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LIVRO DE RESUMOS

LIBRO DE RESÚMENES

ABSTRACT BOOK

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*Research Center in
Biodiversity and
Genetic Resources*

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Tuesday, September 30th, 17:30 h**From historical biogeography and shifting range borders to phylogeography and dispersal**

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Parallel species distributions often reflect similar ecological requirements and are frequently interpreted as evidence for shared evolutionary histories. However, over what time period may this hold true and how deep in time can we look back, really? Early on in my career I adopted the comparative approach to deal with this type of questions and I worked with pairs of amphibian species, such as in the genera *Bombina* and *Bufo*, *Lissotriton* and *Triturus*. A keystone for fixing the history of species in time and space has been ‘enclaves’ – pockets of a species range geographically isolated from the stock by the other species in the pair. In my talk I hope to show how my work, from historical biogeography and shifting range borders to phylogeography and dispersal, has been inspired by enclaves and other remarkable biogeographical patterns.

Life in extreme habitats. Lizards of small islands, islets and rocks: some examples from the western Mediterranean

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The Mediterranean region is an important biogeographical "crossroad", where islands represent crucial biodiversity reserves particularly rich in endemic species. Several thousands of islands, islets and rocks, of which less than the 3% is permanently inhabited by Man are found in this area. Paleo-geographical issues are not always the most important factors in explaining the present faunal composition of these islands, because man intervention has also played an important role. Strong and extended effects of human activity are therefore one of the current distinctive features of Mediterranean faunas. According to the present knowledge, this effect has been acting since the Neolithic times starting in the Near East and extending westwards. Since then many of the characteristic biogeographical elements of the region have undergone a process of exploitation which has continued across into historical times till now. The exploitation of habitats, of native species as well as the voluntarily/involuntarily introduction of aliens led to the extinction of several island autochthonous species. At present the terrestrial herpetofauna of many of the Mediterranean islands displays a partially homogeneous composition of elements. It also consists of species that are more or less common to the present fauna of all the Mediterranean islands, showing in part a generic continental origin influenced by the faunal composition of the nearest mainland. Endemic amphibians mostly occur on the biggest western Mediterranean islands while endemic reptiles survived also on secondary, sometimes quite inaccessible and "insignificant" small islands. Some geckoes and some lizard species (e.g. *Podarcis*) are able to inhabit, even in very low densities, tiny islet (area < 5000 m²) where no other terrestrial vertebrates would be seemingly able to live. Within reptiles the highest rate of endemism is attained by lizards, in particular in micro-insular contexts. These endemic small island populations that survived therefore became "uniqueness"! Within vertebrates the terrestrial herpetofauna can be considered one of the main indicators for island faunistic population dynamics. Thus the study of the eco-ethological characteristics of this fauna, developed over thousands of years on small islands and islets, become of fundamental importance not only for the identification of proper conservation strategies but also because of the opportunity given by these evolutionary laboratories to test and investigate peculiar ecological traits as well as evolutionary routes.

NA2RE Project: The new atlas of amphibians and reptiles in Europe

NEFTALÍ SILLERO

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A precise knowledge of the spatial distribution of taxa is essential for decision-making processes in land management and biodiversity conservation, both for present and under future global change scenarios. This is a key base for several scientific disciplines (e.g. macro-ecology, biogeography, evolutionary biology, spatial planning, or environmental impact assessment) that rely on species distribution maps. The Societas Europaea Herpetologica (SEH) published in 1997 the first atlas of European amphibians and reptiles with 50×50 km resolution maps and ca. 85,000 grid records. Since then, more detailed species distribution maps covering large parts of Europe became available, while taxonomic progress has led to a plethora of taxonomic changes including new species descriptions.

For these reasons, the SEH decided in 2006 through its Mapping Committee to implement the New Atlas of Amphibians and Reptiles in Europe (NA2RE: <http://na2re.ismai.pt>) as a chorological database system. Initially designed to be a system of distributed databases, NA2RE quickly evolved to a Spatial Data Infrastructure, a system of geographically distributed systems. Each individual system has a national focus and is implemented in an online network, accessible through standard interfaces, thus allowing for interoperable communication and sharing of spatial-temporal data amongst one another. A Web interface facilitates the access of the user to all participating data systems as if it were one single virtual integrated data-source. Upon user request, the Web interface searches all distributed data-sources for the requested data, integrating the answers in an always updated and interactive map. This infrastructure implements methods for fast actualisation of national observation records, as well as for the use of a common taxonomy and systematics. Using this approach, data duplication is avoided, national systems are maintained in their own countries, and national organisations are responsible for their own data curation and management. The database could be built with different representation levels and resolution levels of data, and filtered according to species conservation matters. This system is implemented using only open source software: PostgreSQL database with PostGIS extension, Geoserver, and OpenLayers.

The NA2RE system uses the data compiled by the Mapping Committee of the SEH until 2013. We used different data sources: published in books and websites, ongoing national atlases, personal data kindly provided to the SEH, the 1997 European Atlas, and the Global Biodiversity Information Facility (GBIF). Databases were homogenised, deleting all information except species names and coordinates, projected to the same coordinate system (WGS84) and transformed into a 50×50 km grid. The newly compiled database comprises more than 384,000 grid and locality records distributed across 40 countries. We calculated species richness maps as well as maps of Corrected Weighted Endemism and species distribution types (i.e. groups of species with similar distribution patterns) by hierarchical cluster analysis using Jaccard's index as association measure. We also identify taxonomic and geographic gaps of knowledge that need to be filled, and we highlight the need to add temporal and altitudinal data for all records, to allow tracking potential species distribution changes as well as detailed modelling of the impacts of land use and climate change on European amphibians and reptiles.

Taxonomic and Functional Responses of Reptiles to Fire in the Mediterranean Basin

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Fire is a common disturbance in many regions of the world, and a fundamental element to understand ecosystem functioning and structure. Over the short term, fire may act as an environmental filter that selects species better adapted to the restrictive postfire conditions. Early postfire succession increases open areas; in these environments we expect that many reptiles would be favoured. According to the habitat-accommodation model of succession, reptiles are expected to replace each others in parallel to post-fire changes in habitat structure. Most studies to validate this model have been conducted in Australia, identifying early, medium and late reptile species on regard of their occurrence with respect to the time since fire. To test this model in the Mediterranean basin is challenge since responses may vary depending on particular life-history traits of species. During the last seven years, I focused my research on analyzing responses of reptile assemblages in six localities at the Western Mediterranean, in order to tackle the following questions: Are all reptile species favoured by fire? Which functional traits are selected by fire? How ecological specialization affects responses to fire? Are threatened species particularly affected by fire? Does fire affect Mediterranean and Atlantic species in a similar way? Does the reptile community follow the habitat-accommodation model of post-fire succession? My final objective is to translate my results to stakeholders and conservationists to design adequate guidelines to protect species from fire, especially under a future scenario of increasing fire frequency and extension.

Ecopathology and zoonosis in herpetology

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Since the initial descriptions of diseases in herpetology during the 70s, this concept has acquired growing relevance. The study of ecopathology gained importance with the increased detection of invasive amphibian and reptile species. In consequence, the number of diseases has grown over the last years. In Spain and Portugal, self-limiting diseases in wild species have been reported, including melanomas in *Calotriton arnoldi* or haemoparasites in lacertid lizards (described in *Gallotia*, *Podarcis*, etc.). Furthermore, without an adequate sanitary control, the commerce and translocation, either legal or illegal, and even the scientific studies and official releases of specimens can be a mode of dispersal and introduction of pathogens. Pathogenic agents like *Batrachochytridium*, *Saprolegnia* or *Basidiobolus* in amphibians, as well as *Herpesvirus* or *Ranavirus* in turtles or *Iridovirus* and *Adenovirus* in lizards are emergent diseases that can seriously compromise the projects of recovery of threatened species. Moreover, it is well known that, in captivity, parasites can alter the equilibrium that they maintain with their hosts in the nature, and thus the released animals will receive parasites that are naturally present in the ecosystem. These factors must be taken into account in reintroduction projects. In Spain and Portugal some pathogens to be considered in any management action involving autochthonous herpetofauna have begun to be isolated. The most important are parasites like trematodes Spirorchidae in *Emys* from Galicia or *Serpinema microcephalum* in exotic turtles (*Trachemys*, *Ocadia*, *Graptemys*) naturalized in Barcelona; fungi like *Batrachochytridium* in amphibians from Madrid, Doñana or Balearic Islands; bacteria like *Mycoplasma testudinis* in turtles coming from France or Morocco; virus like *Ranavirus* in introduced newts (*Mesotriton alpestris*) in Catalonia, or *Herpesvirus* (TeHV-1 y TeHV-3) in captive terrestrial turtles (*Testudo graeca*) in Murcia. Some diseases are still being investigated like certain dermatomycoses in *Alytes* from central Catalonia, or erythrocytic virus in *Podarcis bocagei* and *P. carbonelli* from northern Portugal. Most of these diseases affect only reptiles and do not represent a sanitary risk to humans (zoonosis). Zoonotic risks are relatively small and focused almost exclusively in occurrences in children, elderly and immunodepressed people (some cases reported in Barcelona, Valencia and Bilbao between 2010 and 2012). Herpetologists must be considered as population at risk against diseases transmitted by manipulation (*Salmonella*, *Mycobacterium* or *Candida*, among others). In consequence, we must keep in mind that scientists can also be the cause for pathogen dispersion. Hygienic measurements must be taken not only to avoid zoonosis but also to impede the dispersion of diseases towards the ecosystems.

When amphibians become a problem – a balance of the first four years of the control and eradication plan of the African clawed frog on the streams of Oeiras Municipality

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As a result of introductions out of its native range, the African clawed frog, *Xenopus laevis*, is one of most widely distributed amphibians in the world. It was introduced in four continents, with considerable success in temperate and Mediterranean-climate regions, where its direct and indirect impacts changed the ecology and dynamics of invaded ecosystems. This species may also be an asymptomatic vector for pathogens (namely the fungus *Batrachochytrium dendrobatidis*), that may thus be transmitted to native amphibians.

Reproducing populations of this species were found for the first time in Portugal in the Laje stream (in 2006) and in Barcarena stream (in 2008). Both streams run through Oeiras Municipality. A control and eradication plan is taking place since 2010, under a protocol signed by the Institute for the Conservation of Nature and Forests, I.P., the Oeiras Municipality, the Centre for Environmental Biology/ FCUL, and the Instituto Gulbenkian de Ciência.

In this talk, I will present the main results of the plan thus far. At the end of the fourth year of the plan, the state of the *X. laevis* populations in both streams is contrasting. At Laje, the species was very abundant at the start of the plan, and in 2013 we only captured 7 adults, with no signs of reproduction in all the stream. On the contrary, at Barcarena the species reproduced in at least two stream stretches, its main nucleus occupies a relatively large stretch of the stream, and 845 individuals (the large majority of them 1-year old juveniles) were captured in 2013. The capture of 1-year olds means that probably a single successful year (2012) could rapidly repopulate a significant portion of the stream. Taking into account its invasive potential, the risk of colonization of other areas, and the success of the plan at Laje thus far, the plan will continue in the next years, and new methodologies will be tested.

Friday, October 3rd, 17:45 h**Transferring herpetological knowledge to conservation biology**

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The knowledge of biology, corologia and genetic diversity, in the field of herpetology, increased exponentially in the last two decades in Portugal. Nevertheless, the transfer of this knowledge to the Scientific Instruments of Land Management and especially for the management programs of nature conservation and environmental assessment is inexpressive. The lack of long-term monitoring and segregation of human resources between management and research centers, programs are still obstacles to be overcome.



