple, whole mitochondrial genomes to resolve complex phylogenetic relationships. The Araneae (spiders) are a diverse order containing over 40,000 described species distributed among 109 families. However, spiders have been relatively understudied from a higher-level

molecular systematic perspective, and very little is known about inter-family relationships. My research is focused on resolving these relationships using mitochondrial phylogenomics to better understand the evolutionary mechanisms underlying speciation and diversification.

Tree Independent Identification of Evolutionary Rates to Overcome Systematic Errors

Carla A. Cummins and James O. McInerney

NUI Maynooth, Co. Kildare, Ireland.

Removal of fast evolving sites in an alignment has long been used in an attempt to enhance phylogenetic signal in data (Rodriguez-Ezpeleta et al., 2007) and to overcome such problems as long-branch attraction (LBA). Many approaches have been made to identify fast-evolving sites, many tree based. Tree based methods, however, often lead to systematic bias towards the given tree, whether correct or incorrect. Our new software - Tree Independent Generation of Evolutionary Rates, or TIGER, overcomes these systematic errors using simple analysis of split support and split conflict (Wilkinson, 1998) to detect varying degrees of compatibility amongst the data. Analysis of both a ribosomal RNA dataset that is known to manifest extreme compositional bias and a dataset that has been used in order to infer the ancestry of the mitochondrion endosymbiont demonstrates how systematic biases can occur when identifying rapidly evolving sites. Our approach can clearly identify rapidly-evolving homoplastic sites and improve the chances of recovering the correct phylogenetic tree.

The search for a robust phylogeny of malacostracan crustaceans; evaluating the utility of new nuclear protein-coding genes

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Despite over a century of research into the systematics of the Malacostraca a higher-level phylogeny remains intensely controversial. In this study, species have been selected to span the diversity of the group, encompass rare species, and mirror sampling in morphological studies to facilitate comparison/combination. Sequencing efforts focus on a group of hitherto minimally sequenced, broadly conserved, nuclear protein-coding genes, which have been shown to be informative for high-level phylogenetics. The amplification protocol for these genes has been modified for use with genomic DNA rather than cDNA. Here we present the results of two of these markers: Alanine-tRNA synthetase and Glutamyl & prolyl-tRNA synthetase. We find that these genes are phylogenetically informative for unravelling the history of this ancient group. I present results for both loci separately and in combination, and compare these with previous molecular and morphological hypotheses/efforts.

Macroevolutionary trends within Metriorhynchidae (Crocodylia): an integrated quantitative approach

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Metriorhynchid crocodylians represent the pinnacle of marine specialisation within Archosauria; not only were they a major component of the Middle Jurassic to Early Cretaceous marine ecosystems, but provide further examples that extinct crocodylians did not all resemble their modern extant relatives. Here a varied toolkit of techniques, including cladistics, geometric morphometrics, discrete character disparity analysis and biomechanical finite element analysis, is used to examine the macroevolutionary history of this clade. All analyses demonstrate that metriorhynchids became more divergent in terms of biodiversity, form and function up until the Jurassic-Cretaceous boundary, after which there is no evidence for recovery or further radiations. A clear evolutionary trend towards hypercarnivory in *Dakosaurus* is supported by phylogenetic character optimisation, morphometrics and FEA, which also support specialised piscivory within *Rhacheosaurus* and *Cricosaurus*. Within Metriorhynchidae there is a consistent trend towards increasing marine specialisation, with the hypermarine *Cricosaurus* exhibiting numerous convergences with other Mesozoic marine reptiles.

Investigating the mechanism of papillomavirus-host association

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The dynamics of virus-host co-evolution is important in understanding pathogenic evolution, modelling viral evolutionary patterns, and identifying emerging pathogenic threats. Some viruses (such as HIV and influenza) have undergone numerous shifts of host while others, such as the slowly-evolving papillomavirus, are believed to shift hosts rarely or not at all, specialising in different hosts only as these hosts speciate. We observe incongruities between the papillomavirus and host phylogenetic trees, suggesting the possibility of host shift. These incongruities, however, can also be explained by viral divergence prior to host speciation followed by segregation along the different host lineages. We have examined the fit of host divergence times on the papillomavirus phylogeny to determine between different scenarios of host-virus association. We find that the most likely scenario involved multiple ancient mammalian papillomavirus lineages which co-speciated with their hosts. This suggests that papillomaviruses may be far more diverse and widespread than currently known.

