19th International Symposium of Subterranean Biology 2008



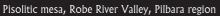
Symposium Abstracts



Held under the auspices of the International Society for Subterranean Biology

Fremantle, Western Australia 21st-26th September 2008









Inland calcretes, Mt Narryer Mid-west Western Australia



Calcrete palaeodrainage, Yilgarn craton

ABSTRACTS

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ABSTRACTS

Abstracts for posters and oral presentations are listed together in alphabetical order by first author.

Oral presentation Molecular phylogeography and phylogenetics of the Parabathynellidae (Crustacea: Bathynellacea) of the Yilgarn region, Western Australia

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The order Bathynellacea is an ancient group of subterranean aquatic (stygobitic) crustaceans. The Yilgarn Region of Western Australia displays a striking diversity of species from the bathynellaceans family Parabathynellidae, which inhabit calcrete aguifers that lie along palaeodrainage channels. This study used DNA sequence data to explore phylogenetic relationships among species genera the and of bathynellaceans from Western and eastern Australia. The study confirmed the monophyly of the order Bathynellacea and provided new evidence for short-range endemism amongst calcrete aquifers in the Yilgarn. Morphological data suggests that most species are restricted in their distribution to a single calcrete, and this was tested and corroborated in this study, using sequence data based on a region of the mitochondrial DNA cytochrome oxidase 1 gene. The lack of evidence for gene flow indicates high levels of endemism between calcrete populations of parabathynellids and is comparable to that of subterranean dytiscid beetles, amphipods and isopods from the Yilgarn region, thus providing support for the general hypothesis that calcretes are equivalent to closed 'island habitats' comprising narrowly endemic taxa. These characteristics render short-range endemic parabathynellid stygofauna, vulnerable to perturbations of species, and other groundwater, which has significant implications for their management. The conservation value of these parabathynellids is a high priority not only because of their uniqueness, but also because of their role in biofiltration and as bioindicators of groundwater quality. These results emphasize the conservation importance of these unique habitats.

Ontogenetic and phylogenetic aspects of regression processes in cave fish: immunohistochemical studies on the blind fish *Phreatichthys andruzzii* Vinciguerra, 1924

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Obligate cave-dwelling animals evolve a common phenotype that is most conspicuously characterized by the loss of functional eves and pigmentation. One of the phylogenetically oldest known cave fishes is the troglobite cyprinid *Phreatichthys andruzzii* from Somalia. This species is characterized is by some extreme phenomena like depigmentation, lack of scales and strong regression of eye, pineal and brain structures. The morphogenesis of these structures was examined using light and electron microscopic techniques. The complete degree of eye regression, which ends as an anophthalmic adult cave fish, with no trace of optic nerves and reduced optic lobes as well as a lack of pineal organ. Photoreceptor cells and lens rudiments, however, are never found in the adults of these well-adapted cave fish. Ontogenetic eye development can be detected, but it does not result in a complete lens and in differentiated retinal layers. The development of lens and neuroretina is strongly influenced by degenerating processes like apoptosis or necrosis. The pineal organ of fish is known to be a light sensitive system. In contrast to the pineal organ of mammals which has lost its light sensitivity, the pineal organ of non mammalian vertebrates contains normal photoreceptor cells which are very similar to those photoreceptor cells in the retina of the lateral eves and they have opsin. In contrast to the main function of eves, the pineal organ does not act as an optical system but it is known as a complex system that is involved in timekeeping mechanisms. Some blind cave fishlike Astyanax retained a functioning pineal organ although this species is neither exposed to light nor to circadian temperature cycles. In contrast, the pineal organ is reduced in total in the adult *Phreatichthys* andruzzii, while in the very young larvae a pineal organ can be detected immunohistochemically with the occurrence of opsin as a photoreceptive substance.

Oral presentation Molecular phylogeny and historical biogeography of cave crickets from the Southern end of the world

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In this study we reconstructed the molecular phylogeny and attempted to infer historical biogeography of a sample of cave cricket species subfamilv belonging to the Macropathinae (Orthoptera. Rhaphidophoridae) which shows a clear Gondwanan distribution. We sequenced fragments of 4 genes (12S rRNA, 16S rRNA, 18S rRNA and 28S rRNA), for a total of 2000 bp. The Macropathinae is considered to be among the most primitive sub-families of Rhaphidophoridae. It includes numerous species, belonging to 30 genera and inhabiting Australia (mainly in its southern regions), Tasmania, New Zealand and surrounding islands, South America and Africa, with only a single species in the caves of Cape Peninsula. The Macropathinae show a narrow range of ecological adaptations, being restricted to temperate and humid habitats and mainly linked to caves. We analyzed a sample of taxa from South Africa, South America, Tasmania and New Zealand. The use of relaxed molecular clocks by means of Bayesian analysis allowed us to date the main cladogenetic events after calibration of molecular clock based alternatively on the plate disjunction of Africa-South America, or Australia -SouthAmerica. Results suggest that the main cladogenetic events occurred in the Macropathinae phylogeny could be explained by vicariance hypothesis, related to the Gondwana fragmentation.

Oral presentation An overview of subterranean fauna and habitats in the Napier Ranges, Kimberley, Western Australia

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A prominent feature of the Kimberley is a massive fossil algal and stromatoporoid reef of Devonian age now exposed above the general land surface. The Devonian Reef is exposed in the east Kimberley (Ningbing Range and Jeremiah Hills) but it is much more extensively exposed in the west Kimberley where it forms a series of separate exposures (the Limestone Ranges). The Napier Range lies at the north-western end of this exposure. These now exposed reefs generally have undergone extensive karstification and it is these limestones that are a significant habitat for the subterranean fauna. The Napier Range is a prominent karst feature and contains such well-known tourism sites as Windjana Gorge, an incised river through the range, and Tunnel Creek Cave, both contained within a National Park. The majority of the Range is within unallocated crown land and operating as a pastoral lease. Speleologists have visited this area for around 20 years, documenting caves and karst features. The karst features range from giant grikes intermittently open to the surface, to small fissure oriented caves and to extensive submerged passages accessible by diver. In addition to locating new caves, speleologists have also assisted the Western Australia Museum to document the subterranean fauna. This paper will summarise the key sites where aquatic and terrestrial subterranean fauna has been observed and collected, detailing significant sites and outlining the main fauna that has been documented.

Poster presentation Ord River Irrigation Area Stage 2 Expansion Groundwater Fauna Survey

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The Ord River Irrigation Area (ORIA) is located in the eastern Kimberley region of north-eastern Western Australia and straddles the Northern Territory border. The Ord River Irrigation Scheme (ORIS) was constructed on the Ord River in 1963 and was opened in 1972. The scheme created Lake Argyle which is Australia's largest reservoir with a storage capacity of 10,760 million cubic metres of water. The ORIA includes approximately 14.000 hectares of developed land which is harvested to produce a diverse range of crops. The development of the ORIA Stage 2 M2 Project will potentially encompass an area of land that covers approximately 78,000 hectares. The land is proposed for agricultural development and comprises farmland (30,500 ha) and conservation corridors (44,500 ha). Environmental approval of the Stage 2 M2 development requires a Flora and Fauna Protection Plan to be developed which includes stygofauna. Aquifers support interstitial assemblages of bacteria and associated biofilms as well as specialised groundwater dependent fauna termed stygofauna. Groundwater dependent animals (stygofauna) were first found in the M2 Project area by the Western Australian Museum between 1994 and 1998, and as irrigation in the ORIA potentially threatens stygofaunal populations through increasing the level of the water table, and subsequently increasing salinity, this project aimed to collect baseline data prior to the Stage 2 M2 development so that any future impacts on groundwater fauna could be assessed. A combination of netting and pumping techniques were used to sample 30 bores within the ORIA, with bore selection based largely on geographic location and available hydrogeological information. This paper presents the findings of the stygofauna survey conducted in the ORIA and was commissioned by the Western Australian Department of Industry and Resources.

Relationship among cave catfish populations, genus Rhamdiopsis (Ostariophysi: Siluriformes: Heptapteridae), from a karst area of northeastern Brazil

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Brazilian cave catfishes of *Rhamdiopsis* genus are represented by 11 populations distributed in an extended subterranean aguifer from Chapada Diamantina, northeastern Brazil. Part of the aquifer is accessible via caves, located in two limestone lenses, interposed by sandstone and guartzite lenses. These latter ones can represent a barrier for cave organisms dispersion and our aim is to know if the sandstone and quartizite lenses are effectively isolating the *Rhamdiopsis* cave populations. To test this hypotheis we chose an external group, represented by an epigean species of Rhamdiopsis. The genus *Rhamdiospis* belongs to a monophyletic group (Heptapteridae clade, sensu Bockmann & Guazelli, 2003), which possibilities a robust comparison for this kind of question. We tested the phylogenetic relationships among different populations from the two limestone lenses by generating RAPD bands through low stringency PCR with random primers. PCR products were run on an ABI 3730 automated sequencer to bands and record their fluorescent size the intensities. Presence/absence of specific RAPD bands was treated as discrete characters and analyzed using both parsimony and neighbor-joining algorithms. Few samples from each population were tested, from five to 10, and, because of this, our results are preliminary yet. Our results showed Moreno cave population clusters with Natal cave population and then Moreno/Natal cave populations clusters with Lapa do Bode population. Interestingly these populations are in the same limestone lense. These three clusters are very different than Canoa Ouebrada cave population, which is located in the another limestone lense. Other result is the weak clustering between the Poco Encantado and Canoa Ouebrada cave populations, corroborating the idea that the lenses of sandstone and guartzite may play a role in the isolation of cave catfish populations from Chapada Diamantina.

Glaphyropoma sp. n. (Siluriformes: Trichomycteridae): the first subterranean copionodontine catfish in Brazil with distribution and ecological data

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Perhaps the most intriguing discovery in trichomycterid diversity in recent decades has been the subfamily Copionodontinae. The group was originally described to include the genus Copionodon de Pinna, 1992 with two species, and *Glaphyropoma* de Pinna, 1992 with one. Copionodontine occur exclusively in the Chapada Diamantina, a vast and complex plateau composed of Proterozoic terrain in the State of Bahia, northeastern Brazil. We present herein a new species of copionodontine, genus Glaphyropoma, from subterranean waters in the Diamantina Plateau, with data on its distribution and ecology. This is the first troglomorphic species of Copionodontinae, distiguished from other copionodontines by the presence of opercular odontodes, and further distinguished from its only congener, G. rodriguesi, by the reduction of dark integumentary pigmentation. The new species shares the single synapomorphy previously proposed for *Glaphyropoma*, and indirect character evidence also supports its inclusion in the genus. The presence of opercular odontodes in the new species, indicate that the absence of opercular odontodes in previously-known copionodontines is secondary, rather than primitive. Preliminary population data based on visual censuses were obtained in January and July, 2007, corresponding respectively to the rainy and dry seasons in the area. In a 150 m of long stream reach and 1 m of width. 61 individuals were visually counted in January and 59 in July of 2007, and respectively population densities of 0.41 ind.m⁻² (Jan. 2007) and 0.39 ind m^{-2} (Jul. 2007), indicating little if any seasonal oscillations in population size. Most individuals were concentrated in guiet isolated pools. The subterranean course of the creek extends for at least another 2,000 meters downstream, where, apparently the population is distributed. Most of the caves in the upper sector of Chapada Diamantina were heavily impacted by diamond mining in the past, an activity which extended to the early 1990's.

Oral presentation Are cave populations evolutionary dead ends?

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Adaptation to cave life entails mal-adaptation to life above ground. Surface species evolve into cave species, not vice versa. Caves and karst have finite life spans, so troglobites eventually go extinct without phylogenetic issue. Nevertheless, it is theoretically possible for gene flow between cave and surface populations of the same species to be bidirectional. Thus, cave specific alleles might have evolutionary futures in surface populations. Previous work on microsatellite variation in cave and surface populations of the Mexican cave fish, *Astyanax mexicanus*, suggested that gene flow between surface and cave populations was twoway and that the rate of flow from caves to surface was greater than the other way around. The present study on specific genes documents a high rate of allelic flow from the El Abra cave populations of this species into nearby surface populations, with allelic movement tending to be eastward, downstream, and gravitational.

Nutrient recycling by protozoan predation can enhance overall bacterial degradation of organic matter in groundwater ecosystems

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The combined influence of predation by flagellate protozoans and type of nutrient limitation on biodegradation of organic matter was studied mathematicallv and experimentally. Mathematical modeling. subsequently confirmed experimentally, indicated that predation can enhance carbon mineralization, and leads to higher overall activity (activity per unit sample), under nitrogen limiting conditions. Under carbon limitation, predation had a negative influence on organic matter degradation, in comparison with when predation was absent. The information obtained in this study can be of significance for the degradation of organic contaminants in oligotrophic ecosystems (e.g. aquifers), where nutrients like nitrogen or phosphorus generally become rapidly depleted. By their predation on bacteria, protozoa can recycle limiting nutrients and may stimulate indirectly pollutant biodegradation.

Comparative mtDNA phylogeography of subterranean amphipod (Chiltoniidae) species from a single Yilgarn calcrete aquifer

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Abstract: The arid Yilgarn region of Western Australia contains numerous subterranean calcrete aguifers (calcretes) with unique assemblages of invertebrate species (stygofauna), including stygobytic amphipods. Studies of the Yilgarn amphipod's mtDNA reveal monophyletic groups to be restricted in their distribution to single calcretes, with speciation occurring as a result of their geographical isolation in the groundwater system. The aim of this project is to investigate the potential for an allopatric mode of speciation to also have operated within groundwater calcretes due to their dynamic nature. Past population fragmentation events should be detectable in the gene trees of stygofauna that inhabit these calcretes. A 627-bp fragment of the mitochondrial cvtochrome c oxidase gene was amplified from subterranean amphipods 1 (Chiltoniidae) distributed across a 3.5 km² borehole grid in a calcrete at the Sturt Meadows pastoral property. Phylogenetic analyses revealed the existence of three monophyletic haptotype groups, with sequence divergences between them of >11%. They are not sister clades and are more closely related to other Yilgarn chiltoniids than to each other, suggesting that the three species were already reproductively isolated prior to colonisation of the calcrete. A large number of fixed allozyme differences were found between the amphipod groups, indicating that there are three morphologically cryptic species endemic to the Sturt Meadows calcrete. Fine scale sampling of the amphipod fauna showed only two of the species to be abundant. No common phylogeographic structure between them was seen, with one species showing evidence for a past bottleneck in its population history and one species showing an east/west phylogeographic break across the aguifer. Inclusion of nuclear loci in the intra-population study of the amphipods showing the east/west division of haplotypes is required to answer questions about historical processes involved in their population differentiation.

Changes in the stygobiont polychaetes communities due to the bioturbative activity of *Bonellia viridis* in the lagoon cave of Jameos del Agua, Lanzarote

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The marine lagoon cave of Jameos del Agua is a natural site conditioned as a tourist place. It is characterized by a semi illuminated environment due to the collapse of the ceiling, that it is known as "jameo", and by having shallow marine waters with tidal cycles. The entrance of light allows the growth of a carpet of diatoms on the bottom to 0.5-2 m depth. The communities of interstitial polychaetes inhabit the carpet of diatoms and the bottom of lava debris with sand and gravel. In the last decades has been producing a great increase of the population of the echiuran worm Bonellia viridis. Its activity has changed partly the natural conditions of the bottom. The whitish ejecta of the echiurian form domed patches that often contrast strongly with the structure of the background. These patches have been colonized by an abundant stygoxen macrofaunal and meiofaunal specimens that compete with the stygobiont endemic species. The community of diatoms carpet are characterized by populations of meiobentonic species Leptonerilla diatomeophaga, a stygobiont nerillid which feeds essentially of diatoms. Interstitial polychaetes are characteristic of areas with sand and gravel, the most important populations are of Fauveliopsis jameoaquensis, a stygobiont fauveliopsid which has omnivores trophic guilds. This species has also colonized the patches of Bonellia viridis. The majority of the new colonizing species, have established in the domed patches of *B. viridis*. These macrofaunal populations of opportunistic species have established in this new biotope; they are species with trophic guilds deposit-feeders (surface and subsurface): capitellids of the genus Notomastus, the trichobranchid Trichobranchus glacialis, the cirratulid Aphelochaeta marioni and the paraonid Cirrophorus lyra; they are all stygoxen species. Due to the new situation, it is necessary a monitoring of the communities, to value the viability of eradicate most of the population of Bonellia viridis of the lagoon cave, since these changes can affect also the endemic populations that live in the water column.

Oral presentation Bathynellacean biodiversity of groundwater in Western Australia

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Recent studies on the subterranean biotopes of the arid region of Western Australia have revealed diverse crustacean taxa of ancient lineages. Numerous Bathynellacean syncarids have been collected from limestone, calcrete, sedimentary and regolith substrata in both saline and anchialine waters in this region. Ongoing fresh. morphological investigation on this material has resulted in the finding of an unexpected diversity and novel information on the phylogeny and biogeography of this animal group. To date 37 new species of the family Parabathynellidae have been identified, of which 15 species have been published, belonging to five genera (six of Kimberleybathynella Cho et al., 2005; ten of Atopobathynella Schminke, 1973; twelve of Australobathynella gen. nov.; seven of Billibathynella Cho, 2006; two of Brevisomabathynella Cho et al., 2006). In addition, four new species of the family Bathynellidae have been identified. Many species are primitive forms with unusual characters such as large body size (length over 5 mm), the fat-bellied habitus, the possession of ornaments on pleomeres and the prominent uropod. Peculiarly, Billibathynella includes a species, which are comparable with the hypothetical stem species of the Parabathynellidae. The species of *Kimberleybathynella* are restricted to the Kimberley Region and adjacent Canning Basin, while those of three other genera occur widely in Western Australia and show a peculiar distributional pattern. The advanced species of these genera occur in near coastal and anchialine water, whereas the primitive ones in the far inland of Precambrian Cratons (Yilgarn and Pilbara) never inundated by the sea since Palaeozoic. This leads to a conclusion that at least the extant parabathynellids have been derived from surface water limnic ancestor and that the transition to the groundwater could have happened in the Notogaea.

Poster presentation Justification of *Kimberleybathynella* Cho et al, 2005 based on morpho-phylogenetic analysis

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Recent studies on the subterranean biotopes of the arid region of Western Australia have revealed a great diversity of Bathynellacean syncarids. Until now 15 species have been added in science, while three genera have been erected. Kimberleybathynella Cho et al, 2005 including six known species is one these genera. Its erection is based on (1) the six segmented antennule, (2) the flat labrum, (3) five spines of mandibular incisor process, (4) one segmented exopod of thoracopods I-VII, (5) the hemispherical male thoracopod VIII, (6) the two segmented antenna, (7) the maxilla with half fused second and third endites. (8) the distal spine of uropodal sympod being longer and thicker than others and (9) the outer terminal seta of uropodal exopod being longer than inner one. The characters (1), (2), (3), (4) and (5) occur also in Atopobathynella Schminke, 1973, suggesting a close relationship between both genera, while the characters (7), (8) and (9) have been supposed to be synapomorphies of *Kimberleybathynella* (Cho et al. 2005) in the context of this putative relationship. At the time of the erection of *Kimberleybathynella*, however, we knew *Atopobathynella* only from four species. Meanwhile, Atopobathynella is more precisely known through the description of six further species from Australia and one species from India. It is thus of interest to test if the erection of *Kimberleybathynella* is justified phylogenetically. For this purpose 39 adult morphological characters were selected to reconstruct the phylogenetic relationships among the 23 species (including four new species) of Atopobathynella and Kimberleybathynella. A phylogenetic analysis using PAUP 4.0b10 (a heuristic search for the most parsimonious trees) yielded 24 most parsimonious trees each of 158 steps, CI of 0.3608 and RI of 0.5270. In all of these trees six species of Kimberleybathynella and 17 species of Atopobathynella are clearly separated. This separation is retained in one less parsimonious trees supporting monophyly of *Kimberleybathynella*.