COMUNICAÇÕES ORAIS
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Patrones de variación genética en *Discoglossus* ibéricos: implicaciones taxonómicas

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El género *Discoglossus* incluye cinco o seis especies distribuidas por todo el Mediterráneo Occidental. Aunque sus relaciones filogenéticas están relativamente bien resueltas, la posición taxonómica de los taxones ibéricos *D. galganoi* y *D. jeanneae* sigue siendo objeto de debate, y algunos autores las consideran a nivel subespecífico. Estudios previos han identificado clados muy divergentes basados en secuencias de ADN mitocondrial (ADNmt), pero poca estructura en marcadores nucleares de evolución lenta. En este estudio, caracterizamos 12 nuevos marcadores polimórficos (2-21 alelos por locus) de tipo microsatélite y los empleamos para describir los patrones de variación genética en 310 individuos muestreados por toda la Península Ibérica. Elaboramos modelos de favorabilidad ambiental para diferentes grupos de poblaciones basados en los datos de microsatélites y comparamos los resultados con modelos similares basados en la asignación de individuos a clados de ADNmt para evaluar la concordancia entre diferentes marcadores, identificar áreas de contacto y evaluar hipótesis de aislamiento reproductivo entre grupos. De acuerdo con los resultados, que muestran amplias zonas de contacto por todo el área de distribución, sin alelos fijados alternativamente en los diferentes grupos, concluimos de acuerdo con estudios previos que los dos taxones ibéricos deben ser considerados como subespecies (*D. galganoi galganoi* y *D. galganoi jeanneae*).

Patterns of genetic variation in Iberian *Discoglossus*: taxonomic implications

The genus *Discoglossus* includes five or six species distributed over the western Mediterranean basin. While their phylogenetic relationships are relatively well established, the taxonomic position of the Iberian taxa *D. galganoi* and *D. jeanneae* is still disputed, with some authors considering them at the subspecific level. Previous studies have shown deeply divergent mitochondrial DNA (mtDNA) clades but little structure in slowly evolving nuclear markers. In this study, we use 12 newly developed polymorphic microsatellite markers (2-21 alleles per locus) to address patterns of genetic variation in 310 individuals sampled throughout the Iberian Peninsula. We built models of environmental favorability for microsatellite-based population clusters and compared them with models based on assignment of individuals to mtDNA clades in order to evaluate concordance across datasets, identify areas of secondary contact and evaluate hypotheses regarding reproductive isolation between groups. Based on the finding of extensive inferred admixture throughout the range, with no fixed allelic differences across groups, we agree with previous studies that the two Iberian taxa should be better regarded as subspecies (*D. galganoi galganoi* and *D. galganoi jeanneae*).

Taxonomy, microsatellites, mtDNA, species distribution modelling, phylogeography, *Discoglossus*.

Desvendando uma diversidade extraordinária – ADN Barcoding dos répteis de Socotra

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O barcoding de ADN é baseado na ideia de usar uma sequência genética curta de um marcador padrão (gene citocromo c oxidase 1, COI) para a identificação de espécies. Tem a vantagem de facilitar a identificação das espécies e acelerar a descoberta de espécies candidatas/ crípticas de uma forma bastante fiável e economicamente eficiente, com várias implicações na conservação e gestão. No entanto, apresenta alguns problemas técnicos na amplificação, especialmente de répteis; e problemas metodológicos, como a dificuldade na delimitação dos limiares do intervalo do barcode e do número mínimo de sequências de cada espécie, a dependência da escala e limitações nas inferências filogenéticas. Campanhas recentes de barcoding de répteis foram bem-sucedidas e por isso apressámo-nos para aplicá-lo em Socotra, devido à importância desta região, classificada como Património Mundial Natural, e a importância da conservação da sua herpetofauna única, actualmente com 29 espécies endémicas. Depois de várias campanhas de amostragem intensas no arquipélago, com estações a cada 10 x 10 km, mais de 1300 tecidos foram recolhidos. Sequenciámos com sucesso, utilizando-se três conjuntos de primers, 380 indivíduos de todas as espécies actualmente reconhecidas, com um sucesso óptimo de reconhecimento de 99%, e com um sucesso de barcoding de todas as espécies de cerca de 72% na identificação dos espécimes. Detectámos um caso de parafilia e também elevados níveis de variabilidade genética intra-específica, por vezes maiores que os valores de variabilidade genética interespecífica. O resultado da análise de delimitação de espécies GMYC a diferentes níveis, bem como a análise de intervalos de barcoding usando diferentes limiares, revelou níveis relativamente altos de diversidade críptica, sugerindo que devem ser tidos em conta para a gestão da conservação entre mais 4 a 37 entidades do que aquelas que são actualmente reconhecidas. Os resultados previamente publicados e preliminares, utilizando dois marcadores mitocondriais e nucleares, e os altos níveis de variabilidade genética em COI detectados neste estudo, especialmente dentro de *Hemidactylus* e *Pristurus*, incluindo um caso de parafilia, sugerem que a taxonomia de alguns répteis de Socotra deveria ser revista através de uma aproximação integrativa combinando dados moleculares e morfológicos.

Unveiling extraordinary diversity – DNA Barcoding of Socotra reptiles

DNA barcoding is based on the idea of using a short genetic sequence from a standard marker (cytochrome c oxidase 1 gene, COI) for species identification. It has the advantage of facilitating species identification and accelerating discovery of candidate/ cryptic species in a largely reliably and cost-efficient way, with many implications in conservation and management. However, it presents some technical problems in amplification, especially with reptiles; and methodological problems, such as difficulty in threshold delimitation for barcode gaps and minimum number of sequences per species, scale-dependency, and limitations in phylogenetic inference. Recent barcoding campaigns of reptiles have been successful and so we urged to apply it to Socotra due to the significance of this region, classified as Natural World Heritage, and the conservation interest of their unique reptile fauna, presently with 29 endemic species. After several campaigns of extensive sampling in the archipelago with stations each 10 x 10 km, more than 1300 tissues were collected. We have successfully sequenced, using three sets of primers, 380 individuals of all presently recognised species with a best match success of 99%, and with all species barcode success of about 72% of specimen identification. We detected one case of parafily and also high levels of intraspecific genetic variability, sometimes higher than values of interspecific genetic variability. The result of GMYC species delimitation analysis at different levels, as well as a barcode gap analysis using different thresholds, unveiled relatively high levels of cryptic diversity, suggesting that between 4 and 37 more entities than those presently recognised should be taken into account for conservation planning. Published and preliminary results using both mitochondrial and nuclear markers and the high levels of genetic variability in COI detected in this study, especially within *Hemidactylus* and *Pristurus*, including one case of parafily, suggest that the taxonomy of some Socotra reptiles should be revised using an integrative approach combining molecular and morphological data.

Barcoding, Socotra, reptiles, COI, species discovery.

Filogenia de três espécies de *Chalcides* endêmicos de Marrocos: *C. manueli*, *C. montanus* and *C. polylepis*

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A Bacia do Mediterrâneo, da qual Marrocos faz parte, é um dos Hotspots Globais de Biodiversidade. Características específicas deste país, como o clima e topografia, permitem grandes níveis de riqueza e endemismo, particularmente de herpetofauna. O género *Chalcides* compreende 28 espécies, muitas das quais estão presentes em Marrocos e países adjacentes, sendo caracterizados pelo tronco alongado e membros reduzidos. A taxonomia deste género foi revista diversas vezes, mas ainda continua a ser complicada a identificação de determinadas espécies no campo, parecendo provável que a taxonomia atual não reflita completamente a sua história evolutiva. Estudos anteriores relativos à filogenia do género, baseados no DNA mitocondrial, identificaram quatro grupos principais. Neste trabalho, o foco de estudo são as relações filogenéticas entre três espécies do "Clado Ocidental": *Chalcides manueli*, *C. polylepis* e *C. montanus*. Genes mitocondriais e nucleares das populações mais conhecidas foram analisados e os resultados indicam que a taxonomia atual não reflete a diversidade genética.

Molecular phylogeny of three Moroccan *Chalcides*: the *C. manueli*, *C. montanus* and *C. polylepis* clade

The Mediterranean Basin, in which Morocco is included, is one of the Global Biodiversity Hotspots. Specific characteristics of Morocco, such as the climate and topography, allow high levels of richness and endemism, particularly in herpetofauna. The skink genus *Chalcides* comprises 28 species, many of them present in Morocco and surrounding areas, and these are characterized by having elongated bodies and reduced limbs. The taxonomy of *Chalcides* has been revised numerous times, but many species remain difficult to identify in the field, and it seems likely that the current taxonomy still does not fully reflect their evolutionary history. Previous studies regarding the phylogeny for the entire genus based on mtDNA identified four major groups. In this work we investigate the molecular phylogenetic relationships of the three taxa within the "Western clade": *C. manueli*, *C. polylepis* and *C. montanus*. Mitochondrial and nuclear genes of most of the known populations of these three species were analysed. Our results indicate that current taxonomy does not reflect genetic diversity.

Chalcides, phylogenetic analyses, phylogeography, haplotype networks, mtDNA, MC1R.

Perspectiva genética da dinâmica espacial da hibridação numa zona de contacto entre duas espécies de lagartixas

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Compreender o isolamento reprodutivo e as barreiras ao fluxo génico é essencial para entender a especiação. Desta forma, as zonas híbridas (ZH), áreas onde espécies geneticamente diferentes entram em contacto e se cruzam, permitem o estudo da diferenciação entre espécies porque proporcionam uma oportunidade de identificar potenciais barreiras ao fluxo génico, estudar os padrões genómicos da selecção e introgressão, bem como para identificar os caracteres fenotípicos sob selecção. *Podarcis bocagei* e *P. carbonelli* constituem um bom modelo de estudo para a especiação uma vez que já foram descritos cruzamentos entre ambas, apesar de manterem a sua identidade genética e morfológica na sua restrita área de contacto. Neste projecto propusemo-nos a estudar a ZH entre as duas espécies utilizando uma estratégia multidisciplinar. Especificamente, combinamos análises de genética populacional e de morfologia com um mapeamento geográfico a fina escala de cada indivíduo, de forma a compreender a dinâmica espacial desta zona. Para a análise molecular utilizou-se DNA mitocondrial e um conjunto de microssatélites para estimar o nível de miscigenação de cada indivíduo. Estes marcadores moleculares permitiram a identificação de 144 indivíduos de *P. carbonelli* e 33 de *P. bocagei*, assim como 18 híbridos, resultando numa taxa de hibridação de 9.2%. Foi possível identificar diversas classes de hibridação, entre as quais F1, F2 e retrocruzamentos com as classes parentais. Apesar da maior abundância de *P. carbonelli* na amostragem, os híbridos apresentam, maioritariamente, linhagem materna de *P. bocagei*, indicando uma possível direcção de cruzamentos entre fêmeas de *P. bocagei* e machos de *P. carbonelli*. A combinação da identificação genética com a localização dos indivíduos permitirá a avaliação da dinâmica espacial desta ZH e a realização de uma análise de clines. Através da análise espacial, detectamos uma zona de contacto estreito, onde estão localizados a maioria dos híbridos. No entanto, também encontramos alguns híbridos recentes no sul da zona de contacto, indicando dispersão fora da área de simpatria. Além disso, foi utilizada uma combinação de caracteres foliódicos para analisar os possíveis efeitos da hibridação na morfologia das espécies, explorando as características morfológicas dos híbridos. Por outro lado, analisamos os níveis de assimetria de diversos caracteres bilaterais para testar se a hibridação aumenta a instabilidade no desenvolvimento com potenciais efeitos do fitness dos híbridos. Com este estudo poderemos ter uma melhor compreensão da forma como as barreiras reprodutivas e a selecção aumentam/mantêm a divergência entre estas duas espécies. Por fim, os resultados obtidos poderão esclarecer alguns mecanismos evolutivos envolvidos nos processos de especiação com fluxo génico no sistema modelo de *Podarcis* Ibéricas e do Norte de África e na diversificação biológica em geral.