Dating the origin of the 1918 'Spanish flu' pandemic virus

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The devastating 'Spanish flu' of 1918 killed an estimated 50 million people worldwide, ranking it as the deadliest pandemic in recorded human history. Analysis of the genome sequence of the 1918 influenza virus suggested that the virus originated from an avian source. The proposed avian origin of the virus has been the subject of a heated debate, with no clear resolution about how and when it entered the human population. We use maximum likelihood estimation and a non-homogeneous model of sequence evolution that takes into account differences in the substitution patterns between nucleotide sequences of viruses in avian and mammalian hosts to date the host transfer event that led to the

formation of the pandemic virus. We show that the eight RNA segments that became the 1918 viral genome were introduced into a mammalian host around 1899 (1887-1914). This virus split into swine and human H1N1 lineages around 1913 (1912-1914). This indicates that the virus that led to the 1918 flu was already in the human population at least between 2-5 years before the actual pandemic. Although a single whole virus transfer from an avian to a mammalian host leading to a stable viral lineage in humans is the most parsimonious explanation for the origin of the 1918 virus, successive reassortment events involving avian-like segments prior to the formation of the 1918 virus cannot be ruled out. Our results also suggest that the virus underwent an unexpectedly long period of adaptation in a mammalian host. We suggest this mammal might have been swine, where the virus evolved for several years before being introduced into the human population, causing the pandemic a few years later. These results have important implications for assessing the likelihood of a future bird flu pandemic in humans.

Infrageneric relationships and character evolution of SE Asian *Begonia*

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The pantropical genus *Begonia* (>1550 spp.) has a centre of diversity in SE Asia (>520 spp.). Infrageneric relationships of SE Asian *Begonia* are only poorly understood and the circumscription of some sections remains doubtful. Phylogenetic relationships between 72 taxa were investigated with maximum parsimony analyses of cpDNA sequence data (*trn*_{LUAG}-*rpl32*, *rpl32-ndhF*). The results indicate the presence of two major clades. Clade A comprises sections *Sphenanthera*, *Platycentrum*, both having centres of diversity on the Asian mainland, and mainland samples of the polyphyletic section *Diploclinium*. *Sphenanthera* (predominantly fleshy-fruited) is paraphyletic and forms two clades nested in *Platycentrum* (predominantly rain-ballist fruits). Clade B comprises predominantly anemochorous taxa with dry capsules: sections *Coelocentrum*, *Reichenheimia*, Philippine *Diploclinium*, and the Malesian sections *Ridleyella*, *Bracteibegonia*, *Petermannia*, and *Symbegonia*. Section *Symbegonia* is nested within *Petermannia*. Several characters traditionally used for sectional delimitation (including 2-locular ovaries, fleshy fruits, and rain-ballist fruits) were identified as homoplasious within Asian *Begonia*.

Phylogenetics, African biodiversity and intercontinental dispersal in *Polystachya* (Orchidaceae)

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The genus *Polystachya* (Orchidaceae: Epidendroideae) contains approximately 240 species distributed mainly in tropical Africa, but with several species in the Neotropics, Indian Ocean islands and Asia. It is one of few truly pantropical orchid genera and this, together with high morphological variation in Africa and variation in chromosome number throughout its range, makes it a good subject for phylogenetic studies. Using plastid, nuclear ribosomal and nuclear low-copy gene sequences, we find five main clades correlating with morphological and geographical trends. Species endemic to western Africa are deeply separated from those endemic to eastern Africa, but closely related to many species with more widespread distributions. One species group has undergone recent intercontinental dispersal, and this appears associated with polyploidy. Analysis of nuclear low-copy genes is giving information

about the origins of polyploids and this will be important in considering taxonomic boundaries in the group, and its dispersal out of Africa.

What happened in the past? - Power to detect rule changes from reconstructed phylogenies

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Molecular systematics has given rise to an ever increasing number of reconstructed phylogenies recording historical relationships, but including only extant taxa. There has been some study on the power of such phylogenies to reveal the history of past diversification but these have generally made simplistic assumptions of constant speciation rates and often do not consider the effect of extinction and its potential to erode important signal. I am using simulations to produce trees grown under more realistic parameters – I use a model of logistic density-dependent cladogenesis with both speciation and extinction rates varying with clade size. Under this model, I introduce a rule change – a change in clade carrying capacity – and I wish to characterise under which parameters such a rule change is detectable. Plate tectonics and climatic fluctuations throughout Earth's history have repeatedly altered the areas and habitats available for diversification; my aim is to determine which of these past changes can be recovered using data only from present-day species.



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