Oral presentation The subterranean biodiversity of China: some recent examples

Arthur Clarke

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The known karst of mainland China is extensive and still largely unexplored; its cave fauna habitats and subterranean biodiversity are diverse with new and undescribed species, and the potential for many more. Tectonic activity has resulted in large blocks of structurally isolated and hydrologically separated cave systems and the geomorphic processes acting on limestone with depths up to 1,000 to 1500m has resulted in development of massive fossil cave systems above the present day active stream level. The author has been actively engaged in the hypogean fauna studies of defined areas in three Provinces (or Autonomous Regions): Guangxi, Hubei and Jiangxi, Examples of his recent co-authored descriptions include *Macrobrachium lingyunense* (a palaemonid prawn shrimp from Guangxi), Jiangxiaphaenops longiceps (a trechine carabid beetle from Jiangxi) and Chetoneura shennonggongensis (a non-bioluminescent keroplatid fly from Jiangxi). Two other cavedwelling carabid beetles from Guangxi have also been named in his honour: Giraffaphaenops clarkei and Jujiroa clarkei. Added to the range of troglobitic fauna: carabid beetles, springtails, cave crickets, cockroaches, isopods, spiders, harvestmen, pseudoscorpions and mites, plus some recently determined millipedes, the diversity of stygofauna is enormous but yet still largely unknown or undescribed. The partially determined aquatic fauna of caves in China includes gastropod snails. planarians, worms and leeches, copepods (four families), crangonyctoid amphipods, atvid shrimps, isopods (including styloniscids), crabs, shrimps (or prawns), dytiscid beetles and three families of cavefish. There is also the potential for discovering other species of microfauna such as ostracods and possibly bathynellid syncarids. The discovery of some tinyeved gigantic tadpoles up to 14-15cm in length located several kilometres away from known surface entrances has raised some interesting possibilities for research into cave dwelling anurans. The presence of "large transparent tadpoles with reduced eyes" in Chinese caves was first reported in 1988 by a Belgian expedition. Considered an unlikely example of neoteny amongst anurans, these tadpoles are presumed to be the larval stage of a megophryid frog, possibly Oreolalax rhodostigmatus, known to breed in the springs and stream pools of limestone caves.

Poster presentation Defining cave micro-habitats: a Tasmanian perspective with new species examples

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Cave zones, associated habitats and the region where cave adapted species occur are traditionally classified by the linear distance from cave entrances and/ or the perceived extent of surface influences. Aside from climatic attributes, variations in seasonal humidity, air flow and hydrological influences (stream recharge), factors such as cave shape/ entrance morphology and the presence/ actual location or redistribution of natural or derived organic inputs need to be considered. Using the term "karst bio-space" encompassing the total living space for all hypogean species in the saturated or unsaturated zones of caves and other karst-like cavities, crevices or voids, the ecology of wild caves and tourist caves in two karst regions of Tasmania (Hastings and Ida Bay) can be described in five cave zone regions, ten macro-habitats and 44 microhabitats. The karst bio-space includes inter-relationships of species and predator-prey relationships within the subterranean food chain, together with cave bacteria and other micro-organisms in all cave zones; the dependence of cave species on these micro-organisms has not been studied here in Tasmania. In most wild caves of Tasmania, e.g., Ida Bay, organic material is naturally derived as gravity fall or stream inputs, but at Hastings where tourist caves have been developed, much of the organic matter has been introduced to the cave. The source of organic input in Tasmanian tourist caves includes forest products introduced in the form of tree trunk poles, rough sawn timber and Manfern trunks used in the construction of stairs, bridges and fern log pathways, plus the litter "inadvertently" carried in by humans or dumped as refuse during the continuing development of caves for tourism. Examples of the new cave dwelling aquatic and terrestrial fauna from Tasmanian caves include recently described and undescribed species: hydrobiid snails, copepods, ostracods, bathynellid syncarids and phreatoicids; plus springtails, beetles, spiders and millipedes.

Is cold tolerance a general ecophysiological response in the subterranean crustacean *Niphargus rhenorhodanensis*?

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Subterranean organisms live in an extreme ecosystem and share common features linked to this biotope (e.g. hypoxia and starvation tolerance, adaptation to darkness, long life cycle). Living in a thermally buffered biotope, hypogean organisms should exhibit stenothermal characteristics. Niphargus rhenorhodanensis is an aquatic subterranean crustacean which lives in karstic, porous and fissured aquifers. Ecophysiological studies showed that one population of this species (Chatillon-la-palud, France), survived long-term exposures at low temperature $(0^{\circ}C)$, and exhibited eurythermal characteristics. Surprisingly, this population was able to maximize performance over a large temperature range, including endured in its natural habitat. Moreover, temperatures never phylogenetic analyses demonstrated that this morphologically defined species can be divided into 6 independent evolutionary units. The aim of this study was thus to investigate if responses to cold exposure reported for one of the six evolutionary units could be observed in the other Several ecophysiological parameters lineages as well. (oxvgen consumption, ventilatory and locomotory activities) and metabolic responses (carbohydrates and amino acids concentrations) were measured on 7 populations. These measurements were performed on kept organisms at 11°C (i.e. biotope temperature) and 3°C. We also performed genetic analyses to corroborate N. rhenorhodanensis phylogenetic structure. We found that the 7 populations studied displayed a cold hardiness and belonged to 3 different evolutionary units. A plausible explanation links this cold tolerance to historical heritage as all locations were covered by ice during the Riss and the Würm glaciations in the Jura Mountains (France).

Identification and expression of the first inducible Hsp70 gene from a subterranean crustacean, *Niphargus rhenorhodanensis*

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Current scenarios of global warming predict an increase in average temperature of 2 to 6 °C in the next 100 years. Recent contributions have shown the consequences of such an elevation of temperature on the abundance and the geographic distribution of several taxa. However, and although they live in extreme biotopes which are thermally buffered, reaction to global warming in hypogean organisms has never been studied. Organismal reactions and evolutionary consequences to temperature increase being rather difficult to characterize, we defined a molecular marker allowing us to quantify responses to thermal variations in subterranean field-collected organisms. The protein family HSP70 is a marker of thermal stress, and expression of these proteins can be induced by an increase of the temperature. We characterized one copy of the Hsp70 genes in Niphargus rhenorhodanensis, which is a common subterranean amphipod crustacean. Using quantitative RT-PCR, we demonstrated that this copy is an inducible one and that it could be used to quantify the cellular stress endured by N. rhenorhodanensis. Finally, we evaluated N. rhenorhodanensis responses to a +2 and a $+6^{\circ}$ C temperature elevation to provide first evidences of global warming consequences in subterranean taxa.

Comparative phylogeography of crustacean stygofauna from calcrete aquifers of central Western Australia: are they climate relicts?

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Numerous groundwater calcrete aquifers of the Yilgarn region of central Western Australia have been shown to contain a diverse subterranean invertebrate fauna (stygofauna), including dytiscid water beetles and a variety of crustacean groups, such as Amphipoda, Isopoda, Bathynellacea and Copepoda. Previous morphological and genetic analyses of the dytiscid beetle fauna indicated that subterranean lineages evolved independently from surface lineages following the onset of aridity on the Australian continent during the Late Miocene/ Pliocene. This study supported the "climatic relict hypothesis" for the evolution of the dytiscids and further suggested that individual calcretes are equivalent to closed island habitats. Here we consider these hypotheses further by investigating the evolutionary history and timescale for diversification of amphipod and isopod crustaceans from the Yilgarn calcretes. Mitochondrial DNA (mtDNA) phylogeographies of crangonyctoid and chiltoniid amphipods and isopods (Haloniscus), based on Cytochrome Oxidase subunit 1 (CO1) sequence data, revealed a striking pattern of distinct and divergent mtDNA lineages, each restricted to a single calcrete aquifer. Morphological analyses further suggest that many of these mtDNA lineages correspond to distinct species. Such patterns of mtDNA and morphological diversity indicate that there has been longterm genetic isolation of calcrete populations. We present new molecular clock analyses of the likely time when calcretes became colonized by different crustacean species and assess whether aridification of Australia was the driving force leading to the diversification of species. The role of additional isolating factors in shaping the diversification of species, such as palaeodrainage channels (rivers that largely stopped flowing in the Palaeocene) and drainage divides will also be discussed.

Oral presentation New insights into subterranean life: superficial subterranean habitats

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The standard view of subterranean habitats is that they are food-poor, aphotic, and without temporal cues, making successful colonization extremely difficult. Superficial subterranean habitats (SSH) do not fit this paradigm. A variety of habitats, including talus slopes and regolith (milieu souterrain superficial - MSS), superficial ground emerging in small (hypotelminorheic), cracks, fissures in subcutaneous zone seeps (epikarst), and shallow tubes in lava, are aphotic, but with relatively high resources and temporal variability. All of these superficial subterranean habitats harbor species modified for subterranean life (eyeless and without pigment, and with elongated appendages), similar to species found in caves. This suggests that the absence of light is the primary selective factor in the evolution of the distinctive morphology of subterranean animals, a view supported by the finding of "overexpressed" genes in populations of the cavefish Astyanax fasciatus as well as a major reorganization of the opsin and other gene pathways. This is the crux of adaptation to subterranean life. Species in superficial subterranean habitats may give rise to species in deeper subterranean habitats such as caves. Preliminary analyses of morphology of SSH species compared to cave species in the amphipod genus Stygobromus shows somewhat less antennal elongation occurs in SSH species, but the differences are not large. Colonization of superficial subterranean habitats may be the result of adaptive shifts or the result of unfavorable conditions in surface habitats.

Oral presentation Mulun karst, a hot-spot of subterranean biodiversity in China

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Southern China has been the focus of major biospeological investigations during the last two decades, which give us a fairly good overview of the faunistic composition of its rich subterranean communities (Lips et al 1993, Pouilly 2003, Clarke 2006, Lynch & Duncan 2006, Duncan & Wu 2007, Latella in press, Deharveng in press). In the frame of a conservation project aimed at surveying subterranean and soil biodiversity in order to evaluate the possibility of incrasing the surface of karstic nature reserve of Guangxi, we performed in 2005 and 2007 intensive sampling in several large karst areas of the province: Bapen, Longgang, Longshan, Yachang (near Leve) and Mulun National Reserve. In this paper, we expose the results of our survey in the caves of Mulun NR, which was sampled for the first time during this project. Samples were made in parallel inside and outside caves in order to ascertain the ecological status of species. On a total of 96 collected taxa, 34 were likely to be obligate subterranean, making of Mulun the richest cave in China, specially regarding the fact that its aquatic fauna was poorly surveyed due to rarity of favourable habitats. The surveyed caves hosted 7 to 16 troglobites. The fauna was characterized by a high richness in Diplopoda (6 species), and high level of troglomorphy in several taxa. In addition, suterranean fauna of a karst close to Mulun investigated during this project was largely different from that of the core of Mulun NR, suggesting significant local endemism, and providing biological justification for extending park boundaries, which in turn would facilitate regeneration of surface vegetation in highly disturbed areas.

Comparison of oligochaete fauna inhabiting surface and cave water bodies in the sandstone mountain region of Poland

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The Jaskinia Miecharska cave, situated in the Beskidy Mts., is the longest (1800m) of the pseudokarstic caves formed in the sandstone rocks in Poland. It has a permanent stream entering the cave through small crevices, and a small pool not connected with the stream, formed by water filtering through. In years 2006 - 2007, samples of benthic fauna were taken six times from the stream above, inside and below the cave and in the cave pool. In the surface sectors of the stream, insect larvae, mainly flies (Diptera), stoneflies (Plecoptera) and caddisflies (Trichoptera) predominated, which is typical for small streams in the Beskidy Mts. Inside the cave in both types of water bodies, oligochaetes (Oligochaeta) prevailed. The taxonomic composition of this group is similar in the surface and underground sectors of the stream but a slightly higher number of taxa were found inside the cave, including one stygobiontic species. Species with various ecological requirements: typically aquatic, semi-aquatic and terrestrial ones were confirmed in all studied sites. The Enchytraeidae family (enchytraeids) was represented by the highest number of species, whereas from other families (Haplotaxidae, Naididae, Tubificidae and Lumbriculidae) 1 - 3 species were determined. The highest density of oligochaetes was found in the stream below the cave. In the Jaskinia Miecharska Cave terrestrial polychaete (Polychaeta) - Hrabeiella periglandulata, was confirmed for the first time in the aquatic environment. In karstic areas with streams flowing directly into the caves, the number of species found in underground streams was smaller than in their surface sectors. The stream in the Jaskinia Miecharska Cave is an exception, probably due to the concentration of aquatic and terrestrial taxa from the Enchytraeidae family in its more stable conditions.

New species of cave and forest litter dwelling Trechinae (Coleoptera Carabidae): emerging patterns in morphology, distribution and evolution of Tasmanian cave beetles

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Nine new morpho-species of Tasmanian cave and forest litter dwelling Trechinae are identified from the closely related genera Tasmanotrechus Moore 1972 and Goedetrechus Moore 1972 (Coleoptera: Carabidae). In both genera the identified new species, as well as those species previously described, exhibit differing degrees of troglomorphy, however Tasmanotrechus spp. are less troglomorphic than Goedetrechus spp. Typical troglomorphisms include progressive elongation of legs and antennae, and progressive reduction in eve size. Other modifications are related to the shape of the pronotum, and changes in body size. It is proposed that present day troglobitic trechines are derived from troglophilic progenitors that colonised caves from adjacent forest litter habitats during Pleistocene interglacial periods, while retreat of forests during glacial periods isolated cave populations from surface populations facilitating troglogenesis. Distribution patterns in the troglomorphic species exhibit characteristic short range endemism coinciding with discrete karst areas, or even discrete caves within a single karst area. At regional scale, the most highly troglomorphic species are found in southern karsts, while central and northern karsts appear to be characterised by less troglomorphic species. The Pre-Last Glaciation, which was more extreme than the Last Glaciation, involved extensive ice cover with associated hydrologic, geomorphic and vegetation changes to karsts located in northern and central inland settings, while karsts in southern near-coastal settings were less affected by glaciation. It is hypothesised that the highly troglomorphic species group (Goedetrechus mendumae group) found in southern karsts represent an earlier phase of cave colonisation, while the less troglomorphic species groups (Goedetrechus parallelus and Tasmanotrechus cockerilli groups) found in the central and northern karsts represent more recent colonisation(s). Extinction of older troglobites may have occurred in the central and northern karsts that lay proximal to, or were partly overridden, by glacial ice during the Pre-Last Glaciation.

Terrestrial subterranean diversity in non-karstic Archaean rock terrains: another Aladdin's Cave opening in the Pilbara region of Western Australia

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Until recently most terrestrial troglobites in Australia were known only from large (macro-) caves developed, predominantly, in karstified carbonate rocks, and less commonly from lava tubes developed in basalt. Excepting a few isolated earlier reports, and despite the accumulated evidence from other countries, there had been little searching for troglofauna in smaller (meso-) cave habitats developed in non-karstic rock terrains. This situation changed abruptly a few years ago when diverse communities of short range endemic terrestrial troglobites were discovered during routine stygofauna sampling in Tertiary channel iron deposits in the Pilbara region of Western Australia. This discovery (described elsewhere this symposium), triggered a spate of troglofauna surveys instigated for pre-mining environmental impact assessment. Here we report the discovery of diverse assemblages of terrestrial troglomorphic fauna occurring in Archaean ore-bearing rocks in the north and central Pilbara. The higher level systematic composition of the troglomorphic assemblages in the Pilbara includes arachnids (Araneae, Pseudoscorpionida, Schizomida, Palpigrada), insects (Diplura, Thysanura, Coleoptera, Hemiptera, Blattodea), myriapods (diplopoda, chilopoda) and isopods. The richness of troglomorphic species recorded at some localities (12 to 26 species) is comparable to, or exceeds, that recorded from the most intensively surveyed karst localities and lava caves in Australia. In the Archaean ore-bearing rocks, secondary porosity is developed by tectonic, mineralisation and/or weathering processes, and provides prospective habitat for troglofauna. Examination of diamond drill cores shows air-filled fractures and meso-caverns extending many metres below ground level, while regolith and colluvium may provide additional shallow subsurface habitats analogous to the milleu souterrain superficial that harbours diverse troglofaunas outside Australia. The limited sampling at just a few localities to date has confirmed that the arid Pilbara region harbours a significant diversity of troglofauna, and like stygofauna, this poses research and conservation challenges in the face of increasing demand for mine developments.

Poster presentation Subterranean amphipods in karst and flysh zone of Istria, Croatia

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In order to determine diversity of subterranean amphipod fauna of various water bodies in Istria, field studies were conducted from 2000 to 2008. The coastal area is relatively well studied, but we know much less about the subterranean amphipod fauna in the mountain area of Istria. In order to test species diversity of various eveless amphipod habitats, we have compared the ratios of different distribution types of subterranean crustaceans. The genus *Niphargus* has an extended distribution, occurs in both geological units (karst and flysh), with the higher species diversity in the karst, rich in endemic taxa. This genus has both epigean and hypogean species, where epigean species seem to prefer intermittent flysh springs and streams. There are also some species, which are specialised to brackish waters, but their number is relatively low. Recently some new stygobiotic species have been described from the epikarst zone of the Southern Istria. New records increase both, the number of taxa with restricted area and the number of widespread taxa. A short bibliography of subterranean amphipod fauna of Istria is also included. Most of the data are scattered in taxonomic works. We provided also the new insights into the biogeographic distribution patterns of Croatian part of Istrian cave, spring and stream dwelling eyeless amphipods.

Oral presentation Groundwater ecosystem functioning along a contamination gradient

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Several studies of the shallow groundwater in the coastal lowlands of Canterbury Plains, New Zealand, investigated ecosystem functioning and its responses to contamination from treated sampling array wastewater. The traversed а putative contamination gradient from an upstream (-200 m) control location to 430 m downstream of the contamination source. Water quality improved with distance from the source from some nutrients, but this change was not a simple gradient. Preliminary dissolved organic carbon fluorescence investigations using signatures indicated that organic carbon from wastewater effluent does indeed enter the groundwater, and was most apparent close to the discharge point. Stygofaunal composition and abundance reflected this gradient generally, but not consistently. Preliminary investigations using biofilm cultured on standardised native sediments in a subset of wells along part of the gradient demonstrated increasing denitrification rates (denitrification enzyme activity) with increasing distance from the contamination source. Initial studies using stable isotopes of carbon and nitrogen proved inconclusive, but indicated that dissolved organic carbon from the effluent was incorporated into biofilm, which, in turn, was consumed by some stygofaunal species. Attempts to elucidate trophic levels within the stygofauna were confounded by difficulties in obtaining sufficient biomass of mostly minute species and the predominance of omnivory.