A landscape genetics perspective on the spatial dynamics of hybridization between two species of wall lizards

Understanding reproductive isolation and barriers to gene flow is essential to understand speciation. As such, hybrid zones (HZ), areas where genetically differentiated species come into contact and interbreed to some extent, allow the study of differentiation between species because they provide the opportunity to identify potential barriers to gene flow, to study the genomic patterns of selection and introgression, and to identify the phenotypic characters under selection. *Podarcis bocagei* and *P. carbonelli* provide a good model system to study speciation because they are known to interbreed to some extent, but also to maintain their genetic and morphological identities in their small contact area. In this work we studied the HZ between these two species using a multidisciplinary approach. Specifically, we combined population genetic and morphological analyses with a fine-scale geographic mapping of individuals to understand the spatial dynamics of this HZ. For molecular analyses we use mitochondrial DNA and a combination of microsatellites to evaluate the level of admixture of each individual. These molecular markers allowed the identification of 144 individuals of *P. carbonelli* and 33 of *P. bocagei* as well as 18 hybrids, resulting in a hybridization rate of 9.2%. Hybrids consist of multiple classes (F1, F2, backcrosses with each parental form and higher generation hybrids). Moreover, despite the presence of more *P. carbonelli* individuals in our sample, hybrids have the majority of the maternal lineage belonging to *P. bocagei*, indicating a directionality of crosses where females of *P. bocagei* breed with males of *P. carbonelli*. Genetic identification is crossed with individual location in order to look for spatial dynamics in the HZ and to perform cline analyses. Through the spatial analysis we were able to detect a narrow contact zone where the majority of the hybrids are located. Nevertheless, we also

detected some recent hybrids at the south of the contact zone, indicating dispersion outside de sympatric area. Further, we used a combination of pholidotic traits to examine the effects of hybridization on the morphology of the species by exploring the morphological properties of hybrids. Also, we examined the level of asymmetry of several bilateral traits to test whether hybridization increases developmental instability with potential effects on the fitness of hybrids. Through this study we will be able to obtain a better understanding of how reproductive barriers and selection enhance and maintain the divergence between these two species. Further, the results obtained will provide insights to the evolutionary mechanisms involved in the process of speciation with gene flow in the model system of Iberian and North African *Podarcis* and in biological diversification in general.

Hybridization, landscape genetics, *Podarcis* sp.

Evolução e filogeografia do complexo de espécies *Acanthodactylus scutellatus* no Norte de África

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Eventos geológicos e paleoclimáticos são duas forças essenciais que atuam sobre os processos evolutivos na natureza. Estes são fenómenos pouco estudados no Norte de África, apesar da sua grande diversidade de habitats, paisagens heterogéneas, e histórias climáticas e geológicas complexas. Os padrões atuais de biodiversidade do Sahara e do adjacente Sahel provavelmente resultaram de oscilações fortes no clima e paisagem. O complexo de espécies *Acanthodactylus scutellatus* inclui importantes elementos da herpetofauna de ecossistemas áridos no Norte de África, e bem adaptados a condições xéricas. Apesar da sua notável diversidade, a taxonomia dentro do complexo é controversa, e com a exceção de dados morfológicos, pouco é sabido acerca destes organismos. Observações de indivíduos com morfologia intermédia em áreas de simpatria sugerem hibridação entre estas taxa. Este estudo pretende: 1) inferir relações filogenéticas dentro deste grupo e identificar as linhagens principais; 2) compreender os padrões filogeográficos das linhagens do Nordeste de África; 3) inferir o fluxo génico contemporâneo numa zona de contacto na Mauritânia. As análises filogenéticas e filogeográficas basearam-se em cerca de 450 indivíduos sequenciados para ambos os genes mitocondriais 12S e Cyt-b (756 total bp) e para o gene nuclear C-mos (513 total bp). Cerca de 210 indivíduos da zona de contacto foram genotipados para 17 microssatélites. Os nossos resultados preliminares mostram que as linhagens principais recuperadas não coincidem com a sistemática atual, sugerindo que taxonomia e sistemática necessitam de revisão. A estrutura genética encontrada é geograficamente coerente e possivelmente explicada pelas oscilações passadas no clima e extensão do deserto. As análises de genotipagem confirmaram as linhagens históricas encontradas e mostram ausência de fluxo génico entre elas, não sendo detetados híbridos apesar da grande amostragem e vasta cobertura geográfica. A falta de fluxo de genes observado levanta questões acerca do potencial papel de barreiras reprodutivas e seleção do habitat na prevenção de hibridação. As ferramentas moleculares aplicadas permitiram maior compreensão sobre os limites definindo as espécies, relações evolutivas, história e diversidade dentro deste grupo, para além de contribuir para o conhecimento sobre fluxo de genes no Sahara-Sahel.

Evolution and phylogeography of the *Acanthodactylus scutellatus* species complex in North Africa

Geological and paleoclimatic events are two main forces driving evolutionary processes in nature. These are poorly studied phenomena in North Africa, despite its great diversity of habitats, heterogeneous landscapes, and complex climatic and geological histories. Modern biodiversity patterns of the Sahara and the adjacent arid Sahel likely resulted from strong oscillations in climate and land-cover. *Acanthodactylus scutellatus* species complex comprises important elements of the herpetofauna of arid ecosystems in North Africa, and well adapted to xeric conditions. Despite their remarkable diversity, the taxonomy within the complex is controversial, and with the exception of morphological data, little is known about these organisms. Observations of morphologically intermediate individuals in sympatry areas suggested hybridization between taxa. The present study aims to: 1) infer phylogenetic relationships within this group and identify major lineages; 2) understand the phylogeographic patterns of north-western African lineages; 3) infer contemporary gene flow in a contact zone in Mauritania. Phylogenetic and phylogeographic analyses were based on about 450 specimens that were sequenced for both 12S and Cyt-b mitochondrial genes (756 total bp), and for the C-mos nuclear gene (513 total bp). About 210 individuals of the contact zone were genotyped for 17 microsatellites. Our preliminary results show that recovered major lineages do not match with current systematics, suggesting that taxonomy and systematics need revision. The genetic structure found is geographically coherent and it is likely explained by past oscillations in climate and desert extent. Genotyping analyses confirmed the historical lineages found in the studied contact zone and show absence of gene flow between them, with no hybrids detected despite the large sampling and broad geographic coverage. Lack of observed gene flow raises questions about the putative role of reproductive barriers and habitat selection in preventing admixture. Finally, the molecular tools applied allowed for insights on the species boundaries, relationships, history and diversity of this group, in addition to contributing to the knowledge about gene flow in the Sahara-Sahel.

Acanthodactylus scutellatus, North Africa, phylogeny, phylogeography, gene flow.

Padrões filogeográficos da osga endêmica de Marrocos *Saurodactylus brosetti*

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Há vários casos de estudo onde a informação de sequências de DNA foi usada para inferir variação genética entre espécies na Europa. No entanto são poucos os estudos filogeográficos no Norte de África. Muitas espécies de répteis são, na realidade, complexos de espécies, em particular osgas. *Saurodactylus* é um género que contém três espécies actualmente reconhecidas, todas endémicas da região de Maghreb no Norte de África. Os estudos feitos neste género foram inicialmente feitos de forma geral, embora se tenha identificado uma diversidade considerável com DNA mitocondrial. Em particular, *Saurodactylus brosetti* mostrou altos níveis de diversidade, apesar da limitada amostragem. Além disso, populações aparentemente isoladas no extremo sul e este de Marrocos não foram ainda analisadas. Por outro lado, a distribuição actual desta espécie não é clara e, recentemente, populações anteriormente desconhecidas foram detectadas através de novas amostragens. Neste casos, a modelação por Sistemas de Informação Geográfica tem sido bastante usada, permitindo prever a potencial distribuição da espécie, o que mais tarde irá ajudar a direccionar o trabalho de campo e, consequentemente, a interpretar padrões de diversidade genética. Usando *Saurodactylus brosetti* como modelo e comparando DNA mitocondrial e nuclear, aferimos i) se é um complexo de espécies ii) se há barreiras geográficas que separaram as linhagens genéticas e iii) qual a potencial distribuição da espécie e quais os fatores ambientais que mais influenciam a sua atual distribuição.

Phylogeographic patterns of the Moroccan lizard-fingered gecko *Saurodactylus brosetti*

There are many case studies where DNA sequence information was used to infer genetic variation within species across Europe. However, phylogeography in North Africa is less well studied. Many reptile species are apparently species complexes, in particular geckos. Regarding *Saurodactylus*, a genus containing three currently recognized species all endemic to the Maghreb region of North Africa, studies were primarily at a generic level, although considerable diversity was identified with mtDNA. *Saurodactylus brosetti* in particular showed high levels of diversity, although sampling was limited. Furthermore, apparently isolated populations in the far south and east of Morocco were not yet been examined. On the other hand, the actual distribution of this species is unclear with recent fieldwork detecting previously unknown populations. In such cases, GIS modeling has being widely used allowing prediction of the potential distribution of the species, which can then help with more directed fieldwork and in interpreting patterns of genetic diversity. Employing *Saurodactylus brosetti* as a model and comparing mtDNA with nuclear DNA, we assess i) if it is a species complex ii) if there are any geographic barrier with separation of genetic lineages and iii) what is the potential distribution of the species, and what environmental variables matter most regarding its current distribution.

Saurodactylus brosetti, phylogeography, ecological modeling.

Hotspots evolutivos de anfíbios da Península Ibérica

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A diversidade genética e os processos evolutivos estão na base da geração e persistência da biodiversidade. Como tal, há um crescente reconhecimento de que é necessário identificar e ter em conta os padrões espaciais da variação genética no desenho de redes de conservação. A Península Ibérica faz parte do hotspot de biodiversidade Mediterrânico, caracterizado pela ocorrência de diversas espécies endémicas. Esta área é particularmente importante para a conservação de anfíbios, uma vez que contém várias espécies com uma elevada variação genética intraespecífica, a qual se apresenta frequentemente espacialmente estruturada. Neste estudo, pretendeu-se identificar hotspots evolutivos de anfíbios da Península Ibérica, i.e. áreas de ocorrência de múltiplas linhagens intraespecíficas e áreas de ocorrência de múltiplas zonas de contacto. Foram compilados dados relativos a sequências de DNA mitocondrial e a respetiva localização geográfica das amostras de tecido, a partir da literatura científica, para 14 espécies de anfíbios da Península Ibérica. Para complementar lacunas de amostragem geográfica, sequenciaram-se novas amostras de outros locais. As relações filogenéticas dos principais linhagens dentro de cada espécie foram estimadas através de inferência Bayesiana. Cada árvore filogenética foi usada para calcular uma matriz de distâncias genéticas, a partir das distâncias cofenéticas entre diferentes haplótipos. Foi também calculada uma matriz de distâncias geográficas entre os locais de amostragem. Para agrupar os diferentes haplótipos em linhagens, recorreu-se a uma implementação Bayesiana do modelo general mixed Yule-coalescent (bGMYC). Em seguida, as matrizes de distâncias genéticas e geográficas foram usadas para identificar as áreas de ocorrência de cada linhagem em toda a Península Ibérica, bem como potenciais zonas de contacto entre linhagens, recorrendo a uma versão modificada da interpolação kriging. Por fim, identificaram-se hotspots evolutivos para as 14 espécies somando o número de linhagens distintas (riqueza de linhagens) e o número de zonas de contacto potenciais (riqueza de zonas de contacto). Os resultados têm importantes implicações para o planeamento da conservação, uma vez que a preservação de hotspots evolutivos contribuirá para a persistência de anfíbios da Península Ibérica de forma mais eficiente.

Amphibian evolutionary hotspots in the Iberian Peninsula

Genetic diversity and evolutionary processes underpin biodiversity generation and persistence. Thus, there is a growing recognition that spatial patterns of genetic variation should be identified and accounted for in the design of conservation networks. The Iberian Peninsula is part of the Mediterranean biodiversity hotspot, where several endemic species occur. This area is particularly important for the conservation of amphibian species, as it comprises several species exhibiting high intra-specific genetic variation, which is often spatially structured. In this work, we aimed at identifying evolutionary hotspots of Iberian amphibians, i.e. areas of occurrence of multiple intra-specific lineages and areas of occurrence of multiple contact zones. We compiled mitochondrial DNA sequence data and the respective geographic location of tissue sampling from the literature for 14 Iberian amphibian species. We also complemented geographical sampling gaps by sequencing new samples from other locations. We estimated phylogenetic relationships of major lineages within each species using Bayesian inference. Each tree was then used to calculate a genetic distance matrix using the cophenetic distances between distinct haplotypes. We also calculated a matrix of geographic distances between sample locations. We grouped distinct haplotypes into main lineages by using a Bayesian implementation of the general mixed Yule-coalescent model (bGMYC). We then used the genetic and geographic distance matrices to identify areas of occurrence of each lineage across the Iberian Peninsula and of potential contact zones between distinct lineages, by using a modified version of the kriging interpolation. Finally, we identified evolutionary hotspots by summing the number of distinct lineages (lineage richness) and the number of potential contact zones (contact zones richness) across the 14 species. Our findings have important implications for conservation planning, as the preservation of evolutionary hotspots will contribute cost-efficiently to the persistence of Iberian amphibians.

Phylogeography, lineage distribution, contact zones, kriging, conservation.

Diferentes orígenes de la población de tortuga mora de Doñana como consecuencia de su manejo histórico

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La tortuga mora (*Testudo graeca*) en el Mediterráneo Occidental se distribuye principalmente por el Norte de África, pero presenta algunas poblaciones pequeñas y aisladas en la Península Ibérica (Doñana y sureste Ibérico) y en las islas de Mallorca, Cerdeña y Sicilia. La ausencia de registro fósil en el oeste de Europa y la escasa diferenciación mitocondrial entre estas poblaciones y las del Norte de África hizo pensar que se originaran por introducciones en tiempos históricos. Sin embargo, algunos estudios genéticos recientes han identificado un origen más antiguo para la población del sureste ibérico, datado unos 20.000 – 30.000 años atrás. En este trabajo utilizamos información genética para estudiar el origen de la otra población ibérica, la albergada en el Parque Nacional de Doñana. Las citas más antiguas de la especie en Doñana se remontan al siglo XVIII, pero también se tiene constancia de refuerzos durante el siglo XX con animales procedentes de Marruecos. En consecuencia, el patrón genético de esta población podría reflejar diferentes orígenes. Muestreamos 70 tortugas de Doñana para obtener secuencias mitocondriales del gen citocromo b y utilizamos además trece marcadores microsatélite. Incluimos en los análisis datos moleculares de trabajos previos que abarcaron las poblaciones del sureste ibérico y del norte de África (Norte de Marruecos y Argelia). Para analizar las diferencias entre estas poblaciones utilizamos principalmente descriptores de genética de poblaciones, redes de haplotipos y análisis Bayesianos para la inferencia de grupos genéticos. Las secuencias mitocondriales evidenciaron la presencia de dos subespecies en Doñana. Encontramos haplotipos de *T. g. marokkensis* y de *T. g. graeca*, siendo estos últimos exclusivos y más frecuentes (compartidos por el 88,5% de las muestras). Los microsatélites reflejaron además la hibridación de las dos subespecies, mostrando señales de la introducción reciente de *T. g. marokkensis* en Doñana por encontrar un cuarto del genotipo de unos pocos individuos atribuido a este linaje. Aunque nuestros resultados indican que el manejo de la especie durante el último siglo ha alterado parcialmente el patrón genético inicial, futuros análisis utilizando aproximaciones Bayesianas para la inferencia de procesos demográficos podrían dilucidar posibles relaciones pasadas entre las dos poblaciones Ibéricas y sus originarias en el Norte de África.

Mixed ancestries of spur-thighed tortoises in Doñana as consequence of their historic management

Mixed ancestries of spur-thighed tortoises in Doñana as consequence of their historic management The majority of the western Mediterranean distribution of the spur-thighed tortoise (*Testudo graeca*) lies in North Africa, with a few small and isolated western European populations confined to the Iberian Peninsula (Doñana and south-eastern Spain) and on a few islands (Mallorca, Sardinia and Sicily). In general, western European populations are thought to be the result of historical introductions, which is supported by the absence of any fossil record in Western Europe and the low mitochondrial differentiation found between the European and North African populations. However, recent studies identified an ancient origin for the south-eastern Spanish population, which was dated 20,000-30,000 years ago. Here we use molecular data to shed light to the origin of the other Iberian population, the population located in Doñana National Park. The earliest references of tortoises inhabiting Doñana trace back from the 18th century, but several introductions of Moroccan animals during 20th century are also well documented. In consequence this population could have mixed ancestries as consequence of its historic management. We sampled 70 tortoises from Doñana to obtain mitochondrial sequences from the cytochrome b gene and genotypes derived from thirteen microsatellite loci. The generated dataset was analysed together with molecular data from previous works which involved south-eastern Spanish and North African tortoises. We mainly used population genetic descriptors, haplotype networks and Bayesian clusters analyses to assess the differentiation among the tortoises from Doñana, south-eastern Spain and North Africa (North and East of Morocco and North of Algeria). Mitochondrial data revealed the presence of two subspecies in Doñana. We found haplotypes attributed to *T. g. marokkensis* and *T. g. graeca*, being these last ones exclusive and more frequent (shared by 88.5% of samples). Microsatellite data reflected the hybridization among the two subspecies, showing signals of the recent introduction of *T. g. marokkensis* in Doñana by identifying a quarter of the genotype of a few animals attributed to this lineage. Although our results indicate that the management of the species during the last century have partially altered the initial genetic pattern, further analyses using Bayesian demographic approaches could help to date the origin of the previous *T. g. graeca* population and to decipher possible past relationships among the two Iberian populations and their common ancestors in North Africa.

Spur-thighed tortoise, *Testudo graeca*, Doñana, introductions.

Divergência Plioceno-Pleistocénica no noroeste do Magreb e atuais padrões de estrutura e diversidade genética no cágado-mediterrânico (*Mauremys leprosa*)

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A bacia do Mediterrâneo alberga um elevado número de endemismos e riqueza específica como resultado de uma complexa história paleogeográfica. Eventos como a Crise Salina do Mediterrâneo, formação e/ou transformação de características orográficas e as alterações climáticas do Quaternário tiveram enorme impacto na distribuição e diversificação das espécies na área. Atualmente, os efeitos antropogénicos (p. ex.: translocações, comércio de animais de companhia e para alimento) podem provocar impactos severos na história das espécies. O cágado mediterrânico, *Mauremys leprosa*, ocorre em toda a Península Ibérica e na maioria da região magrebina. Apenas duas subespécies de *M. leprosa* são reconhecidas atualmente: *M. l. saharica* (localizada a sul das Montanhas do Atlas até à Tunísia) e *M. l. leprosa* (localizada a norte das Montanhas do Atlas e na Península Ibérica). Neste trabalho pretendemos explorar o efeito que as alterações climáticas do passado e as barreiras geográficas produziram nos padrões atuais de diversidade genética e estrutura de *M. leprosa*. Sequenciámos mtDNA (citocromo *b* e a região de controlo) e genotipámos nuDNA (11 loci polimórficos de microssatélites) em cerca de 500 amostras. Os nossos objetivos são: 1) identificar a origem geográfica de diversificação; 2) determinar a estrutura genética dentro de cada subespécie; avaliar o efeito das grandes barreiras geográficas no fluxo génico (Estreito de Gibraltar e Montanhas do Atlas); 4) identificar possíveis zonas de contacto entre as subespécies e 5) estabelecer uma base de dados genéticos para alocar indivíduos de origem desconhecida. Identificámos um elevado grau de subestruturarão em África (três sublinhagens para *M. l. leprosa* e quatro para *M. l. saharica*) originadas durante o Plioceno-Pleistoceno. Ao longo do Rif e Médio Atlas detetou-se uma zona de contacto secundário entre as duas subespécies. Uma expansão rápida e recente ocorreu por toda a Península Ibérica e as potenciais translocações de animais poderão ter resultado no elevado grau de mistura genética, impedindo uma alocação precisa dos indivíduos desconhecidos. Globalmente, este estudo atualiza o conhecimento sobre qual o papel que as características geográficas e o clima produziram nos padrões de diversidade genética em *Mauremys leprosa*, complementando o conhecimento atual relativamente à importância do Norte de África como refugio no último máximo glacial.

Pliocene-Pleistocene divergence in North-Western Maghreb and current patterns of genetic diversity and structure of the Mediterranean pond turtle (*Mauremys leprosa*)

The Mediterranean basin harbours a high degree of endemism and species richness as a result of a complex palaeogeographic history. Events such as the Messinian Salinity Crisis, the formation and/or transformation of orographic features, and the Quaternary climatic oscillations have strongly influenced the distribution and genetic diversification of species occurring in the area. Currently, species' histories are influenced by anthropogenic effects (e.g. translocations, pet and food trade), which can produce severe impacts. The Mediterranean pond turtle, *Mauremys leprosa*, occurs widely throughout the Iberian Peninsula and most of the Maghreb region. Two *M. leprosa* subspecies are currently recognized: *M. l. saharica* (located south of the Atlas Mountains till Tunisia) and *M. l. leprosa* (north of the Atlas Mountains and in the Iberian Peninsula). In this work we aim to explore the effect of past climatic oscillations and landscape barriers produced in the current patterns of genetic diversity and structure in *M. leprosa*. We sequenced mtDNA, (cytochrome *b* and control region) and genotyped nuDNA (11 polymorphic microsatellite loci) in ca. 500 samples. Our goals are to: 1) identify the geographic origin of diversification; 2) determine the genetic structure within each subspecies; 3) evaluate the effect of major barriers (strait of Gibraltar and Atlas Mountains) to gene flow, 4) identify potential contact zones between both subspecies; and 5) establish a genetic dataset to allocate individuals of unknown origin. We identified a high level of substructuring in Africa (three sublineages for *M. l. leprosa* and four to *M. l. saharica*) originated during the Pliocene-Pleistocene. A secondary contact zone between the two subspecies occurs along the Rif and Middle Atlas mountains. A recent and rapid expansion occurred throughout the Iberian Peninsula, and potential translocation might lead to the observed high degree of admixture, preventing the accurate allocation of unknown individuals. Overall, this study sheds new light into the role of both geographical and climatic features in the genetic diversity patterns of *Mauremys leprosa*, and complements the current knowledge on the importance of North Africa as refugia in the last glacial maximum.

Biogeographic barriers, *Mauremys leprosa*, Mediterranean Basin, phylogeography.

Efeitos do clima presente e passado na distribuição actual e diversidade genética da salamandra-de-costelas-salientes (*Pleurodeles waltl*)

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A salamandra-de-costelas-salientes (*Pleurodeles waltl*) é um endemismo ibero-magrebí com populações a norte e a sul do Estreito de Gibraltar. Com o objectivo de inferir a sua história evolutiva, recolheram-se amostras de 478 indivíduos provenientes de 55 populações espalhadas pela área de distribuição da espécie, incluindo a Península Ibérica e o Noroeste de África, e analisou-se uma série de marcadores mitocondriais (ND4, 856 pb) e nucleares (10 microssatélites polimórficos) de forma a descrever a sua estrutura e diversidade genética. Construíram-se depois modelos lineares generalizados da distribuição da espécie, com base tanto nos pontos de ocorrência recolhidos neste estudo, como nos dados registados nos atlas herpetológicos de Portugal, Espanha e Marrocos em quadrículas UTM de 10x10 km. Os descritores ambientais da distribuição da espécie foram selecionados a partir de um conjunto de 19 variáveis bioclimáticas, utilizando critérios de informação e significância. Os modelos foram avaliados com base na sua calibração e capacidade de discriminação, e projectados ao passado para inferir as áreas climaticamente favoráveis durante o último período inter-glacial e durante o último máximo glacial de acordo com dois cenários climáticos hipotéticos: CCSM e MIROC. Finalmente, comparou-se as áreas climaticamente favoráveis postuladas por estes modelos com a diversidade genética actual das populações da espécie, medida pela diversidade nucleotídica, riqueza alélica, alelos privados e heterozigidade. A diversidade genética mostrou-se significativamente correlacionada com a intersecção entre a favorabilidade ambiental durante o último período inter-glacial e durante o último máximo glacial segundo o cenário CCSM – isto é, as populações actuais de salamandra-de-costelas-salientes são geneticamente mais diversas em regiões que, de acordo com estes modelos, se mantiveram ambientalmente favoráveis através das glaciações. Estas regiões podem, portanto, ter servido como refúgios glaciais a partir dos quais as áreas atualmente habitadas foram colonizadas.