Phylogeny and convergence in the two cavernicolous genera Spelaeonethes and Titanethes (Crustacea, Isopoda, Oniscidea)

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In Italy the trichoniscid genus Spelaeonethes Verhoeff, 1932 includes three cavernicolous species: S. brixiensis Brian, 1938 from Lombardv. S. nodulosus Verhoeff, 1932 from Veneto and Trentino, and S. mancinii (Brian, 1912) from western Liguria and Tuscany. During recent investigations in some caves of Monte Pisano, north of Pisa, Tuscany, a further species of Spelaeonethes was discovered, morphologically very similar to S. mancinii, from which it is distinguished by the presence of two sexual glands on the pleon of the males. The same kind of glands is present also on the male pleon of a species of *Titanethes* Schioedte, 1849, T. albus (Koch, 1841) from caves in north-eastern Italy and Slovenia. We used mtDNA sequence data (COI and 16S) to reconstruct the molecular phylogeny for 16 populations belonging to the genera Spelaeonethes and Titanethes. The phylogeny inference was analysed using four different approaches: Maximum Likelihood (ML), Maximum Parsimony (MP), Neighbour-Joining (NJ) and a Bayesian analysis (BI). The genetic results have been compared with the morphological characters, with a particular attention to the male sexual glands on the pleon. The genetic distances obtained by the molecular analysis showed that the populations from Monte Pisano caves with pleonal glands have a nucleotide divergence from the populations of S. mancinii. The species T. albus, with similar sexual glands on the pleon, showed a very high genetic distance compared to species in the genus Spelaeonethes, demonstrating that such glands arose convergently from independent lineages.

Oral presentation Morphological and molecular diversity of subterranean nocticolid cockroaches of the Pilbara

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The Pilbara region of northwest Western Australia has a diverse troglobitic community that includes isopods, palpigrads, spiders, schizomids. pseudoscorpions, millipedes, centipedes, pauropods, silverfish, cockroaches and beetles. Previous morphological and molecular work on schizomids and mites inhabiting pisolite mesa landforms along the Robe Valley of the western Pilbara show contrasting patterns. The schizomids comprise multiple divergent lineages, each restricted to individual mesas, suggesting that dispersal is restricted between these discontinuous patches of habitat. In contrast, multiple highly divergent lineages of mites occur sympatrically at single mesas, suggesting that dispersal between mesas is less restricted for the mites. Cockroaches are relatively abundant in the eastern Pilbara, and have representatives from at least two families, with the nocticolids being most abundant. Most animals were juvenile and individuals differed in the presence or absence of eyes. Specimens from seven deposits at three sites in the eastern Pilbara were examined for morphological and molecular diversity. We aimed first to test morphological hypotheses using molecular markers, and second to test whether the local geology presented barriers to dispersal. Two of the sampling sites were located in the Packsaddle and Jirrpalar Ranges, separated by a narrow valley approximately 2 km wide. The third site occurred approximately 250 km to the east in the Ophthalmia Range. Individuals were sequenced for variation at the mitochondrial gene COI. Phylogenetic analyses showed two major lineages. The two lineages did not strictly correspond to the presence or absence of eyes. Lineage 1 comprised haplotypes from all three ranges, whilst lineage 2 comprised haplotypes from the nearby Packsaddle and Jirrpalar Ranges. Multiple minor lineages within the two main lineages corresponded to separate deposits, suggesting that dispersal generally occurs at the local level, but recent or historical dispersal accounts for the widespread distribution of lineage

Teaching speleobiology for understanding biology: an ecology case study (interaction of surface and subterranean amphipods in springs)

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Slovenia is rich in karst (40% of its surface) and in interstitial waters outside it. The first report on cave animals from the region is more than 300 years old. Therefore, karst presents an important natural and cultural heritage of Slovene population. Speleobiology is a subject of the undergraduate course of Biology at the Univerza v Ljubliani. The subject covers several fields of biology that were explored in a number of graduation, master and doctoral theses. Undergraduate students can approach general and complex topics from different subjects of biology. Carefully selected cases and appropriately guided practical work can be used for educational and research purposes. The present study was supervised by teachers of zoology and ecology and realized by a group of undergraduate students; the first part of results has already been published in a cited journal. The aim of these observations was ecological characterization of a surface (Gammarus fossarum Koch) and an eutroglophilous (Niphargus timavi S.Karaman) amphipod, to establish what interactions between them occur and how they affect their microdistribution. Both species were found in several sampling sites below the spring throughout the year, but the abundance of the two varied along the stream spatially and temporarily. Gammarus differs from *Niphargus* in higher reproductive potential. Furthermore, *Gammarus* preferentially shelters in leaves, while Niphargus invades sand and leaves in roughly the same proportions. Stomach contents of both showed no differences. Laboratory observations found Niphargus as the more powerful predator and cannibal than Gammarus. In short, preferences for a certain substrate allow coexistence of both species. Similar food preferences can be a basis for interspecific competition whose results most probably depend on different reproductive potentials, predation, microhabitat availability, and stochastic events (drift). Its lack of habitat preference and its predation effectiveness explains the presence and success of Niphargus in subterranean waters.

Oral presentation Three distinct clades of the stygobiotic isopod Antrolana lira (Crustacea, Cirolanidae) in the Shenandoah Valley of the Eastern United States

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We obtained 14 unique haplotypes from nucleotide sequences of a 658 bp segment of the COI gene from 70 individuals of the stygobiotic cirolanid isopod Antrolana lira collected from 9 phreatic sites spanning 150 km along the Shenandoah Valley of the eastern United States. The haplotypes partitioned geographically into northern, southern and western groups of sites with some haplotypes shared among sites within groups but no haplotype shared among groups. Phylogenetic analyses identified three distinct clades that corresponded exactly to the three geographic groups. Analysis of molecular variance showed that over 99% of sequence variation occurred among the clades. Average sequence divergence values were less than 1% within sites and within clades, about 11% between clades, and about 23% between A. lirg and the outgroup Cirolanides texensis, a stygobiotic cirolanid isopod from Texas. The occurrences of the three clades match the geology of the valley. The A. lira sites are karst features developed along two northeast-southwest trending bands of exposed Ordovician age carbonates bisected by a band of shale. Sites of the western clade occur west of the shale. Sites of the northern and southern clades occur east of the shale, and are separated by a narrow constriction of the carbonates. We suggest that the three clades represent three separate episodes of colonization of the phreatic habitat from surface marine ancestors left stranded by receding sea levels at the end of the embayment of the Shenandoah Valley in the early to mid-Miocene. We did not detect any morphological difference among the three clades.

The cave-dwelling millipede fauna of Vanuatu (Espirito Santo Island): preliminary results and perspectives on diplopod biodiversity, with comments on the millipede families Pyrgodesmidae, Haplodesmidae and Doratodesmidae (Diplopoda: Polydesmida)

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We propose here the first results related to the cave-dwelling millipede fauna recorded on Espirito Santo Island in Vanuatu, Melanesia, southwestern Pacific, collected during a scientific expedition managed by the Muséum National d'Histoire Naturelle (Paris, France) in 2006. Among the diplopds collected by the 'Karst Group' (L. Deharveng et al. Coll. Leg.), we recognised the first new species of the family Pyrgodesmidae to be recorded there. Lobiferodesmus vanuatu Golovatch et al. was described from Vanuatu, differing from both previously known congeners, L. papuasicus Silvestri, 1920 (type species) and L. superans Silvestri, 1920, from Papua New Guinea, by the presence of 19 body segments instead of 20, the small size <4 mm versus 9-10, the absence of porosteles from segment 7, the more evident lobulations at the front margin of the collum, the gonopod coxa provided with an apicolateral lobe, and the telopodite divided distally into two short branches, with the seminal groove passing distally between the bases of both these branches to terminate on top of a very small solenomere located at the bottom of a median cavity. In accordance with additional data obtained from taxa newly decribed from China and Southeast Asia, the polydesmoid family Haplodesmidae Cook, 1895 was reviewed and shown to be a senior subjective synonym of the family Doratodesmidae Cook, 1896. The Haplodesmidae therefore encompasses six genera and 30 recognizable species, including six new species: two from Vietnam; one from Vanuatu, Melanesia, and three from China. According to this statement, several new synonymies and new combinations were proposed, updating the taxonomic status of many related taxa. Additional data provided by the 'Forest Group' of the Santo 2006 Expedition have been included in our present results, aiming to propose a preliminary checklist of millipede species of Espirito Santo Island and Vanuatu and a first scheme of cave-dwelling and soil-dwelling diplopod biodiversity of the island and archipelago.

Groundwater fauna as indicators of the hydrological exchanges between river and groundwater at the landscape scale

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The evaluation of hydrological interactions between rivers and groundwaters is of paramount importance for water management in the face of the constraints induced by anthropic activities and climate changes. In the Rhône river basin in France, some studies were initiated within a research programme in the framework of "Rhône Basin Workshop Zone" in different geographical areas. In order to identify the interactions zones between surface waters and groundwaters, we developed a method to characterize the hydrological origins through the signature of stygobiotic species. Two complementary approaches were used: (i) the use of all stygobiotic species to characterize exchanges direction and intensity; a water exchange index that considers the frequency of stygobiotic species in populations varies in accordance with the direction and intensity of exchanges and (ii) the search for the species of different aguifers: different faunistic characteristic assemblages can be thus observed between the various types of habitats (karst, alluvial groundwater and hyporheic zone). A sector in the Upper Rhône River (Brégnier-Cordon) has been one of the most studied areas over the last decades. Fifty-five sampling sites for interstitial fauna have been listed here. The study of the fauna has made it possible an inventory of around 100 taxa. The spatial distribution of stygobiotic species along the Rhône bypass channel corresponds to the hydrological and geomorphological characteristics, i.e. the sedimentary dynamics and circulation mode of waters in the alluvium. Hence five areas characterized by the ecological preferences of the stygobies were identified. Integration of these data in a future decision support system should help water agencies to consider appropriate long-term water policies like preservation of groundwater for potable water supplying and/or mitigation of pollutions risks.

Oral presentation Molecular biogeography of cave crickets: phylogenetic inference and evolution of the Eastern Mediterranean Dolichopoda

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This study focuses on the phylogenetic relationships among all known species of East-Mediterranean cave crickets species belonging to Dolichopoda; primarily a Mediterranean genus. Forty populations belonging to 30 cave crickets' species were included in this study: D. araneiformis from Croatia, D. remvi, D. hussoni, D. annae, D. thasosensis and D. graeca from Northern Greece; D. steriotisi. D. gasparoi, D. ithakii, D. patrizii, D. pavesi from Ionian Islands; D. giachinoi, D. kiriakii and D. sp. from Central-Western Greece; D. vandeli, D. insignis, D. petrochilosi, D. cassagnaui and D. makrykapa from Central-Eastern Greece; D. naxia and D. sp. from the Eastern Greek Islands Samos and Kalimnos; D. matsakisi, D. sp. and D. dalensi from Northern Peloponnisos; D. unicolor and D. sp. from Southern Peloponnisos; D. parakevi and D. sp. from Crete Island. One more species from south-west Turkey was considered (D. sbordonii). Three additional taxa within the same family were considered as outgroups (Troglophilus cavicola, Hadenoecus cumberlandicus and Euhadenoecus insolitus). We sequenced 1967 base pairs of mitochondrial DNA, corresponding to three fragments of the small and large subunit of the ribosomal RNA (16S and 12S rRNA) and to the subunit I of cytochrome oxidase (COI). Based on Bayesian methods and using the substitution rates previously considered for the same genes in the West Mediterranean species, we applied a relaxed molecular clock to estimate the divergence times between the lineages. Results revealed an older origin of the Eastern Mediterranean species compared to the Western Mediterranean ones. The end of the Messinian salinity crisis seems to be the main factor responsible of the speciation of Dolichopoda species in Greece, while the speciation of the western Mediterranean species appears to be mostly influenced by the climatic changes of the Plio/Pleistocene. The temporal sequence of cladogenetic events seems to agree with the geographic distribution of the study taxa, supporting the hypothesis that speciation events have been strictly allopatric and mainly determined by a multi-step sequence of geographic and/or bio-climatic separations of populations.

Oral presentation **How much is enough?**

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In the context of climate change and water resource scarcity, a major challenge now looms over environmental regulators at the nexus between groundwater resource development and ensuring conservation of its dependent ecosystems. I am working at this interface between management and science on the North West Cape of Western Australia. A subterranean fauna of international significance inhabits the groundwater in this arid, remote yet increasingly popular tourism destination. At Exmouth, the Water Corporation is responsible for the service delivery of potable water to a permanent population of approximately 3-6,000 people (off-peak and peak periods, respectively) and sees over 100,000 visitors to the area each year. A conflict exists between the need to meet current and future demands for human consumption and the prospect of restricting supply to meet environmental needs and thus sustainable management principles. The Water Corporation has undertaken stygofaunal monitoring of 21 bores over the past 9 years, making it one of the longest programs of its kind in Australia. However, analysis of this data set raises a number of issues, such as:

- 1. What sampling is required to achieve the management objectives set by environmental regulators?
- 2. Does this long-term sampling program achieve these objectives?
- 3. Is the balance between science and monitoring appropriate?

In my talk, I analyse these points briefly in the context of a case study, particularly the adequacy of compliance criteria set to observe detrimental changes to population assemblages in both space and time.

Poster presentation

The eyeless cave amphipod crustacean *Niphargus ictus* Karaman 1985 responds to light

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Amphipods of the genus Niphargus are wide-spread and abundant in interstitial waters and caves throughout Europe. One distinguishing characteristic of the genus is that, regardless of habitat, they all lack (http://web.bf.uni-li.si/bi/zoologija/cene fiser/niphargus/intro. eves htm). A series of casual observations of Niphargus ictus, endemic to the Frasassi Cave system at Genga, the Marches, Italy, suggested that in spite of the absence of eyes, the animals became more active in the presence of light. Accordingly, this was tested in the laboratory. Animals were collected from the field and maintained in ambient water and temperature (14.5°C). Each animal was placed in a clean culture dish with fresh ambient cave water, and allowed to acclimate until it had not moved for at least one minute. Then an LED light was held 6 cm over the animal, and either switched on or not. The number of seconds to the first movement during one minute of observation was recorded. Each animal was tested two times, at random: once with the light on, and once with the light off. Significantly more animals moved in the light than in the dark (X_{1}^{2} = 10.76, p < 0.01); further, of the animals that moved, the number of seconds to the first movement was significantly less when animals were exposed to the light than when they were not (paired t =8.69 p < 0.01, df= 29). Thus, the animals responded strongly to the presence of light. While some other eveless cave dwelling crustacea respond to light, to our knowledge this is the first report for cave amphipods. It is an important observation, as amphipods are frequently the dominant macrofauna in European caves, and it seems likely that light influences dispersion and migration patterns for this species, and perhaps for other Niphargus species as well.

Poster presentation Analysis of taxonomic diversity of stygofauna of the Pilbara Region, Western Australia

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The Pilbara region of Western Australia has been widely recognised as an important and highly-diverse area for subterranean fauna. Recently, increasing emphasis has been placed on stygofauna due to increased mining activity in the region. This, in turn, has allowed for greater opportunities to systematically sample groundwater bores for the presence of subterranean fauna. The findings of three surveys conducted in the Pilbara region relating taxonomic diversity to distributional information are presented here. Results from a similar survey conducted in the north eastern Goldfields region of Western Australia are also included as a comparison of regional differences in taxonomic groups. Analysis of the data reveals that up to 12 invertebrate fauna groups are often uniformly collected, with crustacean groups dominating the samples. Species identification was hampered by the paucity of taxonomic references for many of the taxa. However, Copepoda and Ostracoda species were able to be identified by specialist taxonomists. At each of the three Pilbara sites, similar copepod species richness was encountered, however species assemblages displayed only minor overlap. The copepod assemblages from the Goldfields site did not share any of the same species as sites from the Pilbara. Ostracod species assemblages were also distinct between all four sites. There were number of new records and new species in the samples. These findings add to a growing understanding of the taxonomic diversity of subterranean fauna in the region and highlight the need for greater collaboration between taxonomists, private industry and the state government.

Oral presentation Identifying potential mechanisms of speciation in Australian stygofauna

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The calcrete aguifers of the Yilgarn region, Western Australia, represent 'subterranean islands' that contain high numbers of endemic fauna. Stygobitic dytiscid diving beetles are a dominant component of the calcrete fauna, with 80 species now described from 34 isolated calcretes. Commonly, three to four distinct size classes of dytiscid species are found within calcretes, many of which have been identified as sister taxa. This has raised a number of questions regarding beetle origins and the modes of *in situ* speciation that have governed their evolution. The aim of this study was to investigate current levels of genetic diversity and population structure within species to understand the potential for allopatric speciation within calcrete systems. In order to identify the presence of isolating and/or diversifying factors within a calcrete that may result in speciation we investigated levels of genetic diversity, population genetic structure and spatio-temporal phylogeography of beetle species within a single calcrete. We carried out a comparative approach using three beetle sister species to strengthen the power of our observations in the event that all three species might share a common spatio-temporal history. The results, based on DNA sequence data from the mitochondrial gene Cytochrome Oxidase subunit 1, revealed extraordinarily high levels of genetic (haplotype) diversity and pockets of population genetic structure over a very small spatial scale (3.5 km²) within the aquifer. We also observed concordant phylogeographic patterns of population expansion and isolation by distance amongst all three species suggesting that similar factors and pressures have operated on all beetle species living in the Sturt Meadows calcrete. Here we provide an interpretation of results that reflects the unexpected levels of genetic complexity amongst stygofauna that we observed within a single cave system.

Oral presentation Perspectives of troglofauna conservation assessment and short range endemism

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Troglofauna and stygofauna are uniquely significant in Western Australia because, in addition to their high local species richness, they have become one of the standard issues examined in environmental assessment. The mining industry, in particular, has been affected by this issue because mines must remove large quantities of subterranean habitat and the processes of mineralization and enrichment appears to create favourable habitat for subterranean fauna. The involvement of mining companies in subterranean fauna has brought a very different perspective from that of researchers primarily interested in documenting diversity and is perhaps leading to fresh ideas. In this talk, we present preliminary results of survey work undertaken on behalf of BHP Billiton Iron Ore Ltd (BHPBIO). BHPBIO has recently begun surveying troglofauna across all of its active and potential mine sites in the Pilbara region of Western Australia. The objective is to provide information about troglofauna distribution in the context of enabling iron ore mining to develop in a way that does not damage conservation values of the region. There is an expectation of frequent short range endemism in troglofauna that implies each mine is likely to support unique troglofauna species. which would make it difficult to preserve all species while mining. Our preliminary results suggest that, while troglofauna species are endemic at the regional level, a proportion of species extend considerable distances along the hills and ranges in which mines occur and that the species also cross valleys between ranges. Most of this information would not have been available from a traditional approach to mine assessment where each site is investigated separately and, because of poor taxonomic frameworks, few cross-sites comparisons are ever made. Complementary genetic work detailed geological mapping and expert geological interpretation have been important additional resources in the program.