Present and past climatic effects on the current distribution and genetic diversity of the Ribbed Newt (*Pleurodeles waltl*)

The Ribbed Newt (*Pleurodeles waltl*) is an Ibero-Maghrebian endemic with populations north and south of the Strait of Gibraltar. With the aim of inferring its evolutionary history, we collected samples of 478 individuals from 55 populations throughout its range, including the Iberian Peninsula and north-western Africa, and analysed both mitochondrial (ND4, 856 bp) and nuclear markers (10 polymorphic microsatellite loci) to describe patterns of genetic diversity and structure. We then built generalized linear models of this species' distribution, based both on the point records we collected and on presence/absence data on UTM 10x10-km cells as documented in the Portuguese, Spanish and Moroccan herpetological atlases. Predictors from a pool of 19 bioclimatic variables were selected for each model using information and significance criteria. We further evaluated the models using calibration and discrimination measures. We then projected these models back in time, to infer climatically favourable areas during the last interglacial period and the last glacial maximum, using the same bioclimatic variables according to two hypothetical climatic scenarios: CCSM and MIROC. Finally, we compared the inferred past favourable areas with the current genetic diversity of Ribbed Newt populations, as measured by nucleotide diversity, allelic richness, private alleles, and heterozygosity. Genetic diversity was significantly correlated with the intersection between environmental favourability in the last interglacial period and in the last glacial maximum according to the CCSM scenario – i.e., current Ribbed Newt populations are genetically more diverse in regions that, according to these models, have remained environmentally favourable through the glaciations. These regions may thus have served as glacial refugia from which currently inhabited areas were subsequently colonized.

Climate change, distribution modelling, glacial refugia, intra-specific diversity, phylogeography.

Fragmentação em populações urbanas: efeitos do isolamento recente na estrutura populacional e demografia em *Salamandra salamandra* na cidade de Oviedo

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A perda e fragmentação do habitat devido à crescente urbanização têm um grande impacto na biodiversidade, apesar de algumas espécies terem conseguido sobreviver nas cidades. A presença de salamandra de pintas amarelas (*Salamandra salamandra*) na cidade de Oviedo (Espanha) constitui um caso incrível de adaptação a áreas urbanas. A viviparidade (nascimento de juvenis completamente desenvolvidos) evoluiu durante a época do Plioceno-Pleistoceno, o que lhes permitiu colonizar ambientes mais severos, tais como as cidades. Esta espécie persiste em pequenos grupos de indivíduos em parques ou pequenos jardins, estando hipoteticamente isolados devido à sua baixa capacidade de dispersão. Este isolamento reflecte o crescimento urbano desta cidade desde a construção de edifícios eclesiásticos (ex. uma catedral e um mosteiro) rodeados por muralhas desde o séc. VIII. É de esperar que a diferenciação genética tenha aumentado nestas pequenas populações urbanas isoladas durante 1000 anos, devido à deriva genética. A redução de habitat e o baixo tamanho populacional, aliado à informação disponível sobre o desenvolvimento urbano no passado, faz de Oviedo um laboratório natural para testar as hipóteses acima mencionadas.

Neste estudo foram usados 15 microssatélites para genotipar 180 salamandras de pintas amarelas em edifícios históricos e parques urbanos espalhados pela cidade. Nós pretendemos: (1) analisar os níveis de estruturação populacional em Oviedo; (2) avaliar os valores de diversidade genética e consanguinidade nos locais amostrados; (3) estimar parâmetros demográficos nos locais amostrados; e (4) avaliar os efeitos do isolamento histórico e da demografia nos padrões genéticos contemporâneos em Oviedo. Resultados preliminares indicam uma forte estruturação genética na maior parte das populações amostradas. O caso mais evidente corresponde a três populações localizadas dentro das muralhas separadas apenas por 300 m, no qual exibem os níveis de diferenciação genética mais elevados (F_{ST} emparelhado > 0.20). Os resultados obtidos neste estudo são discutidos, tendo o potencial para contribuir para uma melhor compreensão dos efeitos do isolamento recente na demografia e na diversidade genética nas populações naturais.

Fragmentation in urban populations: the effects of recent isolation on population structure and demography of *Salamandra salamandra* in Oviedo

Habitat loss and fragmentation produced by the growing urbanization have a profound impact on biodiversity, although some species have managed to survive within cities. Fire salamanders (*Salamandra salamandra*) of the city of Oviedo (Spain) constitute a remarkable example of adaptation in urban areas. These populations evolved to viviparity (parturition of fully metamorphosed terrestrial juveniles) during Pliocene-Pleistocene times, which allow them to inhabit harsher environments, such as cities. The species persists in small groups of individuals within parks or small gardens, being hypothetically isolated due to their low dispersal ability. This isolation would reflect the urban growth of this city since the construction of ecclesiastic buildings (e.g. a cathedral and a monastery) surrounded by an historical wall since the VIII century. It is expected that genetic differentiation has increased in these small isolated urban populations through genetic drift during the last 1000 years. The reduced habitat and small population sizes, allied with the information available regarding historical urban development, makes Oviedo a natural laboratory to test the above hypotheses.

In this work we used 15 microsatellites to genotype 180 fire salamanders from historical buildings and urban parks throughout the city. We aim: (1) to analyse the levels of population structure pattern in Oviedo; (2) to assess genetic diversity and inbreeding values across sampled sites; (3) to estimate demographic parameters within each sampled site; and (4) to evaluate the effects of historical isolation and demography on current genetic patterns in Oviedo. Preliminary results indicate a strong genetic structuring across most of the sampled populations. The most evident case comprises three populations within city walls which are separated only by 300 m, which exhibit the highest genetic differentiation (pairwise F_{ST} > 0.20). The results obtained in this study are discussed, having the potential to contribute to a better understanding of the effects of recent isolation on demography and genetic diversity of natural populations.

Recent isolation, demography, *Salamandra salamandra*, urbanization, genetic diversity.

Atrapada por el clima: refugio interglacial y reciente expansión poblacional en la víbora Ibérica endémica *Vipera seoanei*

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La variabilidad climática es una de las principales fuerzas que afectan a los procesos de diversificación y restricción de especies a áreas específicas y, por lo tanto, tiene un gran impacto en los patrones biogeográficos de las especies. Este estudio pretende inferir el papel del clima en la historia evolutiva de la víbora ibérica endémica *Vipera seoanei*. Combinamos análisis genéticos con modelos de nicho ecológico. Los análisis genéticos, basados en la secuenciación de dos marcadores mitocondriales (cyt b y ND4) para 60 muestras de tejido de ejemplares de toda la distribución de la especie, incluyen análisis filogenéticos y filogeográficos, interpolaciones espaciales de variabilidad y diversidad genética, e identificación de la procedencia geográfica del ancestro común más reciente de la especie. Los modelos ecológicos, basados en 352 localidades (a 1x1 km), combinaron seis algoritmos y proyecciones a las condiciones climáticas pasadas (Último Período Interglacial -LIG-, Último Máximo Glacial -LGM-), así como la identificación de áreas climáticas estables. La estructura filogeográfica superficial y la distribución de la baja diversidad de haplotipos en *V. seoanei* evidencian tres principales procesos evolutivos: aislamiento de *V. berus* en el Plioceno medio-tardío, persistencia de una sola población en el noroeste de Iberia en el Pleistoceno medio y reciente expansión hacia el noreste de la Península Ibérica. Las proyecciones se ajustan espacialmente a los resultados genéticos, indicando contracciones del rango de distribución hacia el noroeste de Iberia durante el LIG y expansiones durante el LGM. Este estudio ejemplifica cómo la combinación de análisis filogeográficos y ecológicos constituyen eficaces herramientas para inferir escenarios evolutivos y respuestas de las especies a las oscilaciones climáticas del Pleistoceno. *Vipera seoanei* respondió de acuerdo a un modelo templado-frío y se ajusta a un ejemplo simplificado de especie "R" donde los períodos de calentamiento interglacial probablemente causaron importantes reducciones de su rango de distribución con la persistencia en un solo refugio en el noroeste de la Península Ibérica. El único linaje de ADN mitocondrial observado no apoya la diferenciación a nivel de subespecie en *V. seoanei*. Nuestro trabajo pone de manifiesto la importancia del clima en la explicación de los procesos evolutivos y los patrones biogeográficos actuales de especies con distribuciones restrictas.

Trapped by climate: interglacial refuge and recent population expansion in the endemic Iberian adder *Vipera seoanei*

Climate variability is a major force affecting diversification processes and restricting species to specific areas and thus, it has important impacts on species biogeographic patterns. This study aims to infer the role of climate in the evolutionary history of the endemic Iberian adder *Vipera seoanei*. For this purpose, we combined genetic analyses with ecological niche-based modelling. Genetic analyses, based on sequencing of two mitochondrial markers (cyt b and ND4) for 60 tissue samples from specimens covering the whole species distribution, include phylogenetic and phylogeographic analyses, spatial interpolations of genetic variability and diversity, and identification of putative geographical origin of the most recent common ancestor of the species. Ecological modelling, based on 352 distributional records (at 1x1 km of resolution), involved the combination of six modelling algorithms and projections to past conditions (Last Inter Glacial -LIG-, Last Glacial Maximum -LGM-) and the identification of climatic stable areas. The shallow phylogeographic structure and distribution of the low haplotype diversity evidence three main evolutionary processes, starting with long term isolation from *V. berus* at the middle-late Pliocene, posterior single population persistence in north-western Iberia at middle Pleistocene, and recent expansion to north-eastern Iberia. Projections to past periods spatially fit genetic results, indicating range contractions to north-western Iberia during the LIG and expansions during the LGM. This study exemplifies how the combination of phylogeographic and ecological niche-based models are powerful tools for inferring evolutionary scenarios and responses of species to Pleistocene climatic oscillations. *Vipera seoanei* responded accordingly to a cold-temperate model and fits a simplified example of "R" type species where interglacial warming periods during the Pleistocene probably caused major range reductions with persistence in a single refuge in northwestern Iberia. The single mtDNA lineage observed in this study does not support the differentiation at subspecific level in *V. seoanei*. Our work highlights the importance of climate in explaining evolutionary processes and current biogeographical patterns of species with restrictive ranges.

Ecological niche-based models, geographic variability, phylogeography, Pleistocene, spatial interpol.

Los mecanismos asociados con la especiación: descripción de la dieta de los reptiles de Socotra

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La isla de Socotra, de origen continental, es una de las más aisladas del mundo y cuenta con una increíble comunidad de reptiles, compuesta por 27 especies endémicas. Estudios filogenéticos demostraron monofilia en algunos linajes de reptiles a partir de un ancestro continental con posterior especiación dentro de la isla. Las especies endémicas de los géneros *Haemodracon* y *Hemidactylus* representan un excelente modelo de investigación ya que se distribuyen a lo largo de toda la isla y muestran una gran variabilidad en tamaños corporales y hábitats ocupados. Este estudio tiene como objetivo describir la dieta de seis especies de geckos de los géneros *Haemodracon* y *Hemidactylus* y sus correlaciones con el tamaño corporal, el hábitat que ocupan y la filogenia, con el fin de explorar los mecanismos que producen especiación en los reptiles de Socotra. El estudio se basó en el análisis de muestras fecales recogidas en dos expediciones científicas en 2013 y 2014. La similitud en la composición de las dietas se analizó mediante el índice de Bray-Curtis y ANOSIM. Los resultados indicaron que los geckos presentaban una amplia variedad de presas en su dieta (10 órdenes de artrópodos), un hecho que sugiere hábitos alimenticios oportunistas. El orden Coleoptera fue el taxón más consumido. El análisis de similitudes (ANOSIM) indicó diferencias significativas entre especies en sus dietas ($R=0.081$, $p=0.015$). El dendrograma mostró cuatro grupos de especies en función de sus similitudes en la dieta: (1) *H. granti*, especie que vive en las montañas, (2) *H. pumilio*, especie de pequeño tamaño que vive en el suelo, (3) *H. trachyrhinus*, especie de pequeño tamaño que vive en la vegetación y (4) un grupo compuesto por tres reptiles grandes que comúnmente utilizan hábitats verticales (*H. riebeckii*, *H. dracaenacolus* y *H. inintellectus*). Este resultado indica que la dieta entre los seis geckos no se ha preservado dentro de la filogenia, sino que ha variado según el tamaño de las especies, su distribución geográfica y los hábitats que ocupan.

The mechanisms associated with speciation: diet description of Socotra reptiles

The Socotra Island, of continental origin, is one of the most isolated islands of the world and has an amazing reptile community composed by 27 endemic species. Phylogenetic studies demonstrated monophyly in some reptile lineages from mainland ancestors with further intra-island speciation. Endemic species of the genera *Haemodracon* and *Hemidactylus* represent excellent models for research as they are distributed along the whole island, and show a high variability in body size and occupied habitats. This study aimed at describing the diet of six species of geckos of the genera *Haemodracon* and *Hemidactylus*, and its correlation to body size, habitat, and phylogeny, in order to explore the mechanisms that produced speciation in Socotran reptiles. The study was based on analysis of faecal samples collected in two scientific expeditions in 2013 and 2014. The similarity in the composition of the diets was analyzed using the Bray-Curtis index and ANOSIM. The results indicate that geckos presented a wide variety of prey items in their diet (10 orders of arthropods), this fact suggesting their opportunistic dietary habits. The Coleoptera Order was the most consumed prey taxon. The analysis of similarities (ANOSIM) indicated significant interspecific differences in diet ($R=0.081$, $p=0.015$). The dendrogram showed four groups of species according to their dietary similarities: (1) *H. granti*, a species that lives in mountains, (2) *H. pumilio*, a ground-dwelling small-sized species, (3) *H. trachyrhinus* an arboreal/shruboreal and small-sized species, and (4) a group composed by three big reptiles that commonly use vertical habitats (*H. riebeckii*, *H. dracaenacolus* and *H. inintellectus*). This result indicates that the diet among the six geckos was not preserved within the phylogeny but varies depending of their morphologies, habitats, and geographic distribution.

Speciation, trophic ecology, reptile, Socotra.

Análise biogeográfica de *Psammophis schokari* no Norte de África: Um modelo para avaliar corredores trans-Saarianos

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Desde o surgimento do Saara há cerca de 7Ma, o Norte de África tem tido repetidos ciclos de condições húmidas e áridas. Para muitas espécies tais oscilações resultaram em eventos de isolamento e contacto secundário ao longo de zonas de contacto. Vários corredores hipotéticos compostos de terras altas, sistemas fluviais e áreas costeiras podem ter facilitado a dispersão Norte-Sul de espécies. *Psammophis schokari* distribui-se do Maghreb até à Índia, com populações a Norte e Sul do Saara e várias isoladas nas montanhas centro-Saarianas. Actualmente estão identificadas quatro linhagens, três no Maghreb e uma em Israel. O objectivo geral deste estudo foi explorar a hipótese de corredores do deserto, especialmente o Saara Atlântico, num enquadramento filogenético e ecológico com *P. schokari* como modelo.

Através de amostragem melhorada, uso de marcadores moleculares mitocondriais e nucleares, e modelação ecológica do nicho, propusemo-nos a responder às seguintes questões: 1) Como está espacialmente distribuída a variabilidade genética?; 2) Que factores ambientais estão presentemente relacionados com a ocorrência da espécie?; 3) Onde se localizam as áreas climáticas adequadas à ocorrência da espécie no Presente?; 4) Onde se localizavam as áreas climáticas adequadas à ocorrência da espécie no Último Máximo Glaciar (LGM) e Último Inter-Glaciar (LIG)?; e 5) Onde se localizavam as rotas de dispersão mais prováveis e zonas climáticas estáveis para a ocorrência da espécie?

Até agora detectámos três linhagens aparentemente alopátricas de *P. schokari* no Norte de África. As linhagens Mauritânica e Marroquina aparentam contactar na região do Cabo Barbas. O posicionamento filogenético dos espécimes da Argélia e Tunísia sugere refúgios locais e contacto recente de populações costeiras e das montanhas Saarianas. Os modelos ecológicos indicam alta conectividade de populações através das montanhas Centro-Saarianas durante o LGM, e isolamento no Presente e LIG. Os resultados para o Saara Atlântico e corredor do Mar Vermelho sugerem uma reduzida conexão N-S em períodos mais quentes. A costa Mediterrânea e montanhas Sul-Argelinas e do Norte do Chad são áreas estáveis de ocorrência prevista e provavelmente refúgios. Tudo isto concorda com os resultados filogenéticos. Um corredor Saeliano E-W é sugerido, mas não detemos amostras disponíveis para testá-lo. No geral a metodologia mostra potencial para resolver estas questões, mas são necessárias ainda mais amostras e análises.

Biogeographic assessment of *Psammophis schokari* in North Africa: a model for assessing trans-Saharan corridors

Since the first appearance of the Sahara desert around 7Mya, North Africa has suffered a repeated intercalation of arid and humid conditions. For many species such oscillations resulted in events of isolation and secondary contact along contact zones. A series of hypothesised corridors composed by highlands, fluvial systems or coastal areas are thought to have facilitated north-south dispersion of species. *Psammophis schokari* ranges from the Maghreb to India, presenting populations North and South of the Sahara and several isolated populations in central Sahara Mountains. Currently, four main lineages are identified, three in the Maghreb and one in Israel. The general aim of this study was to tap into the hypothesis of desert corridors, especially the Atlantic Sahara, in a phylogenetic and ecological framework using *P. schokari* as a model.

Through increased sampling, use of mitochondrial and nuclear molecular markers and ecological niche modelling, we aim to answer the following questions: 1) How is the genetic variability spatially structured?; 2) Which environmental factors are presently related to the species occurrence?; 3) Where are located the suitable climatic areas for the species occurrence in current times?; 4) Where were located the suitable climatic areas for the species occurrence in the Last Glacial Maximum (LGM) and Last Inter Glacial (LIG)?; and 5) Where were located the most probable routes of dispersal and stable climatic areas for the species occurrence?

Until now we have detected three apparently allopatric lineages of *P. schokari* in N Africa. Moroccan and Mauritanian lineages seem to contact in Cap Barbas region. Phylogenetic placement of Algerian and Tunisian specimens suggests local refugia and recent contact of coastal and central Saharan Mountains populations. Ecological models indicate high degree of connectivity of *P. schokari* populations across central Saharan Mountains during the LGM, and isolation at present and LIG. Results for the Atlantic Sahara and Red Sea corridor suggest a decreased N-S connection in warmer periods. The Mediterranean coast and mountains in Southern Algeria and Northern Chad are stable predicted-occurrence areas and

likely act as refugia. All this is concordant with the phylogenetic results. A Sahelian E-W corridor during the LGM is also suggested, but no samples are available to test this. Overall the methodology shows the potential to help solve these questions, but more samples and further analysis are needed.

Biogeography, Sahara, *Psammophis*, ecological niche-based models, phylogeography, paleoclimatic data.

¿Muestran las salamandras vivíparas (*Salamandra salamandra*) divergencia de nicho ambiental?

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La salamandra común, *Salamandra salamandra*, es una de las pocas especies de vertebrados que muestran variación intraespecífica en el modo reproductor: un modo ancestral, ovoviviparismo, en el cual las hembras paren larvas, y un modo derivado, viviparismo, donde las hembras paren directamente juveniles terrestres. Mientras el primer modo se encuentra ampliamente distribuido por la mayor parte del rango geográfico de la especie, el segundo está restringido, principalmente, a lo largo de la Cordillera Cantábrica y a dos islas gallegas en el litoral atlántico. La transición al viviparismo implica la eliminación de la fase larvaria y la semi-independencia del agua, y se asume que conlleva un aumento en el potencial de colonización de nuevos hábitats. Sin embargo, se conoce poco sobre los potenciales factores (ecológicos, fisiológicos, genéticos) que pueden explicar el origen de esta transición. La hipótesis del “clima seco” sugiere que la posible falta de agua, en el pasado, en las islas y en los suelos kársticos de las montañas cantábricas pudo haber desencadenado este cambio de estrategia reproductiva. Para comprender mejor los factores ecológicos y ambientales relacionados con esta transición hemos (1) analizado la divergencia de nicho entre linajes vivíparos y ovovivíparos en la Península Ibérica, y (2) evaluado la posibilidad de un cambio de nicho en el linaje vivíparo mediante la reconstrucción de su distribución potencial en el pasado. Primero analizamos las relaciones filogenéticas de los principales linajes de *S. salamandra* para rechazar la monofilia del linaje vivíparo y determinar sus relaciones con otros linajes. Posteriormente elaboramos modelos de nicho ecológico para los principales linajes vivíparos y ovovivíparos utilizando 500 muestras georreferenciadas que fueron en su mayoría analizadas con ADN mitocondrial (1100 pb), nuclear (15 microsátélites) y caracteres morfológicos (patrón de coloración). Medimos el solapamiento de nicho utilizando variables geológicas y climáticas. Por último, proyectamos la distribución de cada linaje para el último interglaciar (~130.000 años) y último máximo glacial (~21.000 años). Los resultados preliminares muestran evidencias de divergencia de nicho entre los linajes vivíparos y ovovivíparos. Por otro lado, la ausencia de áreas climáticamente adecuadas en el último interglaciar sugiere la posibilidad de un cambio de nicho en el linaje vivíparo. Discutimos los mecanismos históricos y adaptativos que pudieron haber desencadenado el cambio de estrategia reproductiva en este sistema biológico único.

Do viviparous fire salamanders (*Salamandra salamandra*) show environmental niche divergence?

The fire salamander, *Salamandra salamandra*, is one of the few vertebrate species with intraspecific variation in reproductive modes: an ancestral mode, ovoviviparity (larviparity), in which females give birth to aquatic larvae, and a derived mode, viviparity (pueriparity), in which females give birth to fully metamorphosed terrestrial juveniles. While the former is widely distributed along most of the species range, the latter is restricted to northern Iberian populations along the Cantabrian Mountains and two islands in the Atlantic coast of Galicia. The transition to viviparity implies eliminating the aquatic larval stage and thus semi-independency from water, and is assumed to increase the potential to colonize new habitats. However, the potential factors (ecological, physiological, genetic) explaining the origin of this transition are yet poorly understood. The “dry-hypothesis” has been suggested to explain the transition to viviparity, postulating a potential lack of water in the islands and in karstic limestone along the Cantabrian Mountains in the past that would have triggered the reproductive shift. To get a better understanding of the environmental and ecological factors related to the switch in reproductive modes, we here 1) assessed the potential niche divergence between viviparous and ovoviviparous lineages in the Iberian Peninsula, and 2) evaluated potential niche shifts in the viviparous lineage by the reconstruction of its past range. We used a well-resolved phylogenetic tree including representatives of all major clades in *S. salamandra* to reject the monophyly of the viviparous lineage. Then we derived independent ecological niche models for each major viviparous and ovoviviparous lineage based on ca. 500 georeferenced samples analyzed with mtDNA (1100bp), nuDNA (15 microsattellites) and morphological characters (coloration pattern). We measured niche overlap based on bioclimatic and geologic variables. We projected the distribution of each lineage to the Last Inter Glacial (LIG, ~130,000 yrs) and to the Last Glacial Maximum (LGM, ~21,000 yrs). Preliminary results show evidence for niche divergence between ovoviviparous and viviparous lineages. On the other hand, lack of suitable climatic areas in the LIG suggest the possibility of niche shifts in the viviparous lineage. We discuss the historical and adaptive mechanisms that may have triggered the reproductive shift in this unique biological system.