Oral presentation The dynamics of groundwater invertebrate communities in alluvial aquifers during and following drought

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Many alluvial aguifers in eastern Australia are inhabited by diverse communities of groundwater invertebrates. Often, the alluvial aquifers are hydrologically connected to streams and rivers, which supply oxygen and organic matter to groundwater food webs. However, in periods of prolonged drought many rivers cease to flow, severing the connection with the aquifer. This lost connectivity potentially changes the trophic dynamics of the aguifer, affecting invertebrate community structure. To assess changes in stygofauna community during and following drought, 8 sites in the Hunter Valley were monitored seasonally for two drought years and one post-drought year from 2005 to 2008. Water level, water quality parameters, and nutrients (nitrates, soluble reactive phosphorous, dissolved organic carbon) were also recorded, as were other potential sources of organic matter such as phreatophytic trees, infiltrating rainfall, and surface flow. Preliminary analysis indicates that increased water tables following drought initially dilute faunal densities. Higher invertebrate densities and community richness coincided with proximity to phreatophytic trees, suggesting that tree roots potentially offer food or habitat to stygofauna in alluvial aguifers as they do in cave streams. Communities further from rivers appeared to be less diverse, but more stable than those closer to rivers, probably because near-river communities contained higher proportions of stygoxenes. In periods of drought, exacerbating drawdown through pumping could not only lead to the loss of phreatophytic trees, but also to the stygofaunal communities that they potentially support. These impacts may be further amplified in highly fragmented landscapes where clusters of tree roots become isolated.

A tale of different origins: Western Australian mesas contain subterranean fauna with contrasting evolutionary histories

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The proliferation of mining activity in the Pilbara region of Western Australia has seen substantial increases in biological survey effort mostly focusing on terrestrial biota, but with strong emphases on subterranean ecosystems. The arid regions of Western Australia are now known to harbour large arrays of subterranean organisms, many with highly restricted ranges (short-range endemics). A series of mesas situated along a palaeovalley in the Robe Valley near Pannawonica comprising pisolite deposits were found to contain a wide variety of subterranean invertebrates including schizomids, mites, springtails, pseudoscorpions, diplurans, and millipedes. Whilst some taxa were too rarely collected to permit exhaustive molecular analyses, sufficient numbers of schizomids (Hubbardiidae) and mites (Laelapidae) were collected to study phylogeographic patterns using sequence data. The schizomids exhibited pronounced genetic structuring at a fine spatial scale that closely corresponded to individual mesa landforms. The magnitude of this genetic divergence was consistent with each mesa being inhabited by a unique schizomid species. Species status was also confirmed by small, morphological differences. In contrast, little genetic consistent structuring of sub-surface (edaphobitic) mites was detected. Laelapid mites possessing highly divergent genetic lineages (indicative of separate species) commonly occurred together on individual mesas. In addition, many genetic lineages occurred on multiple mesas across the study area. Gene flow between Robe Valley mesas is ongoing and extensive, and their minute size and haplo-diploid parthenogenetic reproductive modes may facilitate dispersal by wind or water. The mites have broad environmental tolerances and are not restricted to mesa landforms. In contrast the schizomids have very narrow environmental limits and cannot move between mesas due to modem climatic and soil barriers. Troglobitic and edaphic organisms in the arid Pilbara show different evolutionary histories, with ramifications for development proposals and conservation programs.

Cavernicolous planthoppers (Insecta: Hemiptera: Cixiidae) from Hawai'i and Australia: model systems for evolutionary biology and speleogenomics?

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Building on the results of previous research, we here discuss two insect models with regards to their suitability to test hypotheses on genetic change, speciation and the gene regulatory mechanisms underlying cave adaptation and to promote the potential of the field of speleogenomics. On Hawai'i, Oliarus planthoppers are descendants of a single colonizing lineage which has undergone an extensive radiation (ca. 80 species and subspecies) on the six main islands. At least seven evolutionary lines have independently colonized lava tubes on three islands. One of the lineages on Hawai'i Island, the blind and flightless Oliarus polyphemus, has been extensively studied and shown to be a complex of incipient species. In Australia, Solonaima planthoppers have colonized limestone caves and lava tubes in Northern Queensland at least four times independently. The cave-dwelling lineages display varying degrees of troglomorphy, ranging from mildly troglomorphic, troglophilic species to highly modified, eyeand flightless troglobitic species. Amazingly, the less troglomorphic species are found in several million year old limestone caves of the Chillagoe and Mitchell Palmer Karst, whereas the highly troglomorphic species are found in only 190,000 year old lava tubes at Undara. In both systems, the colonization of subterranean habitats is correlated with drastic morphological alteration such as loss of compound eyes, wings and bodily pigmentation (troglomorphies). The process of adaptation, i.e. the adjustment to the environmental stresses such as permanent darkness, permanently high relative humidity, and often scarcity of food, however, above all requires alterations of physiological and behavioural traits to enable a given organism to orientate, to locate food resources and potential mating partners. Comparative developmental genetics may hold the key to understanding the nature and dynamics of this process. The Solonaima and Oliarus species complexes offer the unique opportunity to study the underlying gene regulatory and genomic changes in closely related but ecologically independent taxa.

A surprising new stygobiotic species of the amphipod genus Gammarus (Crustacea, Gammaridae) from groundwater habitats in central Pennsylvania, USA

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Examination of amphipods collected from various groundwater habitats (i.e., springs and cave streams) in several counties in central Pennsylvania has revealed an undescribed stygobiotic species of the genus Gammarus morphologically distinct from the common, widespread Gammarus minus Say, with which it occurs syntopically. However, in contrast to accompanying specimens of G. minus, the new species is characterized by highly degenerate eyes, lack of integumentary pigment, proportionately longer antennae and other important structural differences. Considering that most of the limestone/karst areas of Pennsylvania have been explored biologically for many years and that the groundwater amphipod fauna of this region is apparently well known, finding a distinct, undescribed stygobiotic species is surprising. The new species is recorded from six localities in four valleys floored with carbonate rocks of Ordovician age that are common in the Appalachian Valley and Ridge Province of eastern North America. These valleys are drained to the east by tributaries of the Susquehanna River. Despite significant morphological differences, the new species appears related to G. minus and presumably shares a common ancestry with this species. Precisely what has driven the evolution of the new species is open to speculation. However, it may well have been the influence of Pleistocene glaciation, which did not physically reach this part of the Appalachians but probably came close enough to profoundly lower the temperature of both surface and subterranean waters. In addition to a taxonomic description of morphology, a molecular analysis is planned to determine the genetic differences between these two species of Gammarus and possibly gain some insight into how long they might have been separated from a presumptive common ancestor.

Oral presentation Subterranean biodiversity in Australian shield regions and arid areas: are there global implications?

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Australia was considered, until recently, to contain an impoverished subterranean fauna, largely on account of relative lack of karst and widespread aridity. This has proved illusory and diverse subterranean faunas are being revealed in the Australian arid zone and in areas of basement geology. The onset of aridity in the Tertiary appears to have been a prime driver both of the establishment of the habitat of arid zone subterranean habitat (groundwater calcretes and pisolites) in cratonic areas, and in driving, or drawing, underground lineages that are not prone to establish subterranean populations elsewhere. Subterranean diversity of sedimentary basins is substantial, but most is found on the ancient shield regions. The Western Shield has been emergent as a single landmass since the Palaeozoic and yet the adjacent cratonic blocks it comprises support radically different subterranean faunas. Are there wider implications to the discovery of diverse short range endemic faunas in the groundwater calcretes of arid Australia? Has speleocentricity among subterranean biologists resulted in significant subterranean biodiversity having been overlooked in shield regions outside Australia? Or, are both substrates as major centres of subterranean biodiversity uniquely confined to Australia? The climate necessary for extensive groundwater calcrete formation is restrictive and globally few locations are suitable, the most likely being south-western South Africa. Thus, the diverse subterranean fauna in groundwater calcretes may be unique to Australia. Examination of global distribution of hot-spots of subterranean biodiversity and of the distribution of biodiverse higher taxa indicates that they are predominantly found in areas outside bedrock regions. The occurrence of high biodiversity in bedrock regions of Australia is unusual, but it is implausible that the shield regions of Australia differ profoundly from those of other land masses, which suggests that vast areas of subterranean biodiversity may remain to be in discovered elsewhere.

Oral presentation Biodiversity of cavernicolous crabs and conservation of cave habitats in the Philippines

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The Philippine archipelago is fringed widely with Pleistocene uplifted coral reefs and is composed of older limestone bodies where a large number of cave systems are developed. During the past few years, we surveyed over 60 anchialine and freshwater land caves on 18 islands. We found 19 decapod crustacean species belonging to seven genera in four (Gecarcinidae, Hymenosomatidae, families Sesarmidae and Sundathelphusidae) that inhabit subterranean ecosystem. We identified four of them as new, one of which represents a new genus (Hymenosomatidae), another one is a new species of genus Karstarma and the other two belong to genus Sundathelphusa. Of the 19 species identified, five are cave-obligate species, seven are known and classified as epigean species, while the rest shows various degrees of cave adaptation ranging from slight loss of pigmentation to elongation of legs and reduction of eyestalk. It is of interest that the freshwater crab genus Sundathelphusa alone is comprised of all types of cave species, namely stygoxene, stygophile and stygobite. Much of the caves we surveyed are frequently visited by locals and tourists, and so anthropogenic disturbances are quite evident. While these caves are just a fraction of the total number of all caves in the Philippines, the increasing human pressures to the subterranean habitats are so alarming and thereby threatening the cave biodiversity before they are discovered.

Oral presentation Developmental relationships between eye regression and constructive traits in Astyanax cavefish

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The blind cavefish Astvanax mexicanus is noted for eve and melanin pigment regression but has also evolved a large number of constructive traits relative to conspecific epigean fish. These constructive traits include increased larval jaw size and taste bud number, enlarged hypothalamus and olfactory bulbs, expanded cranial neuromasts, and more extensive fat deposits. Examples of constructive behavioral traits include a change in feeding posture, which allows cavefish to more efficiently sample particles from the substratum, and attraction behavior, in which cavefish rapidly swim toward and investigate moving objects in their surroundings. A current hypothesis to explain cavefish eve regression is indirect selection for constructive traits based on developmental tradeoffs with optic development. Here we will discuss several lines of evidence that support the tradeoff hypothesis. First, we will describe experiments that reveal the mechanisms of cavefish eve degeneration during embryonic development and implicate the morphogen Sonic Hedgehog (Shh) in this process. Second, we will discuss experiments showing that increased jaw size and taste bud number in cavefish is regulated by Shh overexpression. Third, we will describe experiments suggesting an inverse relationship between eve formation and increased jaw and taste bud development regulated by Shh signaling during early embryogenesis. Finally, we will present genetic analysis of attraction behavior that has identified 9 quantitative trait loci (QTL), several of which overlap known QTL for eye regression. The developmental and genetic evidence support the possibility that eyes were lost in Astyanax cavefish indirectly through selection for enhanced morphological and behavioral constructive traits that are adaptive in the cave environment.

From "terra incognita" to the center of biodiversity: The story of Australian Candoninae (Crustacea, Ostracoda)

Ivana Karanovic

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Dry, bare, hot, dusty..., are the words describing 2/3 of the Australian landscape. This is certainly not the place where freshwater crustaceans would thrive. But crustaceans are tough animals, more than 500 million vears old. They have found a perfect niche in such a hostile place: subterranean waters. Discovery of calcrete systems in Western Australia revealed unprecedented crustacean diversity. During the past decade the number of subterranean Candoninae in Australia has increased dramatically, from nil to 95 species. Due to intensive mining activities. subterranean waters of Australia are now sampled on a regular basis, and there are 40 new species awaiting description. So far, this is about 1/3 of the world diversity. Australian Candoninae is classified into 14 genera and four tribes: Candonini, Candonopsini, Danielocandonini and Humphrevscandonini. All species, 13 genera and one tribe (Humphreyscandonini) are endemic to Australia, and most of the species are short range endemics as well. Because the Pilbara region is a hot spot for the Australian mining industry, it is the best explored region and more than 80% of known species were collected from there. Australian Candoninae represents very old lineages in the subfamily. Some of them (tribe Humphreyscandonini in particular) probably evolved from pre-Cretaceous continental connections, as they show close relationships with some Euro-Asian recent and fossil Candoninae. On the other hand, Danielocandonini is an obvious Gondwana tribe, being present in Africa and South America as well. Candonopsini from Western Australia are close to African species, while those from Queensland and New South Wales are close to South America. This old and unique fauna, its phylogenetic connections with the rest of the world fauna and distribution within this continent give new implications for the study of Candoninae evolution, and also raises the need for rigorous protection of their pristine habitats.

Zoogeography of Australian subterranean copepods (Crustacea) contradicts assumptions of monophyly of large continental blocks

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In Stygofauna Mundi, an important reference work for subterranean biologists. Australia was divided into only two zoogeographic regions: Western Australia and "other areas of Australia and Tasmania". In the same book Europe was divided into no less than 34 different regions and subregions ("zones", "provinces" and "districts"). Unfortunately, Australia is not as well explored as Europe, but what little we already know points to the previous lack of data as the only explanation for this huge discrepancy. During the last decade intensive investigations of many subterranean groups in Australia revealed unprecedented diversity and short range endemism, comparable only maybe to that of the European Balkan Peninsula. The difference is that in Australia wherever we start new research we discover another such area. During the last eight years I have tried to document and publish as much of that diversity regarding copepod crustaceans as possible, in many papers and three large monographs. Surprising differences between subterranean faunas of some neighbouring regions in Western Australia prompted me to propose novel such as "the pulsating desert hypothesis". dynamic models, Comprehensive investigations of copepods from the Australian marine interstitial only echoed patterns discovered in inland subterranean waters. Dispersal and vicariance are often considered competing hypotheses in historical zoogeography, but both processes have shaped the recent distribution of Australian copepods. However, what became apparent more than anything is the importance of looking at small-scale patterns when inferring Gondwanan zoogeography. Although the discovery that different parts of Australia have closer affinities to other Gondwanaland fragments than to each other is not new, it has significant implications for further studies of this apparently ancient subterranean group of crustaceans with very poor fossil record. Research done so far on ostracod crustaceans (which have a very good fossil record) and some centipedes all point to the same direction.

Molecular evolutionary divergence and phylogeography among Southern Chinese cave crickets of the genus *Diestrammena* (Rhaphidophoridae)

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From 1995 through 2007 French, Slovenian and Italian biospeleological expeditions have been carried out in different provinces of Southern China. They retrieved a relevant number of cave crickets of the genus Diestrammena. which belongs the worldwide-distributed familv Rhaphidophoridae. The collected populations showed different degrees of specialization to cave life in terms of development of eves (from normally developed to completely blind) and degree of pigmentation. Very little knowledge is currently available on the systematics and evolutionary relationships of the group in the area. Here, we have used sequence polymorphisms at two loci (one mitochondrial, Cyrochrome Oxidase I-COI and one nuclear, Internal Transcribed Spacer I- ITSI for a total of 1246 base pairs) to unravel relationships among 30 Diestrammena populations from Southern China (three provinces: Guizhou, Guangdong, Jiangxi). For further comparisons we have also included and representatives of the genus Eutachycines (Rhaphidophoridae) from Laos and Thailand. Molecular data revealed a profound level of genetic divergence. Results yielded from multiple phylogenetic methods can be summarized as follows. The genus *Diestrammena*, as currently defined, is not a monophyletic taxon since the three Eutachycines populations included in the study are embedded within it. Blind and reduced-eved populations are not monophyletic either; rather dependence upon caves is likely to have evolved multiple times independently. The strictly cave populations are placed basal in the phylogeny of the group. The same area (province) hosts a mixture of lineages with from normal to slightly reduced eyes that are separated by deep phylogeographic breaks. This geographical distribution might be explained by envisioning an evolutionary scenario based on zones of secondary admixture among allopatrically evolved lineages. Conflicts between topologies based on both markers and their impact on the final species tree are also discussed.

Oral presentation The Tethys enigma - Optimization of DNA sequence data in paleogeographic analyses

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Many groups of obligatory groundwater organisms have similar disjunct geographic distributions, both at regional and global scales. Stygobiotic taxa with disjunct large-scale distribution patterns are often considered as remnants of ancient faunas. However, the biogeographic history of each group is unique and often quite complex, for example, involving multiple transitions from marine to brackish or freshwater conditions (and vice versa), and (re)-colonization events at different times. Hypotheses that propose a Meso- or even Paleozoic origin for stygobionts with disjunct distributions have been traditionally based on comparative morphology and vicariance. In recent years, analyses of DNA sequence data have become an increasingly powerful tool to evaluate such hypotheses by estimating molecular divergence times. Despite the growing number of publications in this field, the currently used methodical standard is far from optimal. Apart from taxon sampling and the choice of markers, the optimization of multiple sequence alignments is a critical, but frequently neglected factor in phylogenetic and phylogeographic analyses.

Oral presentation Stygofauna in northern New South Wales - unpredictable but interestingly diverse?

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In rural Australia, groundwater is a valuable resource, providing much of the domestic, agricultural and industrial water needs. At the same time, agricultural practices threaten shallow aguifers through pesticide contamination and nutrient addition. However, there remains little information on the ecological significance of agricultural landuses on groundwater ecosystems. The aims of this study are to identify spatial patterns in the stygal and microbial assemblages of shallow alluvial aguifers in areas of intensive irrigated agriculture. We focus on irrigation region of the Gwydir River Catchment in NW New South Wales Australia. We hypothesise that irrigation practices will facilitate the leaching of pesticides and nutrients to groundwater, which in turn will affect groundwater ecosystems and causing changes in the groundwater biota relative to non-irrigated areas. Groundwater fauna and water quality were sampled in 18 bores across the Gwydir River Valley in both summer and winter. Stygal specimens were identified to genus were possible, however as knowledge of groundwater biota in this region is limited, many specimens are currently only identified to family level. Microbial activity was measured using cotton strip assay and Biolog TM Ecoplate methods. Heterogeneity of the stygal assemblages limited our ability to detect patterns related to agricultural practice, however, a diverse array of stygofauna were found, with copepods, amphipods, syncarids, mites and oligochaetes being widely recorded. Microbial activity did not significantly vary between irrigated and non-irrigated sites. There were no differences found between water quality tests completed at irrigated sites compared to non-irrigated sites. These findings may be related to the lack of agricultural activity in the region due to the widespread drought currently being experienced in the region. The region has seen cotton (the main crop in the region) being planted at a 60 year low last year, due to the drought.

Poster presentation Quantifying the heterogeneity of microbial activity and diversity in aquifers

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The effective management of groundwater requires specific guidelines for sampling and assessing groundwater ecosystems. Groundwater biota, including microbes are often overlooked in the assessment and management of groundwater at contaminated sites. This study reports the preliminary results of microbial sampling from existing shallow bores in aquifers in south eastern Australia. We investigate the enzyme activity (lipase/esterase (FDA) & B-glucosidase) and metabolic fingerprint of microbial communities within aguifers of different geologies and origin (fractured rock, restricted area - pristine; sandy, metropolitan; alluvial, agricultural) across space and time. The variability of microbial activity within an aquifer limited our ability to detect differences among aquifers, with site to site variability being, in some cases, greater than aguifer to aguifer variability. The alluvial aguifer, located in a predominantly agricultural region had similar levels of FDA microbial activity (0.06-0.1 ng/mL/h) among sites despite their considerable difference apart. However, closely located sites in the sand-bed and fractured rock systems showed relatively large variability. Repeat sampling showed a slight decrease in the level of enzyme activity over time. Although variable, microbial enzyme activity remained lower on repeat visits compared to initial samples. With water guality in the bores remaining relatively stable over time, the role of above ground activities. sampling artifacts, and the groundwater-surface water relationships are being investigated as possible causes of this variability. The importance of this variability to the assessment of aguifer condition is considered.

Islands underground: species limits, phylogenetics, and conservation of *Neoleptoneta* spiders in Texas caves

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Cave-adapted invertebrates are characterized by their unique morphology and extremely limited geographical distributions. Their patterns of endemism have made them conservation priorities, especially in the Southwestern USA, where rapid population growth threatens sensitive cave habitat. Here we present a phylogeny of Neoleptoneta spiders in Texas, encompassing specimens from all published and several new cave localities, including the endangered species Neoleptoneta microps and Neoleptoneta myopica. Results show that several species are more broadly distributed than previously known, and genetic data suggests the presence of subterranean connections between caves. Alternative models of speciation for cave organisms are presented and compared to our phylogeny in order to test for support of the climaterelict model. The phylogenetic history of *Neoleptoneta* shows a pattern of repeated, independent invasion of caves within a relatively short geologic time scale. The majority of troglobitic lineages share closely related surface relatives, which fails to support the climate- relict model.

Oral presentation Does subterranean fauna have altered rates of molecular evolution?

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The rate of molecular evolution is subject to factors such as body size, generation time and effective population size. In related species where these factors differ due to adaptations to life in different habitats one expect that differences in molecular rates are detectable accordingly. Generally subterranean fauna (stygobites and troglobites) are expected to have longer generation times, lower fecundity and reduced effective population sizes compared to their epigean congeners. However, the expected effect of these individual factors on the molecular rates may not be all in the same direction: e.g. increased generation times will slow down the molecular rate, while decreased population size and lower fecundity may speed up the molecular rate. Here I test if subterranean lineages have an altered rate of molecular evolution by means of independent evolutionary contrasts of epigean and stygobitic lineages of mitochondrial data sets of diving beetles (Dytiscidae) and atyid shrimps (Atyidae). I find that in 14 out of 18 independent epigean and stygobitic contrasting pairs, the stygobitic lineages have an increased molecular rate. The relative importance of the above mentioned factors will be discussed. The finding of increased rates of molecular evolution in stygofauna is important especially when attempting to estimate divergence times between epigean and stygobitic lineages. Due to increased rates of molecular evolution in stygobitic lineages, divergence times in these lineages may be over-estimated.

Poster presentation

Preliminary data on the prospection of the mesovoid shallow substratum (MSS) on Gran Canaria (Canary Islands, Spain)

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The preliminary results of an ongoing research on the invertebrate fauna of the MSS on the island of Gran Canaria are presented. The study of the terrestrial hypogean fauna in the Canaries had been usually carried out in the four western islands, with relevant results on its high diversity and island speciation. However, in the eastern islands this subject had been hardly accomplished because of the apparent worse conditions of their caves, either very dry (Lanzarote and Fuerteventura) or very scarce (Gran Canaria). Indeed, only 5 among the 124 adapted species so far know in the archipelago were described from the eastern islands, three of them from Gran Canaria. Our current sampling in the MSS of Gran Canaria has shown that the absence of caves does not imply an extremely poor fauna. A special type of pitfall traps has been designed for sampling the MSS repeatedly for a long time, with minimal disturbance of the habitat once the system is stabilized. It is made of a PVC pipe with abundant small holes in its lower half, with a jar fitting at the bottom and containing bait and preservative liquid. The pipe is set vertically underground in such way that the holes reach the MSS, and the top remains a few centimetres below surface, closed with a cap and covered by soil. 12 traps distributed in 6 different locations have been working during the last five years with some intervals. Besides some material not yet identified, as much as 11 new species of hypogean arthropods have been found, namely one species of Scotophaeus and one Troglohyphantes (Araneae). one *Dolichoiulus* (Julida), two Medon (Coleoptera: Staphylinidae), four different Oromia and one Laparocerus (Coleoptera, Curculionidae), and an unknown genus of Chthoniidae (Pseudoscorpions).

Poster presentation From 1896 to 2008, a sight of the stygobitic family Stenasellidae (Crustacea, Isopoda, Asellota)

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Discovered in 1896 in gours and in the underground river of Padirac (SW France) and first considered as a "living fossil", this family of stygobitic asellotan Isopods includes now 10 genera and more than 75 taxa, mainly scattered in North-tropical interstitial and/or karstic waters of the Ancient and New Worlds. Their morphology and global repartition indicate that two centers of dispersal may be at te the origin of this underground freshwater fauna: an "Atlantic" one, for American and Western African genera, a "Mesogean" one for the genus Stenasellus, whose species live from SW Europe (Portugal, Spain, France, Italy), the Middle East region (Kenya, Somalia, Oman, Turkmenistan) to the Far East: Cambodia, Thailand (inland and peninsular parts, Phuket isl.), Indonesia (Sumatra, Java, Borneo-Kalimantan isl.) and Sarawak (Borneo isl.). Although low sea-level periods of Quaternary times seem more or less responsible for the local repartition of some species (these ones able to expand in coarse alluvial deposits of hydrographic systems), the global repartition of the second stock (Stenasellus) recalls the geographical position of the mesogean sea where their anophtalmous marine ancestors were supposed to live.

Poster presentation Recent investigations on subterranean animal species in Sardinia, and on their conservation

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From the biogeographic and biospeleological points of view, the subterranean fauna of Sardinia is one of the most interesting in the Mediterranean area: very rich in taxa, mostly endemic to the island, it is well known and currently investigated. Nevertheless, the amount of unexpected discoveries is increasing every year. In the last four years, we focused our attention on some topics. In particular, (i) we examined the larval morphology of several species of troglobitic beetles, and (ii) we monitored temperature variations both in show caves and in not visited caves, with the aim to increase our knowledge of the anthropogenic impact on subterranean animal species. Concerning the latter purpose, we collected data on temperature variations in one of the most visited show caves in Sardinia (the Bue Marino cave), as well as in a cave not open to the tourists. On this subject, we tested a very cheap but effective method that could be used for monitoring caves on a large scale. During these field investigations, we had the opportunity to describe two highly specialized troglobitic beetles (Sardaphaenops adelphus Casale, 2004, and Patriziella muceddai Casale, 2004) and a new species of troglobitic Histeridae (Sardulus sacerensis Casale & Marcia, 2006) and its larva. Furthermore, we described the larval features of the aphaenopsoid carabid Sardaphaenops adelphus, and we are describing the larva of the troglobitic molopine carabid Speomolops sardous. The study of larvae of several species of cave dweller cholevid beetles, belonging to the genera Ovobathysciola and Patriziella, is in progress. Furthermore, during our field activities in caves, we sampled some not yet described species: two carabid beetles (of the genus *Typhloreicheia*), two flatworms, one spider, one Eukoeneniidae palpigrad, one Isopoda, one Diplopoda, one Curculionidae belonging to the genus Amaurorhinus, and some others.

Poster presentation Anchialine fauna from the Canary Islands: a checklist of recorded taxa for the La Corona lava tunnel (Lanzarote)

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We provide a checklist of 49 recorded taxa for anchialine sections of the La Corona lava tunnel, including information on habitats, distribution in the cave and main references for each species. Of the eight major groups recorded, the Crustacea show the highest diversity with 26 species and the highest degree of endemism (92.3%). We found thirteen species of Polychaeta, seven of which also occur in offshore waters (46.2% endemic). Except for two species, the polychaetes are all benthic species (some interstitial), whereas most of the crustaceans are exclusively pelagic or epibenthic. The species-richness of other major groups is limited and lacks endemic species, with one endemic species of Mollusca as single exception.

Distribution patterns of the interstitial Nerillidae (Annelida: Polychaeta) in La Corona lava tunnel (Lanzarote, Canary Islands), with some remarks on the recent colonization processes of the meiobenthic communities of the cave

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The meiofaunal Nerillidae are the species-richest family among the interstitial polychaetes, occupying an extreme variety of habitats worldwide and being also dominant in major cave systems, where Mesonerilla, Longipulpa and Leptonerilla are the most representative genus. Its diversity, together with its direct development and the exclusively interstitial distribution, makes them an excellent group for modelling recent speciation processes of the meiobenthic communities in those marginal habitats. The present study comprises populations of Nerillidae that dwell in La Corona lava tunnel, an extensive cave system, which extends more than 1600 m into the sea, with three different sections formed by natural collapses: Cueva de los Lagos, the anchialine lagoon Jameos del Agua and Túnel de la Atlántida, with the so-called Montaña de Arena, a 40 m high sand dune formed by sea infiltration, at 700 m from the tunnel entrance. The samples were collected during several surveys between 1993 and 2008 and led to the record of five species of Nerillidae, all of them showing different distribution and habitat preferences within the cave. Mesonerilla aff. intermedia. Mesonerilla aff. armoricana, both typical from off-shore species, and Meganerilla sp. were collected at Montaña de Arena, together with a high diversity of interstitial taxa. These collections reflect a high external influence on this locality, which is also inhabited by a number of eutroglobitic species. Leptonerilla diatomeophaga, recorded from the diatom bed in the Los Jameos del Agua, showed a high degree of ecological specialization. Finally, Mesonerilla n. sp., a new nerillid only known from this cave system, was found in all sections of the lava tunnel, even in the most isolated areas from Los Lagos cave. These distribution patterns, together with the abiotical parameters and the diversity of the accompaining fauna in each tunnel section, allow us to make some provisional conclusions on the colonization processes of the Nerillidae in La Corona lava tunnel.

Poster presentation Aquatic fauna in the unsaturated area in caves of Pădurea Craiului Mountains (northwest Romania)

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The local diversity pattern of fauna found in percolation water in the unsaturated area in five caves of Pădurea Craiului Mountains (part of The Apuseni Mountains, northwest Romania) has been investigated. Research has been conducted within a karst area of approximately 3 km² and an average elevation of 450 m a.s.l. Dripping water has been continuously sampled between November 2007 and August 2008. Monthly variation of physical and chemical parameters (pH, temperature, conductivity, Al, Cr, Fe, NO₂, and CCO-Mn) and species distribution pattern have been assessed in order to establish small-scale correlation between caves and between stations inside caves. The fauna of the investigated caves is heterogeneously distributed and its composition varies from one station to another. High diversity of aquatic taxa has been recorded throughout the investigated months; they belong to 10 groups: Harpacticoida, Hvdrachnidia. Cvclopoida. Collembola. Oligochaeta. Nematoda. Amphipoda, Ostracoda, Isopoda Insecta. and The copepods (Harpacticoida and Cyclopoida) are the most abundant in the dripping water and represent up to 49.2% from the reported aquatic taxa. The stygobites identified within the crustaceans are representatives of three fauna groups: cyclopoids [Acanthocyclops reductus reductus Chappuis, 1925; Speocyclops troglodytes (Chappuis 1923)], harpacticoids (Parastenocarididae) and amphipods (Niphargus tenuicaudatus Schellenberg 1940 and Niphargus andropus Schellenberg 1940). One harpacticoid species is endemic for the Apuseni Mountains and one is a new record for Romania. The most widespread groundwater harpacticoid species are: Spelaeocamptus spelaeus (Chappuis 1925) and Bryocamptus (Bryocamptus) caucasicus Borutzky 1930 and the richest harpacticoid genera are Bryocamptus and Maraenobiotus. The factors responsible for the observed pattern of faunistic records in the investigated caves are discussed within the local and regional context.

Oral presentation Bioluminescence in cave glow-worms: signs of altered circadian rhythmicity

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Australian glow-worms are in fact fly larvae (Order Diptera, Family Keroplatidae). They emit light from cells in the malpighian tubules to attract prev into their webs. They are found in suitable wet caves as well as in rainforest settings. Rainforest glow-worms cease glowing on exposure to light so they glow only at night. They possess a circadian rhythm of light output, demonstrated in the laboratory through their ability to maintain cyclical glowing for many weeks in constant darkness. Because glow-worms reach high population levels in caves where they do not receive strong daily resetting stimuli, we investigated whether cave glow-worms are rhythmically bioluminescent. Recording of light output of the Tasmanian glow-worm, Arachnocampa tasmaniensis, using time-lapse digital photography, established that cave glow-worms maintain strong rhythmic light output. The time of peak light output is different to rainforest glow-worms: cave populations glow most brightly when it is daylight outside the cave and most weakly during the night: they are completely out of phase with adjacent rainforest populations. We discuss the possible basis of the phase-shift and synchronization within caves.

Oral presentation Interstitial fauna as indicator for heavy metal pollution

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Aries River (north-western Romania) is a good model for the study of interactions between surface aquatic fauna and interstitial groundwater fauna, in the ecotone zone represented by the riverbank sediments. This river is one of the most polluted in Romania, the area being known as very important in mine exploitation, especially gold and heavy metals. Our preliminary and sporadic results indicated a change in interstitial communities' composition in the polluted river section and the extraordinary flourishing of stygobiontic forms. Beginning with November 2007 we made again a more systematic study, by sampling monthly interstitial fauna, water from surface and interstitial, and sediments. We used a Bou-Rouch pump and sampled at about 1.5 m in depth, filtrated the water and analyzed water and fauna in laboratory. Beside these, superoxide dismuthase activity was determinate in laboratory from organisms' tissues and fluids, as good indicator of stress induced by environmental factors (such as pollutants in our case). Water chemical analyses show increasing pollution down-stream. Surface and interstitial waters have similar chemical composition without reaching critical values, while sediments show high degree of pollutant accumulation. Fauna composition can be also correlated with water quality. So far, only Crustaceans were identified at species level but fauna differences between clean and polluted sections are obvious, and seasonal variations are also discussed. The conclusions are emphasizing the role of competition in groundwater colonization and the highly dynamics of aquatic fauna, both of surface or groundwater origin, in the interstitial ecotone.

Poster presentation Biodiversity on caves of the Valencia Community

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This work examines in depth the biodiversity of invertebrates that harbor the cavities selected in the Life00/NAT/E/7337 project, titled "Conservation of bats in the Valencian Comunity". We obtained information of great interest on the biodiversity to put in order by importance these cavities and decide on the conservation and management appropriate to each. The invertebrate fauna is represented by a wide range of diverse groups, but mostly belong to Arthropods. Many of these species are forms of high endemicity, with very small populations and some of them have high degree of specialization on underground habitats and are regarded as living fossils. Knowledge of invertebrate cave fauna can help to estimate, with objectivity, the conservation interest of a cave to encourage better management of Natural Heritage. The development of this work obtained an inventary of the invertebrate fauna in the cavities, and analyzing these data with statistical methods, we obtain the relative importance of each one depending on the rarity and evolution of its wildlife. The methodology used in this study can be applied to assess the importance of other caves with invertebrate fauna of greater scientific value and in need of special conservation measures.

Poster presentation The first vertebrate from the hyporheic: a new species of Phreatobius Goeldi, 1905 (Siluriformes, Incertae Sedis) from northern Amazon, Brazil

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We describe the first vertebrate known to be a dweller of the hyporheic environment, a new species of *Phreatobius* from the Rio Negro and Rio Branco systems, Amazon basin. The genus Phreatobius currently includes three species, all from the phreatic waters in the Amazon basin: P. cisternarum, P. dracunculus and P. sanguijuela. The new species herein reported differs from all congeners by the dense dark pigmentation along the dorsal and lateral regions of the body; by the opaque dark red color in life; by the pseudotympanus not visible externally; by the five pectoral-fin rays and by the number of caudal procurrent rays, 12 to 20 ventrally and 33 to 40 dorsally. It differs further from P. dracunculus and P. sanguijuela by the presence of eyes. The new species is the first vertebrate reported to inhabit the hyporheic zone, defined as an interstitial habitat bounded above by surface water in the river channel and by the groundwater below. In the Amazon this habitat is composed by deep layers of submerged forest litter and detritus intercalated by layers of sediments in various configurations. Adults individuals of the new species were collected from deep litter banks in the stream channel and juveniles from fine root systems in the hyporheic zone under banks. The hyporheic habitat worldwide has just recently begun to be explored biologically, and is yielding unexpected biodiversity, until now only of invertebrate taxa. It suggests a dynamic evolutionary interaction between riverine ecosystems and the phreatic/aquifer environments in Amazon where the previous Phreatobius species were found.

Poster presentation

Using DNA to resolve the phylogenetic placement of a morphologically aberrant taxon, the stygal catfish *Phreatobius* (Goeldi, 1905) (Ostariophysi: Siluriformes)

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Phreatobius is a peculiar subterranean genus distributed in the Amazon basin inhabiting hyporheic and phreatic ecosystems. Although its inclusion in the order Siluriformes is uncontroversial, the familial allocation of *Phreatobius* is problematic. The highly specialized morphology and scarcity of specimens for study has led to highly divergent ideas about the phylogenetic affinities of Phreatobius. At least five different families have been proposed as related to the genus since its discovery in the early 20th century: - Trichomycteridae, Cetopsidae, Clariidae, Plotosidae, and Heptapteridae - separately or in combination. Comparative morphological data so far clearly have been insufficient in their ability to resolve the phylogenetic position of *Phreatobius*. For this study, samples from different species of Phreatobius were collected in order to obtain molecular data potentially informative to help resolve its relationships. We present results from nuclear Rag1, Rag2 and mitochondrial 16S rRNA gene sequences for P. cisternarum, P. dracunculus and two additional species as vet undescribed. The data on Phreatobius were combined with equivalent sequence information from representatives of several other siluriform families, and analyzed by maximum parsimony. Results strongly indicate that Phreatobius forms a monophyletic group with Pimelodidae and Pseudopimelodidae, but relationships among the three taxa remain unresolved. Our hypothesis suggests that *Phreatobius* needs to be allocated in its own family, as Phreatobiidae (already available at subfamilial level).

Poster presentation A new species of genus *Sphaerosyllis* (Polychaeta: Syllidae: Exogoninae) from the Atlantida Tunnel, Lanzarote, Canary Islands

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The species of the genus *Sphaerosyllis* (Syllidae, Exogoninae) are characterized by having a small and slender body of few mm long, most of them are interstitial and they also have been found associated to biogenic substrates. The new species was collected in interstitial habitat of Montaña de Arena in the Atlantida's Tunnel, a marine lava tube of La Corona volcano (Lanzarote, Canary Islands). The species is characterized and distinguished from all previously described species of the genus for having the combination of lack of eyes, parapodia with four heterogomp compound falcigers with long blades and dorsoventral gradation in its size, solitary dorsal simple chaetae, present from chaetiger 1. Female with ventral incubation of the eggs and development of attached juveniles (epigamy), it only happens in setiger 12. Males with long ventral cirri from setiger 13, without natatory chaetae. *Sphaerosyllis* n. sp. is the first record of a troglobitic species of Syllidae. The species shows a clear adaptation to cave environment.

Oral presentation Geological age and troglobiomorphism in volcanic islands: is there a correlation?