Salamandra salamandra, viviparity, ENMs, phylogeny, niche shift.

Disposición de la distribución de la población de gallipato (*Pleurodeles waltl*) en Cataluña y variables que influyen, atendiendo a su abundancia

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En las sierras litorales del sur de Cataluña se encuentra el límite norte por el Mediterráneo del área de distribución del gallipato (*Pleurodeles waltl*). A pesar de ser catalogado como abundante en 2002 (Montori et al 2002), cuando en el año 2007 se actualizaron los datos por parte del Servicio de Biodiversidad y Protección de los Animales (BIPA) del gobierno de Cataluña se observó una disminución del 50% del área ocupada por la especie, pasando a estar presente en sólo 10 cuadrículas UTM 10x10 respecto a las 20 de los últimos muestreos. Desde entonces, se ha estado desarrollando un seguimiento más exhaustivo de la población hasta este año 2014, con el fin de detectar las causas de dicho declive de efectivos y atajar esta tendencia negativa dentro del territorio catalán.

Tras muestreos repetitivos durante 7 años, en los que se han muestreado y categorizado 130 micro-hábitats dentro de la zona de distribución de la especie, se han determinado las abundancias cuantitativas de la especie en cada punto (un total de 67), detectándose que dichas abundancias de individuos están relacionadas con la disposición de cada una de las charcas dentro del su área de distribución. Por consiguiente, se establece una mayor abundancia en el centro del área de distribución y una disminución de ésta conforme nos vamos alejando del centro. Este fenómeno también nos indica que en dicho centro poblacional se encuentran el mayor número de puntos para la reproducción y que en las charcas reproductivas las abundancias son mayores.

Por otro lado, se han estudiado las variables ambientales que pueden afectar a las abundancias cuantitativas de la población, estableciendo de este modo una relación entre ellas. La relación establecida entre las variables permite crear un modelo que consiga explicar las diferencias entre las abundancias de cada balsa, concluyendo cuáles son los requisitos medioambientales y el número de individuos que tiene que albergar cada micro-hábitat acuático para que la población sea viable.

Disposition of the population distribution of Spanish ribbed newt (*Pleurodeles waltl*) in Catalonia and factors influencing, attending to their abundance

The coastal mountains of Southern Catalonia represent the northern limit of the Iberian ribbed newt (*Pleurodeles waltl*) range in the Mediterranean side. Despite being listed as abundant in 2002 (Montori et al 2002), we observed a decrease of 50% of the area occupied by the species in 2007 when the data was updated by the Biodiversity and Protection of Animals Service (BIPA) of the Government of Catalonia, being found in only 10 UTM 10x10 grids compared to the 20 grids found in former samplings. Since then, a more thorough monitoring of the population has been made until 2014, in order to identify the causes of the decline of effective and halt this negative trend in the Catalan territory.

After undertaking repetitive sampling for 7 years, where 130 micro-habitats within the distribution area of the species have been sampled and categorized, we have determined quantitative abundances of the species at each point (total of 67), detecting that such individual abundances are related to the disposition of each of the ponds within their range. Therefore, a greater abundance is found in the core of its range and it declines as we move away from the core. This phenomenon also indicates that in this population center we find most of the reproduction points and that in breeding ponds the abundances are higher.

On the other hand, we have studied the environmental variables that can affect the quantitative abundance of the population, thereby establishing a relationship between them. The relationship established between the variables lets us to create a model that could explain the differences between the abundances of each raft, concluding which are the environmental requirements and the number of individuals that must accommodate each aquatic micro-habitat for the populations to be viable.

Abundance, disposition, distribution, Spanish ribbed newt.

Análisis de la serie de datos (1993-2013) de marcado de tortuga boba (*Caretta caretta*, Linnaeus 1758) en el Mediterráneo occidental y el Golfo de Cádiz (Atlántico oriental) en el marco del programa de investigación de grandes pelágicos del IEO

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El Centro Oceanográfico de Málaga del Instituto Español de Oceanografía (Ma-IEO) es pionero en el marcado de tortugas marinas en el Mediterráneo occidental y Golfo de Cádiz. Se han utilizado diversos tipos de marcas: marcas de plástico de ICONA y metálicas del ACCSTR (Universidad de Florida); propias (en el proyecto European Marine Turtles Program EMTP, financiado por la UE). La Asociación Herpetológica Española (AHE) en colaboración con el Instituto Español de Oceanografía (IEO) a través de su Centro de Málaga, y la Oficina de Anillamiento (OA) de la Dirección General de Conservación de la Naturaleza, puso en marcha en 1997 un Programa de marcado y conservación de tortugas marinas homologado utilizando marcas metálicas Inconel® aportadas por el M.^o, establece los responsables por áreas geográficas y un responsable de la base de datos y se crean grupos acreditados de expertos gestionados desde la AHE. La AHE responsabiliza al Ma-IEO de la base de datos de marcado y la coordinación del mar de Alborán, Golfo de Cádiz, Ceuta y Melilla (norte de África). Estas investigaciones se realizan en el marco del proyecto de grandes pelágicos coordinado desde Ma-IEO. Durante la primera fase hasta el lanzamiento de EMTP se marcaron 338 tortugas con marcas preferentemente del ACCSTR. Durante el EMTP (1999-2001), se marcaron 551 tortugas. Desde 2001 hasta 2010 se colocaron 333 nuevas marcas, y desde el año 2011 se han colocado 17 marcas metálicas combinadas con microchip. En la actualidad el grupo Ma-IEO sigue realizando marcado de tortugas marinas, preferentemente durante operaciones de investigación y seguimiento de la actividad pesquera con observadores a bordo en las zonas que operan las embarcaciones de pesca de especies de túnidos y afines, en el Mediterráneo occidental y el Golfo de Cádiz. El grupo Ma-IEO continúa hasta el presente con la labor de marcado iniciada en el siglo XX. El último resumen del programa de marcado de tortugas marinas del Ma-IEO fue publicado en 2009. Este documento actualiza los datos de marcado de tortugas marinas aportando nuevos datos de marcado por año y área (Mediterráneo occidental y el Golfo de Cádiz), analiza los resultados del marcado (números por área, año y periodo del año) y presenta los datos de las recapturas obtenidas en ese periodo. Aunque la mayor parte de las recapturas se han realizado en la misma área de marcado, en ocasiones se han obtenido recapturas en aéreas alejadas, tanto del Mediterráneo como del Atlántico. Se analizan los datos de captura y marcado de tortugas marinas en relación con los datos pesqueros (área de pesca, artes, captura, esfuerzo, by-catch de tortugas) y se representan geográficamente los principales resultados.

Analysis of data series (1993-2013) of tagged marine turtle Loggerhead (*Caretta caretta*, Linnaeus 1758) from the western Mediterranean and Gulf of Cádiz (eastern Atlantic) carried out by the IEO Large pelagic research program

The Málaga Center of the Spanish Oceanographic Institute (Ma-IEO) is pioneering in massive tagging of sea turtles in the western Mediterranean and Gulf of Cádiz. Different types of tags were used initially: plastic ICONA tags and metal tags provided by the ACCSTR (University of Florida); own tags (prepared for the European Marine Turtles Program, EMTP, financed by the EU). The Spanish Herpetological Association (AHE) launched in 1997 in collaboration with the Ma-IEO and the Tagging Office (TO) of the Spanish Directorate General for Nature Conservation, a national tagging and conservation program of sea turtles using Inconel® tags provided by the TO, nominated the responsible for each Spanish geographic coastal area and gave the responsibility for managing the joint database to Ma-IEO. Different groups of experts accredited by the AHE were created. The Ma-IEO experts' group was in charge of tagging coordination in the Alborán Sea and Gulf of Cádiz, Ceuta and Melilla (North Africa). These research activities are carried out within the Large Pelagic Project conducted in Ma-IEO. During the first period until launching the EMTP, 338 turtles were tagged with ACCSTR tags mainly. During the EMTP (from 1999 to 2001), 551 turtles were tagged. From 2001 to 2010, 333 new metal tags were used in sea turtles. Since 2011, seventeen microchips have been placed combined with metal tags. Tagging sea turtles was preferably done by on board scientific observers in the western Mediterranean and the Gulf of Cádiz during the research and monitoring of selected fisheries in fishing areas operated by the Spanish vessels targeting tuna and tuna-like species.

The Ma-IEO group continues with the tagging work initiated in the XX century to the present. The latest summary of the tagging program of marine turtles by Ma-IEO was published in 2009. The present document updates the information on sea turtle tagging providing new data by year and area (in the western Mediterranean and the Gulf of Cádiz), analyzes the tagging results (numbers per area, year and period of the year) and presents the data and information from the recaptures obtained in that period. Although most of the recaptures were made in the same area of tagging, some were obtained in distant areas in the Mediterranean and the Atlantic. Data from tagging of marine turtles are analyzed in relation to fisheries data (fishing area, gear, catch, effort, by-catch of turtles) and the main results are represented geographically.

Loggerhead, tagging, Mediterranean, Gulf of Cádiz.

Anidación de tortugas marinas en España: pasado, presente y futuro

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Existen registros de anidación de tortuga careta en el mediterráneo español desde el siglo XIX. En Canarias hay indicios de anidación de tortuga laúd. Recientemente, el número de nidos detectados en playas españolas se ha incrementado sensiblemente y parece que la anidación se concentra en determinados años (2001, 2006, 2011 y 2014). Todos los nidos depositados por tortugas en la mitad sur peninsular se desarrollaron con éxito y con tiempos de incubación similares a otras zonas estables de anidación de *Caretta caretta*. El análisis genético de algunas madres indica un origen mixto atlántico y mediterráneo.

Predicciones de calentamiento del clima publicadas por el IPCC en 2014 confirman que las condiciones de anidación de tortuga careta en Cabo Verde, única zona estable de anidación de tortuga careta en todo el Atlántico oriental no serán adecuadas al final del siglo XXI. Estudios ecológicos de zonas de alimentación y playas indican que el Norte de África y mitad sur de España mantendrían una producción razonable de ambos sexos dentro de 100 años. Estos resultados señalan a playas españolas como lugares potenciales importantes de dispersión de anidación de tortuga careta en respuesta al calentamiento del clima en las próximas décadas. El fracaso de esta dispersión de hábitats reproductores hacia zonas más frías podría provocar la extinción de la Unidad de Manejo Regional de *Caretta caretta* del Atlántico oriental a medio plazo. Se debe planificar y gestionar ya el litoral de la mitad meridional de España, incluyendo Canarias, como zona potencial de anidación de tortuga careta. Se considera relevante estimular y apoyar este importante proceso ecológico cuyo éxito está muy dificultado por impactos ambientales.

Sea turtle nesting in Spain: past, present and future

There are records of loggerhead turtle nesting in the Spanish Mediterranean coast since the XIX century. On the Canary Islands there is also evidence of sporadic leatherback turtle nesting. The number of observed loggerhead nests in the Spanish coast has apparently increased in the past years and seems that nesting concentrates in given years (2001, 2006, 2011 y 2014). All the loggerhead nests observed in the southern half of the Spanish coast have been successful and incubation duration and hatchling sex ratio have been similar to those found on stable loggerhead rookeries. Genetic analysis of nesting females in Spain indicates a mixed Atlantic and Mediterranean origin.