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The Canary archipelago is undeniable of volcanic origin, but the geology of each particular island varies, mainly due to age differences, type of rocks and volcanic structures. These parameters, in conjunction with past and present climate, and geological events such as massive eruptions or huge landslides, have conditioned the presence/absence of hypogean fauna in different areas. Thus, three of the seven islands (Tenerife, La Palma and El Hierro) which are younger, less eroded and with more humid climate than the eastern ones, concentrate 84% of the terrestrial troglobitic species known so far. Since the Canaries are oceanic islands, all troglobites have evolved locally and are endemic to a single island, or even to small areas within an island when isolation depends on geological factors. Many genera include different local troglobites playing similar ecological roles, as it occurs in Loboptera cockroaches or in Trechus, Wolltinerfia and Domene beetles. In other cases, this kind of vicariancy comes from separate lineages with close ecological requirements, usually different genera from the same tribe or subfamily. Often, different degrees of troglobiomorphism are evident within each particular genus, and also within groups of "unrelated" vicariants. Moreover, this degree of adaptation is similar among all related and unrelated species within each local fauna. In some cases this can be linked to the geological age of the areas occupied by the particular faunas, and in others it seems more related to their adaptation to different hypogean environments. In the light of the geological, geographic and molecular phylogenetic data at hand for troglobitic species from the three Canary islands with richer hypogean fauna, we conclude that there is not a general pattern, and either factors related to the evolutionary time or different environmental selective pressures can be determinant in each case.

Oral presentation Red book of cave-dwelling fauna of Croatia

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Since their establishing in 2002 Year, Croatian State Institute for Natural protection systematically works on protection of biological and landscape biodiversity. One of important missions is publishing of Red books, obligate international law document. Till end of 2007 Year six Red books have been published and four more are in preparation, one of them Red book of Cave-dwelling fauna. Croatian cave-dwelling fauna belong to Dinarid Mt. channel, covering 50 pct of state territory, including three Dinaric geotectonic units: Adriatik, Dinarik and Supradinarik, with all three dinaric biogeograpic regions: North, Middle and South Dinaric. Especially is important and endangered Adriatik region, in general belong to Croatia including almost 6000 km east Adriatic coast with more than 1200 karstic islands. Except Dinaric, during recent biospeleological research in continental Croatia cave-dwelling fauna of sub Alpic region have been found same as cave-dwelling fauna of old Oriental land. Till end of 2007 Year from more than 200 caves more than 340 taxa have been described, almost all cave-dwelling animals. Every year new caves and taxa for science are discovered and many taxa descriptions are in preparation. In same time many causes which endangered cave-dwelling and fauna developed. They primary are: urbanization, habitats hidrotechnical and melioration grips, which physically destroyed cave habitats and their fauna; waste deposits and pollutions which contaminated cave habitats and finally pump out of drink water resources. State Institute for Natural protection has recognized importance and value of cave-dwelling fauna and have supported Red book of cave-dwelling fauna project. According to established criterions around 200 taxa will be included, many of them stigobitic. In same time this publication is bidding to other states, first of all richest in cavedwelling fauna, for publishing their Red book of cave-dwelling fauna according to concerted target, conservation of this world important heritage.

Oral presentation Subterranean homesick blues or: how I learned to stop worrying and love Australian cave shrimps

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There are eight genera of the freshwater shrimp family Atvidae in Australia. Of these eight genera, four are subterranean, and so provide an excellent opportunity to study the process of adaptation to caves. We have been working on the molecular phylogenetic relationships of the Australian and non-Australian taxa. Some of these relationships have proven to be complex and unexpected. The widespread cave genus Parisia (Australia, Madagascar, Philippines) appears to be polyphyletic, as one species (P. unguis) forms a strong clade with the Australian endemic monotypic hypogean genus *Pycnisia*. The other species of Australian Parisia (P. gracilis) forms a clade with yet another endemic monotypic cave genus, Pvcneus. Both sets of clades group with completely separate surface species of Caridina, thus implying an independent colonisation of the underworld in Australia and morphological convergence. The fourth hypogean genus, Stygiocaris, is native to Western Australia and is part of the unique cave biota of the coastal, anchialine, limestones of the Cape Range and Barrow Island, most of whose nearest evolutionary relations are found in coastal caves of the distant North Atlantic. Our nuclear and mitochondrial analyses all recovered the amphi-Atlantic cave genus Typhlatya as the sister taxon of Stygiocaris, rather than any of the numerous surface and cave atyids from Australia or the Indo-Pacific region. The two described Stygiocaris species were recovered as monophyletic, and a third, cryptic, species was discovered at a single site. Our findings suggest that Stygiocaris and Typhlatya may descend from a common ancestor that lived in the coastal marine habitat of the ancient Tethys Sea, and were subsequently separated by plate tectonic movements. The Cape Range's geological dynamism, which is probably responsible for the speciation of the various *Stygiocaris* species, has also led to geographic population structure within species, in particular Stygiocaris lancifera.

Oral presentation

Does bad taxonomy favour conservation purposes? The case of the eyeless *Cicurina cueva* (Araneae: Dictynidae) from caves in the vicinity of Austin (Travis Co.) Texas

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Urban development in central Texas is a threat to many habitats. especially caves. Sixteen cave-restricted arthropod species are protected by the U.S. Endangered Species Act, while many others are classified as species of concern. Cicurina cueva Gertsch is an eyeless spider known from only two caves in the vicinity of Austin (Texas). A proposition for a new highway threatens the ecological integrity of Flint Ridge Cave, one of the two known localities for this species. Correctly assessing the distribution and species limits of this taxon is crucial for any conservation decisions. An intense sampling effort resulted in the collection of Cicurina spp. from ~70 caves in Travis, Hays and Williamson counties. About 1kb of mtDNA (CO1) was sequenced for 170 spiders and the phylogenetic approach of Paquin & Hedin (2004) was used to assign species names to juveniles. Phylogenetic analyses (distance, parsimony, likelihood and Bayesian) gave similar results and extended the occurrence of C. cueva from two to ~20 adjacent caves. These results suggest that C. cueva, C. bandida and C. revesi are the same biological entity. Furthermore, spermathecal variation is not correlated with geography or mtDNA phylogeny, providing additional support for the synonymy. NCA analysis provides support for restricted gene flow, with both partial restrictions (isolation by distance) and complete restriction (fragmentation), suggesting a complex population structure as expected in cave systems. Inadequate taxonomy or lack of collections that artificially increases the biological uniqueness of caves or species rarity is not a sound basis for conservation purposes. Long-term strategies require adequate taxonomic basis and knowledge, which is still, unfortunately, largely deficient.

Oral presentation Evolutionary ecology of the Mexican cave fish *Poecilia mexicana* (Poeciliidae) compared to epigean relatives

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The Atlantic molly (P. mexicana) is a widespread freshwater fish living along the Atlantic coast of Mexico and Central America which colonized in southern Mexico two caves. One is the non-sulfidic Cueva Luna Azufre and the other one separated by the valley of the river El Azufre is the sulfidic Cueva del Azufre. Both caves drain into the El Azufre being a sulfidic surface habitat. Current molecular genetic data suggest that both caves were independently colonized from adjacent non-sulfidic surface habitats and harbors today genetically distinct populations being still in contact by migration. Till now only the population of the sulfidic cave has been studied in detail. Both cave populations have smaller but functional eyes, reduced body pigmentation and slender bodies. In the sulfidic population constructive traits have been demonstrated: a genital pad in the female, an increased number of taste buds on the head, a hyperdeveloped cephalic lateral line system and heavier more structured ototliths. This traits improve orientation and reproduction in the lightless habitat. Increased gill filament length, another progressive trait, facilitates in connection with aquatic surface respiration oxygen uptake in the sulfidic hypoxic environment. Besides morphological differences some behavioural ones have been found: reduction of shoaling and aggression, changes in male and female reproductive behavior. P. mexicana in non-sulfidic surface habitats primarily fed on detritus and algae. In the caves on the other hand invertebrates made up the majority of the gut content resulting in a shorter intestinal tract. The food web in the sulfidic cave appears to rely to some extent in situ chemoautotropic bacterial primary production. The main predator in the sulfidic cave seems to be the large aquatic hemipteran *Belostoma* preferring larger males over smaller ones and females of the same size. Life history consequences of our findings will be discussed in detail.

Poster presentation The cave-dwelling fauna of Gran Canaria (Canary Islands, Spain): the importance of artificial caves

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The Canary Islands are of volcanic origin and most of their caves are lava tubes, which are only present in basaltic layas with a maximum age of a few hundred thousand years. The origin and evolution of the Canary archipelago fits with a particular model of hot spot, in which the islands have no subsidence along time, and can have volcanic activity millions of vears after having been upon the hot spot, thanks to lateral plumes of the mantle. The cycle follows i) an initial fast growing of the island, ii) a volcanic guiescence with abundant terrain discharge by erosion, and iii) a post-erosive lesser activity that partially covers the island with a new layer of lavas. Lava tubes are abundant in the first and third stages, but almost absent in the second. Gran Canaria is between ii) and iii), and therefore lava tubes are very scarce. However, abundant artificial caves have been bored to get water, giving a possibility to find cave-dwelling fauna in areas without lava tubes. Our experience has shown that artificial mines can hold an adapted fauna if they are old enough to have stabilized their environment, and if they go trough rock layers (or MSS) with an important network of mesocaverns. Only three among the 130 adapted species known so far in the Canaries occur in Gran Canaria, all collected in the MSS. The results of two year sampling in all known lava tubes and in some artificial mines are provided. The studied material includes eight new species of the following genera: Troglohyphantes and Scotophaeus (Araneae), Pseudoniphargus (Amphipoda), Nicoletiidae n.gen. (Zygentoma), Lymnastis, Medon and Oromia (Coleoptera). It is worth pointing out that artificial mines were in general richer than lava tubes, maybe because the latter are usually dry in this island.

Oral presentation

Molecular phylogeny and new systematics of leptodirine subterranean beetles (Coleoptera, Leiodidae = Cholevidae): first results

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Leptodirine beetles, with more than 170 genera and about 800 species, show one of the greatest radiations among the subterranean biota. The subfamily has a palaearctic distribution with the exception of the enigmatic nearctic *Platycholeus*. Cave adapted species are restricted to the northern Mediterranean Basin and Balkan Peninsula. Species in other regions like Western and Eastern Europe, Caucasus, Middle and Far East are mostly edaphobiontes. Past attempts to understand the evolution and establish a phylogenetic system of the Leptodirine have used morphological characters. None of these attempts could satisfactorily explain the resulting morphological and biogeographical patterns. Our study included 55 leptodirine and three out-group genera mostly from the Dinaric Karst region, but also from the Pyrenees, Alps, Carpathians and southern Balkans. We sequenced about 3.3 kbp from two the mitochondrial (COI and 16S) and two nuclear gene segments (28S rDNA, Histone H3), and analyzed them using standard phylogenetic procedures. Phylogenetic trees from different loci recovered a monophyletic origin of the studied leptodirines, and showed similar phylogenetic relationship among so far unrecognized clades that can mostly be defined geographically. Morphological characters such as body shape, number of male protarsomeres, male protarsi dilatation, antenna insertion position, length ratio of the first two antenomeres, presence of tibial spurs, spines and comb, and mesosternal carina shape, seem to be homoplastic and therefore not useful for the tribal systematic of leptodirine beetles. Most of the traditional tribal and subtribal taxa established on the grounds of these homoplastic morphological characters have to be rejected and replaced by a new subdivision based on the new clades and their supporting apomorphic morphological characters. The structure of the male edeagus internal sac, already used by Jeannel, Laneyrie and al.. with emphasis on the Supraflagellates Giachino et and Infraflagellates, provides a potential source of such characters.

Oral presentation The genetic basis of morphological change in the cave isopod, Asellus aquaticus

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We are interested in studying the genetic basis behind how cave characteristics evolve. The system we are using, the isopod, *Asellus aquaticus*, has both cave and surface dwelling forms. Though these forms can interbreed, they are morphologically dissimilar. The cave form is unpigmented, has no visible eyes, has less bristles, and has longer appendages. These morphological differences and the ability to interbreed the surface and cave forms make this a tractable system in which to study the genetic basis of cave adapted traits. We have been attempting to raise *Asellus aquaticus* in the lab and have mated the cave form with the surface form. Ultimately, we would like to cross these hybrid individuals to each other to generate a cross for genetic mapping. We are also comparing the coding sequence of candidate genes in eye and pigmentation development between the cave and surface forms.

Oral presentation Methane subsidy and stonefly migration in an alluvial aquifer ecosystem

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The discovery of subsurface stonefly nymphs in highly permeable floodplain sediments kilometers from the river channel in western Montana (USA) in the 1970s dramatically demonstrated the large spatial extent of river-groundwater interaction. The geographic range of the hyporheic stoneflies Paraperla and Isocapnia implies a wide distribution of similar alluvial aguifer ecosystems in western North America. New evidence sheds some light on energetic basis of these ecosystems, and may explain the migration of large, long-lived stoneflies into otherwise resource-poor groundwater. A floodplain scale network of monitoring wells was sampled at 1 meter depth intervals over the annual cycle, in the Nyack flood plain, Montana, USA. Invertebrate density varied over 5 orders of magnitude in the top 1 meter interval (<1 to $6 \times 10^5/m^3$ sediment), with the highest invertebrate densities at DO < 2 mg/l. Density declined exponentially with depth, however wells with orthograde oxygen profiles (low DO in the surface interval) had an exponential increase in density at the ecotone between low and high oxygen. The small size classes of the amphibiont stoneflies were almost exclusively collected at this oxycline. Invertebrate abundance peaked in July or August at most sites, but the oxycline showed a peak in September, corresponding with the oxygen minimum. Stable carbon isotope signatures showed strong levels of depletion among most taxa $(\delta^{13}C - 25\%$ to -70%). This signature was conserved in adult and sub-adult stoneflies collected during emergence in more oxygenated sites near the river. These observations indicate the energetic importance of hypoxic zones, with methane-derived carbon alone providing 10 to 90% of invertebrate energy flow. It also implies a positive energy return on investment for amphibiont stonefly migration, with the energetic subsidy from hypoxic zones being sufficient to offset the cost of migration and adaptation to low oxygen.

Oral presentation Origins and distribution of the family Nesticidae (Arachnida: Araneae) in the Mediterranean basin

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Nesticids are represented worldwide by slightly more than 200 species grouped in 9 genera. This family is widely spread across the karstic reliefs of the Mediterranean basin providing a high number of endemic species. All of them are cave dwellers with only a single Rumanian exception (Carpatonesticus lotriensis). The aim of this study is to infer the origin, colonization pathways and phylogenetic relationships of the Mediterranean species of this family in a phylogenetic framework. Species analyzed in the present study comes from all southern European peninsulas. Some representatives from Turkey and Crete Island were also included. Chinese Nesticus sp. as the most distantly related species within the family was also included and Steatoda sp. (Fam. Theridiidae) was used to root the trees. 2000 bp from two mitocondrial (COI & 16S) and one nuclear gene (18S) have been sequenced. Parsimony analyses of the combined data matrices were performed with the program Winclada v.1.00.08. Clade support was assessed via Bootstrap. Preliminary results of the phylogenetic analysis indicate that, at least, 3 independent colonization processes from eastern Mediterranean have occurred, giving rise to a high number of endemic species in the main European southern peninsulas. The results show that specific radiations that have basically contributed to the high level of endemicity of this family, have been relatively recent phenomena (Carpathian clade & West-Balkanian clade). while ancient groups (C. quadridentatus, Relict clade & Iberian clade) hardly present specific radiation processes. These data together with the absence of Nesticid species in North Africa suggest that several extinction independent phenomena may have occurred in the past. Probably the origin of the canarian species (C. quadridentatus) and the relict clade was previous to the glaciations period. The high value of the genetic distances between these species allows us to consider them as climatic relicts.

Poster presentation Biodiversity of stygofauna and stygomicrobes of aquifers in South Australia in relation to environmental factors

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Until recently little was known about the biodiversity of aquifers in South Australia. Knowledge of aquifer biodiversity is crucial to ensure management of groundwater, particularly responsible during anthropogenic alterations of aquifer systems as occurs during aquifer storage and recovery. During the last year, we conducted systematic sampling of groundwater from observation bores, wells, springs and caves in South East South Australia, Mount Loft Ranges, Evre Peninsula and River Murray. Aquifers from various geological histories were sampled including limestone, fractured rock and alluvial aquifers. Stygofauna was sampled in bores using plankton nets and pumped from sediments using the Bou-Rouche method. This revealed a wealth of new stygofauna, with new species belonging to Amphipoda (Bogidiellidae, Chiltonidae, Melitidae, Neoniphargidae), Isopoda (Heterias), Syncarida (Anaspidacae, Bathynellacae), Gastropoda (Hydrobiidae), Copepoda (Cyclopoidea, Harpacticoidea). Ostracoda and Oligochaeta. Flow cytometric analysis of water samples established a wide diversity of abundance and size classes of stygomicrobes (bacteria and virus like particles). In addition, field parameters, such as total dissolved oxygen, conductivity (~salinity), pH and temperature were recorded. Bore parameters (depth, construction and casing type) and aquifer characteristics (confined or unconfined) were also noted. Using multivariate analysis we investigated how the presence of stygofauna and stygomicrobes is related to these environmental factors. This allows for better predictions of groundwater biodiversity.

Poster presentation To colonize subterranean environment without starvation tolerance capacity: the case of fish Astyanax fasciatus mexicanus

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A fish species, Astyanax fasciatus, exhibits both subterranean and epigean populations. The subterranean populations came from tropical caves characterized by a potential high level of nutriments. In general, subterranean environment is characterized by low food level inducing fasting adaptations like hypoactivity, hypometabolism and/or sequential use of energetic reserves. It is commonly accepted that such adaptations allow subterranean fauna to tolerate longer fasting than epigean one. This study aimed to display behavioral, metabolic and biochemical responses of a fish species (Astvanax fasciatus) during a long fasting (2 months) followed by a re-feeding period (12 days). Our results demonstrated a higher potential for fasting tolerance for the hypogean fishes compared to epigean ones (higher energy reserves and hypometabolism at fed state). But surprisingly, the hypogean fishes consumed significantly more glycogen and protein during the starvation period than epigean fishes (respectively 91% vs -30% and -21% vs 0%) and despite drastic decreases in locomotory activity (-40%) and oxygen consumption (-29%). Furthermore, the hypogean fishes activated significantly more compensatory metabolic pathways such as cetogenesis and neoglucogenesis. After 12 days re-feeding, in contrast to hypogean fishes, epigean ones restored their entire glycogen and protein reserves representing an important functional advantage. In conclusion, Astvanax fasciatus mexicanus appears as a paradox demonstrating that a high starvation tolerance is not a compulsory trait to subterranean environment but is correlated to the trophic resources availability of the biotope.

Poster presentation

The aquatic fauna of the karstic cave system 'Piani Eterni' (Dolomiti Bellunesi National Park, Southern Limestone Alps, Italy)

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The results of a three years survey on the groundwater fauna of the karstic cave system "Piani Eterni" in the eastern Italian Alps is reported herein. "Piani Eterni" is a conspicuous karstic plateau located at 1880 m a.s.l. inside a National Park (Dolomiti Bellunesi). The main cave system (named PE10) currently includes over 17 km path length, with a maximum depth of 966 m below entrance. The top 100 m are occupied by snow and ice: beneath the percolating waters form gours, streams, and small lakes; the drainage system includes several springs at an elevation below 500 m a.s.l. Four surveys were carried out during 2004-2007 sampling the different aquatic microhabitats. The groundwater fauna includes nematodes (11 species), polychaetes (1), oligochaetes (8), copepods (6), amphipods (1) and dipteran larvae (2). Among Nematoda, three species are known from groundwaters only. It is possible that Theristus sp., new for Science, is also a stygobiotic species. The other nematodes are known as common in several kinds of freshwater environments. The Oligochaeta include at least three stygobiotic species; one of them belongs to the genus Rhyacodriloides and is new to Science. *Rhyacodriloides* is an ancient naidide genus, known until now with only two species from Lake Baikal. Among Copepoda, three species the genera Lessinocamptus, Bryocamptus and Speocyclops are new to Science; four species out of six are stygobiotic. Amphipoda are represented by a single species, Niphargus similis, which is endemic to alpine and pre-alpine caves in the Southern Limestones Alps, even at high altitudes. The importance of Diptera in this karstic system is very noteworthy. In particular, many larvae of the chironomid Eukiefferiella gr. gracei were found in every kind of groundwater bodies and in a spring, draining part of the cave system, located about 1,000 m below the sampling sites.

These findings allow this chironomid to be considered as a "biological tracer" of the watercourses within the system. The high percentage of stygobiotic species (38%) is noteworthy, because this area was completely covered by the alpine glaciers during the Pleistocene. The recolonization of the cave system by stygobionts following the retreat of glaciers, or their survival in the deep groundwaters during Quaternary glaciations, may explain this unusually high stygodiversity.

Poster presentation Food web in a cave from northeastern Brazil

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Studies on food webs in caves are scarce, especially those integrating terrestrial and aquatic communities. The present study was carried out in Gruna do Enfurnado, Serra do Ramalho karst area, NE Brazil, a 8,500 m long cave, with a considerable diversity of microhabitats and crossed by an autochthonous stream at its distal end and an allochthonous stream at the proximal end. These streams connect during flash floods, when large amounts of organic matter, mainly vegetal debris, are deposited in the cave, forming the basis for the food webs. A large population of troglobitic catfish. Rhamdia ernfurnada. lives in both streams. Oualitative and quantitative faunistic surveying was done using transects and quadrats. The diet of R. enfurnada was determined with basis on the analysis of stomach contents of preserved samples; for invertebrates, it based on direct observations and data from literature. was Representatives of 72 invertebrate taxa, most identified at family or genus level, were sampled along two years (three visits per year). These taxa were attributed to three trophic levels: the level of detritivores, mainly oribatid mites, collembolans and *Pseudonannolene* diplopods, the level of omnivores, with Endecous crickets, Eusarcus harvestmen and Camponotus ants, and that of strict predators, mainly spiders (Theridium, Loxosceles e Isoctenus) and heteropterans. R. enfurnada is an opportunistic feeder, with a varied diet composed by 17 % of aquatic items, 25 % of terrestrial items and 58% of particulate organic matter. possibly ingested when the fish foraged at the bottom, grubbing the substrate. The percentage of terrestrial items for *R*. *enfurnada* is higher than the observed for epigean congeners and also for other troglobitic catfish such as *Pimelodella kronei*. Due to its large population density, *R*. enfurnada represents an important trophic link between terrestrial and aquatic communities.

Poster presentation Phylogeography of two putative ancient asexual ostracod species (Darwinulidae, Ostracoda, Crustacea) from above and below ground

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A preliminary analyses of the phylogeography of two putative ancient asexual ostracod species, Vestalenula marmonieri and Vestalenula mathilda, was conducted with DNA sequence data from the mitochondrial COI gene. We wanted to test whether there is a geographical separation between populations from below as compared to populations from above ground. Alternatively, specimens from subterranean localities might be less affected by UV and, consequently, be better protected against mutations. In general, V. marmonieri shows much more variability below ground than V. mathilda, which is more typical of interstitial habitats. Considering the phylogeographic patterns. some division is found in V. mathilda as the specimens from the two investigated localities below ground are genetically rather similar whereas the ostracod from above ground is more distant. In V. marmonieri, on the other hand, no clear separation is observed. Furthermore, we observe genetic variability within and between subterranean populations of V. marmonieri. These results provide evidence against our hypothesis that these populations might be better protected against the accumulation of mutations.

Poster presentation A new hotspot of subterranean biodiversity in the Iberian Peninsula. The beasts in the caverns of the "Middle Earth" where "Gollum"-japyx dwells

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Two years ago, in a small karstic region of the Iberian Peninsula's western side, a mysterious japigid diplurian, Gollumjapyx smeagol Sendra & Ortuño, 2006, was described. It turns to be the species with the most extraordinary troglobiomorphic features of the whole Japigidae family, which are characterized by both the body slenderness and the legs length. However, G. smeagol is only the tip of the iceberg of a subterranean environment in a region where a lot of troglodytic and stygobiont forms live: a narrow Mesozoic limestones strip, barely 20 km wide, made up of limited altitude mountain ranges (300 to 500 meters above sea level) parallel to the line of the coast, and separated by graben type valleys (tectonic basins) all along the 80 km coast between Castellón and river Ebro's mouth. We are talking about a geostructural region known as "Coastal Ranges". We can find a total of 34 species, including 30 troglobionts and 4 stygobionts (5 macropredators, 13 micropredators and 16 fungivore and detritivore elements), all of them endemic of the "Coastal Ranges". Among these taxa, eight of them are unique and unparalleled genera: the driptinae carabid *Ildobates neboti* Español, 1966, the anillini carabid Iberanillus vinvasi Español, 1971, the japygid dipluran Gollumjapyx smeagol, the campodeid dipluran Paratachycampa hispanica Bareth & Condé, 1981, the araneid Speleoharpactea levantina Ribera, 1982 and the cirolanid isopodan Kensleylana briani Bruce & Herrando-Pérez, 2005. Moreover, two new genera (in process of description) will soon be added to this list. All of them are relictic elements, relics of a past related to the Iberian Peninsula's archaic isolation. The "Coastal Ranges" subterranean environment is made up of small caves hardly 100 m long. Inside these caves, both a high humidity and a temperature between the 14 and the 17°C (according to altitude) are kept. Among the caves showing a greater number of strictly cave-dwelling elements, "Cova dels Encenalls" (15 troglobionts) and "Avenc d'En Serenge" (14 troglobionts) are remarkable.

Oral presentation

Biogeographic significance of the recent discovery of a new species of *Stygobromus* (Amphipoda, Crangonyctidae) from groundwaters in Eastern Siberia near Lake Baikal

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Recent biological investigation of groundwaters near Irkutsk in the vicinity of Lake Baikal in eastern Siberia has resulted in the collection of a new species of Stygobromus from two springs. Prior to this discovery, only three species of this genus were recorded outside North America in the Palearctic region. Two were described from single sites in Asia east of the Ural Mountains and the third from a single site just west of the Caspian Sea. Despite the apparent rarity of these species and the fact that two of them are based on inadequate descriptions, they nevertheless established the presence of Stygobromus outside the Neartic region. Otherwise, the subterranean amphipod genus Stygobromus is widespread in the groundwaters of North America, where it is represented by 132 described species and as many as 50 provisionally recognized but undescribed species. The locality of the new species occurs approximately 900 km east-northeast of the site for Stygobromus pusillus, which is recorded from Lake Teletskove in the Altay Mountains. The third Asian species, S. kazakhstanica, was described from a spring in the region of Chernorechenskaya Cave northeast of Bishkek (formerly Frunze) in Kyrgyzstan. The new species from near Irkutsk does not appear to be morphologically closely allied with any other species in the genus Stygobromus, although this needs further detailed study. The widespread occurrences of three species of Stygobromus in the Palearctic and the fact that many areas in this vast region have not been carefully investigated for groundwater faunas strongly suggest that additional species of this genus will be discovered there in the future.

Oral presentation Evolution of Hawaiian surface and cave adapted Caconemobius crickets (Insecta, Gryllidae, Nemobiinae)

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Endemic crickets in the genus Caconemobius occur in moist bare-rock environments throughout the Hawaiian Islands, including coastal wavesplash zones, barren lava flows, wet cliffs, caves and mesocaverns. This group shows multiple species shifts to underground habitats, and provides a case study to test hypotheses of adaptive shift (sympatric/parapatric) versus physical isolation (allopatric) evolution. Species have been characterized based on morphology, chromosome differences and habitat. Current work on mtDNA and AFLP analysis is giving a clearer picture of the genetic variation within and among species. One presumed ancestral species, C. sandwichensis was believed to occur in the coastal wave splash zone of all the main Hawaiian islands, but mtDNA indicates two species on Hawaii Island (east side and west side) and a third from all the other main islands. Two lava flow surface species occur on Hawaii Island, C. fori on Kilauea Volcano and C. anahulu on western Mauna Loa. MtDNA comparisons show that these species are extremely close to the two Hawaii Island maritime species respectively, supporting the adaptive shift hypothesis. One cave species (C. howarthi) occurs on Maui and four or five occur on Hawaii (C. varius, C. uuku, C. albus, C. paralbus and at least two undescribed). MtDNA shows the Maui species to be distantly related to the surface species, while the Hawaii Island species show a greater separation from the surface species with a "genetic clock" distance of 6-7 million years (Hawaii Island is less than one million years old). C. howarthi, C. varius and C. uuku have drastic chromosomal reorganization in comparison with the surface species and with each other, showing the possibility of more rapid genetic and chromosomal changes than projected by the standard genetic clock. Further tests are needed to resolve the adaptive shift versus isolation hypotheses for the cave dwelling species.

Poster presentation

Current status of the family Nocticolidae (Insecta, Blattaria); a tropical Old World and Australian group rich in cave adapted species

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The cockroach family Nocticolidae is distributed through the Old World tropics and subtropics from Africa through Australia. Species inhabit damp forest litter, moist rocks, caves and mesocaverns. Cave adapted species occur throughout the area occupied by Nocticolidae, ranging from species with reduced pigment, eyes and wings in shallow cave and mesocavern habitats to highly troglomorphic pigmentless, eveless and wingless species in deep cave and bad air (high CO₂) cave zones. Eight genera and over 30 species have been described, with nearly twice that number remaining to be described. The Nocticolidae show many characters that are different from other cockroach families including an erect posture, head rotated forward, forewings similar to hindwings, wings held outward from the body at rest and highly complex male genitalia. DNA and microscopy studies of Australian species show an absence of the endosymbiont *Blattabacterium cuenoti*, present in all other cockroaches currently studied. Recent DNA studies have placed the Nocticolidae as an early branch of the cockroach order, but its placement in relation to other cockroach families is still ambiguous. The Nocticolidae are a highly promising group for studies of evolution and ecology of cave adapted species. Recent successful rearing of a colony of the partially cave adapted species Nocticola australiensis has provided the first information on the life cycle and reproductive behavior of this group. Further DNA comparison of surface and cave adapted Nocticola species should offer an excellent opportunity for testing adaptive shift (sympatric/parapatric) versus climate shift (allopatric) hypotheses of cave adaptation.

Oral presentation Introgression in Astyanax fasciatus cave populations (Teleostei)

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In Northern Mexico about 25 cave populations of the neotropical freshwater fish Astyanax fasciatus have evolved, each of which derive from at least two independent invasions of surface fish from the south. These northern Astyanax populations can be separated into two groups: A phylogenetically old one consisting of morphologically very similar strongly eye and pigment reduced (SEP) cave populations, e.g. Sabinos and Yerbaniz, which descended from an ancient invasion. The other group is heterogeneous and comprises surface as well as cave populations with variable eve size and pigmentation (VEP), e.g. Micos, Caballo Moro and Chica. This group also includes several SEP populations like Pachon showing complete or nearly complete replacement of their original mtDNA haplotype due to introgressive hybridization. In some caves the co-occurrence of cave and surface fish was observed, suggesting that the morphological variability of eyes and pigmentation in VEP populations was due to ongoing hybridization between old SEP and surface fish. In contrast, the morphological variability of eyes and pigmentation may be explained by a more recent origin of these populations. To distinguish these hypotheses we performed a molecular genetic study using mtDNA cytochrome b sequence data and six microsatellite loci. The F_{ST} -values from the microsatellite loci indicate that no gene flow has occurred between almost all populations studied, including the cave populations like Micos (VEP) and Yerbaniz (SEP) in which the existence of surface fish was observed. Also in Caballo Moro (VEP), where two cavefish groups - a large and a sunken eved one - co-exist, it was found that these can be separated. This study reveals that the VEP cavefish (Micos, Caballo Moro and Chica) are phylogenetically young, sharing the same ancestor with the recent surface populations. Therefore, the morphological variability of eyes and pigmentation is most likely due to their recent and incomplete evolutionary transition. In contrast, the hybridization involving SEP populations like Yerbaniz and Pachon is probably an ancient event.

Oral presentation Discovery of subterranean 'Hidden Treasures' within the Northern Agricultural Region, Western Australia

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The Hidden Treasures Project studied the biodiversity of the Northern Agricultural Region (NAR). Caves and their associated biodiversity are one type of natural asset being considered, and the 2007 Subterranean Biodiversity Inventory is a key component of the Hidden Treasures Project. The project aimed to collect information to allow the highest priority conservation areas in the NAR to be identified. It is anticipated that this prioritisation would lead to the development of a Biodiversity Conservation Plan for the NAR. The study has been funded by the Northern Agricultural Catchment Council (NACC) and delivered by the Department of Environment & Conservation in association with the Australian Speleological Federation (ASF). This paper seeks to summarise some of the key findings of the project in a context that is significant to the management and conservation of these priority areas. The regional areas that will be addressed include Eneabba, Jurien, South Hill River, East Moore River and the Moora caves. Fauna sampling within caves focussed on invertebrate populations and revealed a diverse assemblage of organisms. This new information (in conjunction with historical accounts) has provided an insight into the ecological condition of these karst areas. Future management and conservation of these cryptic ecosystems will draw upon this increased knowledge.

Oral presentation Subterranean terrestrial isopods (Crustacea, Oniscidea) from Western Australia

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To date 26 species of terrestrial Isopoda have been recorded from subterranean habitats in Australia, 21 of which are from Western Australia. Most of these species are trogloxenes or troglophiles, and only Abebaioscia troglodites from caves in Nullarbor region is a troglobite, and Haloniscus longiantennatus, H. tomentosus, H. stilifer and H. pedisetosus from calcrete aquifers in the Yilgarn Region are stygobites. A large collection of terrestrial isopods from karsts and calcretes from Western Australia (Pilbara, Cape Range, Nambung, Augusta, Yilgarn and Nullarbor areas) has been examined and 28 new species of troglobitic and stygobitic Oniscidea have been recognised: one species of Styloniscus (Styloniscidae) and one species of Adoniscus (Olibrinidae) from the Pilbara; three species of a new genus of Stenoniscidae and 13 species of Haloniscus (Philosciidae) from the Yilgarn region; two species of Hanoniscus (? Oniscidae) from Cape Range and Nullarbor, two species of Laevophiloscia (Philosciidae) from caves in Nambung and Augusta areas; one species of Buddelundia (Armadillidae) from Nullarbor; and five species of *Troglarmadillo* (Armadillidae) from Cape Range, Pilbara and Yilgarn. All these species show troglomorphic characters, being blind, colourless and often with long and frail appendages. The new species of Haloniscus, each restricted to single calcrete aquifers in the arid Yilgarn region, confirm morphologically the diversification of this genus already demonstrated with molecular analyses, and support the "subterranean island" hypothesis. The discovery in the Yilgarn calcretes of three new species belonging to a new genus of Stenoniscidae, related to the littoral genus Metastenoniscus known from one species on some Indian Ocean islands, and one species from Venezuela, and the presence of a species of Adoniscus, a genus previously known from Sulawesi, Kenya, Socotra Island, Lebanon and Morocco, seem to demonstrate the affinities of this fauna with the Tethys.

Oral presentation

Urban and rural cave invertebrate communities: Isotopic evaluation of trophic structure (δ^{13} C, δ^{15} N) & the relationship between taxa richness and cover types in central Texas, USA

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The energy regime in small Texas (U.S.A) caves differs significantly from many caves of the better studied eastern United States in that surfaceforaging cave crickets (Ceuthophilus spp.) are major contributors to these systems. The federally listed endangered cave invertebrates of central Texas are dependent on these crickets to transport energy from the surface to the cave environment. Using stable isotope analysis in combination with in-cave counts of animal life we examined cave invertebrate communities in nine caves chosen based on their low, medium, and high levels of human impact. Surface foraging cave crickets do not utilize the same food resources as the invasive red imported fire ant (Solenopsis invicata). The trophic position of the entire cave invertebrate community differed significantly between all three levels of human impact, for both δ^{13} C and δ^{15} N. Numbers of individuals of all cave taxa, including *Ceuthophilus* spp., are correlated with the level of human impact. As the percentage of impervious cover and percentage of impacted area increased, the total number of cave taxa decreased. This trend held true when either 11.2 or 90 acres around the cave entrance were considered in scoring the level of impact. Additionally, the total number of individuals of other taxa recorded from the caves was strongly correlated with the total number cave crickets. Maintaining land in a natural state within the foraging range of cave crickets (C. secretus and C. species B), and controlling the fire ant, S. invicta, are therefore important considerations in the management of Texas' federally listed endangered cave invertebrates.

Oral presentation Using subterranean science in water planning

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The gathering pace of water reform in Australia and internationally demands more urgent delivery of the science necessary to underpin water planning and achieve sustainable and integrated water management. Legislative and policy frameworks to mandate the use of science in water management are in place and water planning is well advanced across Australia, following guidelines set out by the National Water Initiative (NWI). Yet the first review of the NWI found that progress on integrated management of environmental water had not been achieved as envisaged due to inadequate specification of the desired outcomes and shortcomings in the available science. But at the same time researchers repeatedly lament that management approaches fail to recognise fundamental scientific principles. This paper argues that poor uptake of science in water planning is not due to a paucity of science but to imprecise specification of the knowledge needs of water management, resulting in a disjunction between research and its application. Questions derived from water policy are not framed as testable hypotheses that could form the basis of targeted research programmes, and research results lack direct application to water management. A case study is presented of an investigation of the environmental water requirements of an alluvial aguifer connected to a regulated river, and management actions and research questions suggested which could deliver knowledge applicable to an integrated management plan for the alluvium and river. Recasting of policy knowledge needs as specific research questions is offered as a means to better bridge the science/policy divide.

Oral presentation Speleonectes sp. (Crustacea: Remipedia): habitat, abundance and trophic relations in "Crustacea" anchialine system, Quintana Roo, Mexico

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Anchialine ecology is needed for both decision-making and conservation strategies establishment of these vulnerable ecosystems. The Mexican Caribbean coast requires special attention due to explosive urban expansion in response to tourism development. Speleonectes sp. in the "Crustacea" cave shows higher abundance compared to other known sites. We present the results of the "Crustacea" cave assessment. Four sampling stations (A to D) were sampled along a 279 m transect. The water column was characterized by using a multiprobe profiler and discrete water sampling at three depths. Dissolved inorganic nitrogen, phosphorus, particulate organic matter (as %C, %N, δ^{13} C and δ^{15} N) were measured among other factors. Superficial sediment was collected to determine grain size and organic matter content (%C, %N, δ^{13} C and δ^{15} N). Speleonectes sp. abundance was evaluated fourfold along the transect. Fauna was sampled, identified and analyzed for stable isotopes ($\delta^{13}C$, δ^{15} N). Water column is salinity-stratified (10 to 35.7 g/L) dysoxic (2.54 to 0.2 mg/L DO) and pH changes from 6.94 to 6.48 at mixing zone. The nutrient concentration is low (< 4 μ M). The C/N_a ratio of the particulate organic matter increased with depth in the water column (7.23 to 8.17)related to remineralization. The sediment is carbonaceous silt (~70%), rich in organic matter content 16.82-26.75% C, 1.62- 2.62% N with a C/N_a ratio 11.96-12.09 implying nitrogen limitation in the sediment-water interface. In a single dive a total of 243 remipedes were counted along the transect, mostly aggregated at stations C and D (Xi^2 =13.8, p-value < 0.05) indicating site preference. Remipedes are in the upper trophic level $(\delta^{13}C - 35.27 \pm 1.72 \% (n=8), \delta^{15}N 6.71 \pm 0.97 \% (n=8))$ consuming shrimps $(\delta^{13}C-36.29\pm4.66 \% (n=23), \delta^{15}N1.93\pm2.95 \% (n=23))$ and hadziid amphipods (δ^{13} C-38.05±7.05 ‰ (n=2), δ^{15} N0.26±3.73 ‰ (n=3)). The potential food supply for shrimp and amphipods derives from films and particles with a signal of allochthonous and remineralized production under hypoxic conditions ($\delta^{13}C < 35 \%$ and $\delta^{15}N < 5 \%$).

Oral presentation Ecological scenarios in the evolution of troglobites

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Genetic models of evolution of troglobitic taxa generally take into account very simplified population and trophic ecological scenarios. I present two plausible scenarios for evolution of subterranean populations based respectively on a cyclical (paleoclimatic) and a non-cyclical (topographic) model of allopatric differentiation, with fishes as model taxa. Both depart from troglophilic populations of unknown sizes (a great variation is observed in the field). Within the paleoclimatic model, the following sequence of events is possible: 1st step: Favorable climate (wet phases) - Colonization, with establishment of troglophilic populations; 2nd step: Unfavorable climate (dry phases) - Isolation due to local populations (Barr´s extinction of epigean model): Habitat fragmentation/decrease + 1 food availability (due to decrease of nutrient input) \Rightarrow population size $\downarrow \Rightarrow$ accelerated differentiation (bottleneck effects) \Rightarrow adaptations to increase efficiency in the subterranean habitat \Rightarrow population size \uparrow ; 3rd. step: Re-establishment of conditions of less food limitation - Total population $\uparrow\uparrow$, but: reproductive rates \downarrow (K selected life cycle) \rightarrow effective population size \downarrow . The topographic model is based on a progressive alluvial downcutting leading to the fragmentation of an Nsized population into an increased number of smaller ones, due to presence of high waterfalls. The bottleneck hypothesis also applies to these segments, but population dynamics differs: upstream populations would be subject to important emigration but negligible immigration rates, thus higher differentiation rates are expected in comparison with downstream populations, which would receive emigrants from the former. The current population sizes of troglobitic species are determined by a combination of historical patterns and present-day ecological factors, mainly habitat and food availability as perceived by the organisms. Therefore, much caution is need when using these data in genetic models.

Population ecology of *Rhamdia enfurnada*, troglobitic catfish from northeastern Brazil (Siluriformes: Heptapteridae)

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Rhamdia enfurnada, characterized by a high individual variation in reduction of eyes and pigmentation, is known from a single cave in the Ramalho karst area, NE Brazil. Serra do Lapa do Enfurnado, approximately 5,000 m long, is crossed by two streams, with a seasonal input of nutrients, mainly as vegetal debris, carried into the cave during floods and supporting a large population of catfish. The population ecology of R. enfurnada was studied by mark-recapture technique from July 2005 to August 2008. Eight hundred and fifty-four individuals out of 1024 captured were marked and released, of which 153 were recaptured at least once till September 2007; the remaining 169 catfish were either released with mark (too small for marking) or collected for laboratory studies, including genetics, behavior, physiology of pigmentation and diet. All captured specimens were measured (standard length), weighed and the condition of eves and pigmentation was noted. Population size and densities (reaching a maximum of 10 ind.m⁻²) are quite large for cave fish standards. The catfish are relatively sedentary, staying for several months or even years in 100 m long stream reaches; no individual was captured in a stream different from the one where it was marked. Growth occurs is pulses, with growth rates in rainy seasons (in average, 1.9 mm month⁻¹) significantly higher than in dry seasons (0.14 mm month⁻¹) ¹, with many cases of negative growth). Lifespan is also high, with large individuals, up to 250 mm standard length, estimated as being 30+ years old. For the population living in the stream at the distal end of the cave, reproduction takes place in a deep, slow-moving, soft-bottomed lake and recruitment occurs upstream, towards shallower reaches. These catfish are less active than, for instance, Pimelodella catfishes, with longer resting phases.

Oral presentation Adaptive radiation under ground

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The classic view of an evolving troglobiont is that of a linear temporal succession of adaptive and/or neutral phases leading from exapted via slightly troglomorphic to highly troglomorphic. A number of molecular phylogenetic and phylogeographic studies have supported this notion by demonstrating that several closely related subterranean species arose from independent invasions and parallel morphological transformations to troglomorphism. Results from recent molecular phylogenetic and phylogeographic studies, however, suggest that in some areas of the Dinaric Karst subterranean ancestral species gave rise to two or more likewise subterranean species living in the same area (cave) but diverging ecologically. Such cases are provided by leptodirine cave beetles, where independent, geographical clades contain generalist endogean forms together with moderately and strongly troglomorphic species in addition hygropetric filter feeders. specialized cave The stygobiotic to counterpart is a clade of at least four genetically very similar Niphargus (Amphipoda) species from Cave Vietrenica (Bosnia Herzegovina) differing considerably in morphology, feeding behavior, and microhabitat. These findings come close to the definition of adaptive radiation (AR), e.g. by P.R. Grant: "The evolutionary diversification of a single lineage into a variety of species with different adaptive properties". Additional restrictions for such an event to gualify as "subterranean AR" are that (1) the last common ancestor has to be subterranean and (2) it has to take place in an area of contiguous subterranean habitat to preclude the possibility of allopatric speciation. This hypothesis sees interspecific competition as a major driving force leading to troglomorphic adaptation. It follows that different degrees of troglomorphy are not simply steps on a linear evolutionary ladder, but rather morphological signs of niche differentiation in the subterranean habitat. Subterranean AR is the most advanced and independent level in the evolution of subterranean biota, when troglobionts through cladogenesis give rise to new, functionally differentiated troglobiotic species, independent of surface ancestors, climatic relics or adaptive shifts.

Poster presentation Evolutionary biology of cave shrimps (Crustacea, Decapoda, Atyidae) from the Indonesian island Sulawesi

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The evolution of troglobites has fascinated scientists since Darwin's time. A high number of cave-dwelling animals are well studied today, among these several freshwater organisms, e.g. fishes or crustaceans. The freshwater shrimp family Atvidae (Crustacea, Decapoda) also has several subterranean representatives worldwide. But whereas the troglobitic atvids from Australia and Europe have already been comprehensively studied, such data for Southeast-Asian shrimps are still lacking. From the Indonesian island Sulawesi, situated within the biogeographic hotspot area Wallacea, currently 36 species in three genera are known, approx. fifty percent of them endemic. Two genera (Caridina and Atvopsis) only have epigean representatives, while the third and endemic genus Marosina exclusively occurs in subterranean rivers in the southwestern part of the island. So far, this genus only comprises two just recently described species, M. longirostris and M. brevirostris. Comprehensive new collections of both species from several caves of Sulawesi are studied here in detail with morphological and molecular methods as for example already done for the speciose and epigean genus Caridina from Sulawesi. We present data for the subterranean as well as the epigean shrimps in order to derive general patterns of cave colonization and subterranean evolution.

Oral presentation Genes, modules and the evolution of troglobites

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In the 1950's Curt Kosswig discovered the extraordinary importance of the interfertile cave and surface populations of the neotropical characid Astvanax fasciatus (=A. mexicanus) (Teleostei). Several constructive and regressive traits were changed during adaptation from surface to cave life. Genetic analysis of these traits in passing between cave and surface fish revealed that the inheritance was a multiple exhibiting all intermediate stages. Characteristically, each polygene acts quantitatively and provides the same amount of phenotypic expression. A threshold is surpassed after recombination of a minimum number of polygenes leading to an abrupt change of trait size. This process of inheritance occurs in both regressive and constructive traits. The eye provides the possibility to analyze both regressive and constructive evolution of complex traits. Both phylogenetically young cave fish (e.g. Chica and Micos), and F2-crossings between cave and surface fish, exhibit variability in eve size and also well developed lenses combined with undeveloped retinae, and vice versa. This indicates that these two subunits inherit independently reflecting ontogenetic development. Sequence data of a large variety of structural and control genes (e.g. crystalline, pax6, shh) in cave fish mostly do not deviate from those in surface fish. This is concordant with the observation that the large majority of intermediate eve stages in the crossings contain all structures and some are even functional. Therefore, it is deduced that the "eve genes" are upstream top control genes, which determine the size of the first eve primordia. Crossing analyses revealed no genetic linkage between the regressive eye and several constructive and regressive traits (e.g. taste buds, aggressive behaviour, pigmentation). Independent of being regressive or constructive the different traits can be considered separate units of development and evolution, so-called modules. They follow the same mode of inheritance and are also, like the eye, regulated by upstream top control genes. Selection and random mutations are generally accepted as the main factors and driving forces of evolution. However, both are submitted to contrasting principles. Whereas the development and maintenance of adaptations during evolution is ruled by selection, their origin is ruled by chance, because it is based on random mutations at the molecular genetic level. The regressive evolution of light dependent structures, such as eyes, visually triggered behaviour or

pigmentation in cave animals, which have lost their biological function in darkness, would be one of the rare cases, in which the random mutations being characteristic at the molecular basis of evolution are not eliminated by selection, but can accumulate and be manifest phenotypically. By these processes the characteristic variability of regressive structures can be explained.

Oral presentation

Sensory receptor gene family evolution in cryptozoic insects: first insights from the red flour beetle *Tribolium castaneum*

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Universal consequences of long-term adaptation to life in caves include extended life span, increase in mechanoreceptors, reduction of visual organs and body pigmentation loss. Despite documentation across diverse animal groups, the correlated developmental, genomic and gene regulatory consequences have been studied thus far only in cavefish. We have begun to explore the molecular consequences of cryptozoic adaptation in insects by virtue of the recently sequenced genome of the red flour beetle Tribolium castaneum. Morphological and behavioral evidence indicate a state of advanced cryptozoic adaptation in this highly crepuscular grain-store pest. Under laboratory conditions, flight is only provoked by stress such as crowding or heat. Compared to other darkling beetles (Tenebrionidae), the compound eyes of Tribolium species are small and unevenly shaped. Intriguingly, the Tribolium genome contains high numbers of olfactory and gustatory receptor genes while the number of visual receptor genes (opsins) is reduced. Most insects express at least one opsin gene with sensitivity in the blue range. However, only opsins with photoreceptor sensitivities in the green and UV wavelength range are expressed in the *Tribolium* retina. Asking if the implied lack of color vision in Tribolium is ancient or derived, we investigated opsin conservation in darkling beetles with large, highly resolving compound eves: the mealworm Tenebrio molitor and the superworm Zoophobas atratus. We found that these species express the same opsin genes as Tribolium and in the same pattern. Hence, the lack of color vision in Tribolium is old and possibly ancestral in darkling beetles. Our results point at the possibility that energy savings rather than dispensability of vision were the driving force of eye reduction in *Tribolium*.

Poster presentation Distribution of benthic macroinvertebrates in two streams of the Alfredo Jahn Cave, Venezuela

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Aquatic communities in cave streams are primarily fueled by organic matter generated from the surface because photosynthetic primary production is nonexistent due to light absence. The two galleries of the Alfredo Jahn Cave in Venezuela have permanent streams, which remain unconnected through the year, except during flood events during the rainy season. The Quebrada Gallery receives surface water inputs from the Cambural River and is connected to the surface by large openings, while the Río Gallery is primarily fed by percolation water and is isolated surface inputs. In this study, benthic macroinvertebrate from communities were compared between the two gallery steams to examine the influence of organic matter sources on community structure. Water and benthos samples were collected in six stations along the two cave streams. Nutrients, suspended organic matter and inorganic solids concentrations in stream water were higher in the Río Gallery than in the Quebrada Gallery, probably explained to higher guano inputs from bats in the Río Gallery. Higher densities of macroinvertebrates were found in the Ouebrada Gallery stream, where Coleoptera, Diptera and Ephemeroptera were the most abundant groups while Gasteropoda and Oligochaeta were the dominant taxa in the Río Gallery. Most macroinvertebrates were shredders and collectors in the Quebrada Gallery, and scrappers and collectors in the Río Gallery. Multivariate analysis based on taxonomic composition and trophic groups indicated a higher similarity between communities from the same gallery than between galleries which could be related with organic matter sources and hydrological conditions.

Oral presentation Patterns of troglobiotic beetle (Coleoptera) biodiversity compared to sampling intensity and recent environmental correlates in the Western Balkans

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Distribution records of troglobiotic beetles in the karst of Western Balkans (Dinaric karst plus eastern parts of Southern Calcareous Alps) have been extracted from numerous sources, localities georeferenced and arranged in a relational database. Species numbers of three families (Carabidae, Cholevidae, Pselaphidae) were mapped on a grid with square cells 20x20 km and additionally, contour maps of species richness prepared, with the use of spatial interpolation methods. Sampling intensity was measured by the number of sampled subterranean localities with beetles per grid cell. For the study area, the following environmental correlates were available: altitude. mean annual precipitation, mean annual temperature, and vegetation. Besides internet sources, information was obtained from printed maps for the area that were digitized and vectorized for use in GIS. Two differing sources of vegetation cover were used: the information from the satellite recordings as well as the map of potential vegetation. The grid cells with higher numbers of species and the ones with higher numbers of localities mostly do not overlap. Cells with similar numbers of sampled localities had different numbers of species. Sixty percent of variance in number of species in cells can be explained by the variance in number of sampled localities. Species richness patterns of troglobiotic beetles are to a certain extent linked with sampling intensity, but they cannot be attributed to differences in sampling only. The highest statistically significant correlation of species numbers was with precipitation. Altitude showed statistically significant but lower positive correlation. The lowest value of the mean annual temperatures within the grid cell showed low, but statistically significant negative correlation. Numbers of species were significantly positively correlated with the proportion of potential forest vegetation within grid cells, while the correlation with the existing vegetation cover (as recorded with satellite) was not significant. Interpolated maps of species richness were compared visually with maps of precipitation and temperature. Distribution of the areas with mean annual precipitation over 1400 mm and mean annual temperatures under 10°C showed considerable similarity with distribution of the areas where more than three species were expected.

AUTHOR INDEX

Abraham6	
Abrams	
Adams 1	
Aden	8
Allegrucci 9, 3	39
Anderson 10, 9	96
Aotani 7, 16, 24, 43, 6	68
Austin 7, 16, 24, 43, 6	50
Baratti	32
Bedos	26
Bell	
Bennison1	
Bernhardt	55
Berry	
Bichuette 12, 13, 10)2
Bogdan	34
Borowsky 12, 14, 4	11
Brad 15, 67, 6	59
Bradford1	
Brehier	26
Brito17, 65, 66, 7	
Cai 10	
Campagna	
Carchini	
Carlini	
Casale	
Castillo 10	
Cho 18, 1	9
Clarke 20, 21, 2	
Cobolli	55
Cocking4	14
Cocking	35
Contreras	74
Contreras	13
Croom	93
Culver 25, 4	19
De Lange	12
de Pinna 13, 71, 7	72

Deharveng
Di Russo 55
Douady 22, 23
Dracup40
Drake
Dumnicka27 Eberhard28, 29, 33
Eberhard
Edward46 Escobar-Briones
Fabrio
Faría 108
Felice 102
Feng
Fenwick 31
Ferrarese86
Filipelli
Finston
Fišer 34
Fong 36, 49
Fortunato 9
Friedrich 107
Gal34
Gamboa 108
Gardner 40
Geoffroy37 Giachino
Giachino 28, 80
Gibert
Gillespie60
Goater40
Gottstein30
Gowarty41
Gravot22
Gregory42
Griswold60
Guil 102
Gunawardene42 Guzik7, 24, 43
Guzik
Hackley98

Halse	33	, 44	, 89
Hancock			
Hartland			.31
Harvey			. 46
Hedin			77
Hervant	22	, 23	, 85
Hoch			
Hoch Holsinger		. 49	, 92
Horvat			
Hose		. 58	, 59
Howarth		. 48	, 93
Hughes			.76
Humphreys . 16,	18,	24,	43,
46 50 76 97			
Husana			51
Hutchins			.36
lepure		. 67	, 69
lliffe		65,	104
Jalžić			.30
James			.93
Jeffery			. 52
Juan			74
Juan Karanovic		. 53	, 54
Kase			.51
Keber			
Kereži			.34
Ketmaier			. 55
Knott			.40
Koenemann			
Korbel			
Kovačec			
Krejca			
Lategan			. 59
Latella			. 55
Ledford			60
Leijs		. 61	, 84
Levei			. 69
Lo			
López		. 62	, 79
López-Pancorbo			.83

Luštrik	34
Maestre	12
Magniez	63
Marcia	64
Marković	75
Marmonier	38
Martens	89
Martínez 17, 65, 66,	
Martìnez-Ansemil	86
McRae	44
Meleg 67,	69
Merritt	68
Miclean	69
Minggyi	26
Moiler	
Moldovan 67,	69
Montagud 70,	90
Morales	23
Moreno 12,	70
Moškrič	34
Moulds	
Моуа	74
Muirhead	29
Muriel-Cunha	72
Novak Núñez17, 65, 66,	34
Núñez 17, 65, 66,	73
Ogorevc Oromí62, 74,	34
Ortuño 70,	
Ozimec	75
Page76, 1	04
Palandančić	34
Paquin 60,	77
Parzefall	
Patel	
Pérez62, 79,	
Petkovska	
Pipan	25
Piscart	
Plath	78

Polak	
Potočnik	34
Protas	81
Puglisi	42
Pustovrh	34
Rampini 39,	55
Reid	82
Renault	22
Ribera 74, 83,	90
Röling	
Roudnew	84
Saint	24
Salin	85
Sambugar	86
Sansone	88
Sbordoni 9,	39
Scanlon	44
Schön	89
Sečen	
Secutti 1	02
Sendra70,	90
Serov	58
Shafer	49
Sidorov	
Simon	38
Sket34, 1	09
Škrget	34
Slana	34
Stevens	
Stoch	
Stone 48, 93,	
Storey	
Straalen	
Strecker	95

Susac	96
Taiti 32,	97
Takhteev	
Talamante 1	00
Taukulis	42
Taylor	98
Teruel 70,	90
Thomas	
Tiedemann	55
Todisco	
Tomlinson	99
Trajano . 12, 13, 88, 101, 1	02
Trontelj80, 1	03
Turjak	34
Ulloa 1	08
Urios	90
Voituron	85
von Rintelen 1	
Walker	94
Walter	46
Wessel	48
Wilkens 1	05
Wood 1	07
Worsaae	66
Wowor 1	04
Wu26, 1	07
Xuefeng	26
Yamamuro	51
Youbang	26
Yum	19
Zagmajster 1	
Zuanon	
Zullini	86









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