Predictions of global climate warming published by the IPCC in 2014 confirm that nesting and incubation conditions of loggerheads in Cape Verde, the only stable rookery in the Eastern Atlantic coast, will not be suitable for the species at the end of the XXI century. Ecological studies of feeding grounds and beaches indicate that the coast of the northwest of Africa and the southern half of the Iberian Peninsula will maintain in 100 years from now suitable conditions for the reproduction of loggerhead turtles, including the production of hatchlings of both sexes. These results point out to warm Spanish beaches as potential habitat for the dispersal of loggerhead nesting in response to climate warming in the next decades. The failure of this nesting dispersal to colder beaches could cause in the midterm the extinction of the Regional Management Unit of the loggerhead turtle of the Eastern Atlantic.

The Spanish programs of coastal planning and management should consider the southern half of the Iberian Peninsula as well as the Canary and the Balearic Islands as important potential areas for the dispersal of loggerhead nesting. This important ecological process should be supported and reinforced because its success is highly threatened by diverse environmental impacts.

Caretta caretta, nesting, Spain, global warming, dispersal.

Factores implicados en la variación geográfica del tamaño corporal en el tritón ibérico (*Lissotriton boscai*)MIGUEL PEÑALVER ALCÁZAR¹, IÑIGO MARTÍNEZ-SOLANO², FERNANDO SEQUEIRA², PEDRO ARAGÓN¹¹Departamento de Biogeografía y Cambio Global, Museo Nacional de Ciencias Naturales (MNCN-CSIC). José Gutiérrez Abascal 2, 28006 Madrid, Spain.²CIBIO-InBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto. Campus Agrário de Vairão, 4485-661 Vairão, Portugal.

Las causas de la variación geográfica del tamaño corporal en animales han interesado a los biólogos desde el siglo XIX. Sin embargo, en animales ectotermos todavía no hay un consenso en cuanto a las fuentes de variación y los mecanismos involucrados. A nivel intra-específico esta complejidad ha sido abordada mediante diferentes aproximaciones e hipótesis por separado, pero existen muy pocos estudios con una aproximación integradora que examinen simultáneamente varias hipótesis a diferentes escalas y para ambos sexos. El objetivo de este estudio es describir patrones de variación en el tamaño del tritón ibérico (*Lissotriton boscai*) y discernir los mecanismos involucrados. Para ello, se muestrearon 23 poblaciones representativas de todo su rango de distribución. Se tomaron medidas morfológicas, muestras genéticas y se caracterizó el hábitat, tanto a nivel de microhábitat (mediante muestreos *in situ*) como a escala macro-espacial (mediante SIG). Se realizaron diferentes análisis multivariantes para considerar nueve posibles hipótesis causales: principio de conservación del calor, disponibilidad hídrica, disponibilidad hídrica opuesta, disponibilidad de alimento, resistencia a desecación, estacionalidad, resistencia a inanición, depredación y competencia. En conjunto las contribuciones relativas de los predictores utilizados para explicar la variación geográfica de la longitud cabeza-cloaca (LCC) media, máxima y mínima mostraron tanto similitudes como diferencias entre sexos. La temperatura a escala macro-espacial se mostró relevante en ambos sexos, mientras que la estacionalidad de la producción primaria fue más relevante en machos y la competencia intra-específica en hembras. Los residuos de estos modelos no mostraron autocorrelación espacial significativa a nivel geográfico. Los resultados no solo mostraron la existencia de un patrón de variación entre poblaciones, sino también que diferentes mecanismos son responsables de dicho patrón, contribuyendo de forma diferente a la LCC de machos y hembras. Este estudio confirma la importancia de examinar simultáneamente diferentes hipótesis sobre la variación geográfica del fenotipo a diferentes escalas para detectar efectos potencialmente enmascarados.

Determinants of geographical body size variation in the Iberian Newt (*Lissotriton boscai*)

Determinants of geographical body size variation in animals have interested biologists since the XIX century. However, there is no consensus regarding the involved mechanisms or sources of variation in ectotherms. At the intraspecific level, different approximations and hypotheses have often approached this problem separately, but so far few studies have been performed to address it in an integrative fashion, including hypotheses at different scales and for both sexes. Our goal is to describe patterns of body size variation in the Iberian Newt (*Lissotriton boscai*) and to unravel the mechanisms involved. For this, 23 populations representative of the species' distribution were sampled. Morphometric measures and genetic samples were taken, and the habitat was characterized, both at the microhabitat level (*in situ* surveys) and at the macro-scale (using GIS). Different multivariate analyses were performed to address nine likely causal hypotheses, namely: heat balance, water availability, converse water availability, food availability, endurance, seasonality, starvation resistance, predation and competition. Overall, the relative contributions of the predictors used to explain the geographic variation of mean, minimum and maximum snout-vent-length (SVL) showed both similarities and differences between sexes. Temperature at the macro-scale was relevant for both sexes, while seasonality of primary productivity was more important in males and intraspecific competition was relevant in females. Residuals of these models showed no significant geographic spatial autocorrelation. These results not only suggest the existence of a pattern of body size variation among populations, but also that different mechanisms are responsible for this pattern, contributing differently to female and male SVLs. This study highlights the value of considering simultaneously different hypotheses of phenotypic variation at different scales to reveal potentially hidden effects.

Body size variation, newts, intraspecific, microhabitat, macro-scale.

Alometría, modularidad e integración como determinantes de la variabilidad fenotípica: ¿qué nos pueden enseñar las *Podarcis* ibéricas y norteafricanas?

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La comprensión de los mecanismos que subyacen a la generación de la variación fenotípica es un objetivo principal de la biología evolutiva. La alometría de la talla-forma, la modularidad y la integración morfológica, son algunos de los mecanismos de desarrollo principales que determinan el grado de variación en los diferentes rasgos y limitan la covariación entre rasgos. Las lagartijas *Podarcis* ibéricas y norteafricanas son un modelo emergente en los estudios evolutivos y se han utilizado ampliamente para investigar las fuentes de variación fenotípica. En particular, varios estudios han utilizado métodos de morfometría geométrica para estudiar la variación en la forma de la cabeza en relación con, por ejemplo, la ontogenia, el dimorfismo sexual, y el uso del hábitat. Curiosamente, todo indica que la variación de la forma de la cabeza se manifiesta generalmente en una dirección anterior-posterior, independientemente del factor biológico focal. Este hallazgo sugiere que la forma de la cabeza puede estar restringida en este grupo de lagartijas y las convierte en un modelo interesante para investigar cómo diferentes mecanismos contribuyen a la diversidad fenotípica. Utilizamos morfometría geométrica para cuantificar la forma de la cabeza dorsal y lateral en un total de 876 ejemplares, que representan 14 de los 16 linajes del grupo mediante la digitalización de 24 landmarks dorsales y 12 landmarks laterales. A continuación, utilizamos estadística multivariante para investigar la alometría de la talla-forma en ambos sexos y entre linajes. Además, examinamos si la cabeza está estructurada en dos particiones modulares (frontal y distal) e investigamos los patrones de integración entre diferentes regiones de la cabeza dentro y entre linajes. Nuestros resultados sugieren que la cabeza de estas lagartijas se divide en un módulo anterior y otro posterior, con cohesión interna, tanto dorsal como lateralmente. Estos dos módulos presentan una fuerte integración entre sí a través de toda la muestra y dentro de cada linaje por separado. La relación alométrica entre talla y forma aumenta el grado de integración, que sin embargo sigue siendo significativo una vez eliminados los efectos alométricos. Por otro lado, los patrones alométricos y de integración varían ampliamente a lo largo de los diversos linajes, lo cual sugiere un papel importante de estos mecanismos en la determinación de los importantes niveles de diversidad fenotípica observados en las *Podarcis* ibéricas y norteafricanas.

How do allometry, modularity and integration determine phenotypic variation? Insights from Iberian and North African wall lizards

Understanding the mechanisms that underlie the generation of phenotypic variation is a main objective of evolutionary biology. Size-shape allometry, modularity and integration are major developmental mechanisms that determine the degree of variation in different traits and constrain trait co-variation. Iberian and North African wall lizards are an emerging model in evolutionary studies and have been used extensively to investigate sources of phenotypic variation. Particularly, several studies have used geometric morphometric methods to study head shape variation in *Podarcis* wall lizards in relation to e.g. ontogeny, sexual dimorphism and habitat use. Interestingly, all indicate that head shape variation is usually manifested in an anterior-posterior direction, regardless of the focal biological factor. This finding suggests that head shape may be constrained in this group of lizards and makes them an interesting model to investigate how different mechanisms contribute to phenotypic diversity. We used geometric morphometrics to quantify dorsal and lateral head shape in a total of 876 individuals, representing 14 of the 16 lineages of the group by digitizing 24 dorsal and 12 lateral landmarks. After Procrustes superimposition to obtain shape variables, we used multivariate statistics to investigate size-shape allometry across both sexes and across lineages. Further, we tested whether the head is structured in frontal-distal modular partitions and investigated patterns of integration between different head regions within and across lineages. Our results suggest that the lizard head is divided in an anterior and a posterior module with internal cohesiveness in terms of shape variance, both dorsally and laterally. These two modules are highly integrated with each other across the whole sample and within each lineage separately. Size-shape allometry enhances the degree of head shape integration, which nevertheless remains significant once allometric effects are accounted for. However, allometric and integration patterns vary extensively across lineages, suggesting an important role of these mechanisms in determining the remarkable levels of phenotypic diversity observed in Iberian and North African *Podarcis*.

Podarcis, head shape, geometric morphometrics.

An inconvenient truth? Interpopulation variation but not environmental mirroring in the ecophysiology of a temperate lizard

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Interest on vertebrate ecophysiology has recently revived since this evidence is expected to inform mechanistic models to forecast species distribution, especially in the context of climate change. Within a given species, ecophysiological variation across populations depending on the environment represents a chance for adaptation/plasticity in case of environmental shift. Conversely, trait conservativeness may involve range retraction and eventually extinction. Lizards provide excellent experimental models for analysing ecophysiology in terms of adaptation and trait conservatism. Most members of the family Lacertidae inhabit temperate regions under substantial spatial and temporal variation in climate. Within this group, preferred body temperatures (T_p) correlate with several physiological optima and are considered evolutionarily conservative both in the phylogeny and at the interspecific level. However, other lizard families show intraspecific variation related to thermal environment and even to parasitisation (lizard fever). Much less is known about their water ecology, although some studies suggest that body temperature and evaporative water loss (EWL) may trade-off. Here, we tested for ecophysiological adaptation/plasticity in *Podarcis bocagei*, a temperate lizard with a poor phylogeographic substructuring and hence shallow evolutionary history. In spring 2013, we collected a total of 169 adult males belonging to nine populations from Northern Portugal encompassing the spectrum of environmental conditions used by this species (from dunes to agricultural walls, 0-1400 m a.s.l.). Lizards underwent two laboratory tests: 1) the classic T_p experiment using a photothermal gradient during 10 time intervals; and 2) the determination of EWL rates in sealed chambers during 12 hours. For each individual we also scored the prevalence and intensity of parasitisation by hemogregarines (from blood smears). We found highly significant variation and different time profiles across populations for both T_p and EWL, which remained after accounting for snout-vent length (SVL) and body mass, even if EWL was partially explained by lizard size varying within and between populations. Parasitisation also varied between sites, larger lizards tending to attain higher prevalences and intensities but, otherwise, with no effect on ecophysiology. Multivariate analyses including all variables revealed environmental influence on lizard size, EWL and parasitisation but not on T_p . Altogether, these well-supported results illustrate the complexity of lizard ecophysiology while advise against straight-forward adaptive explanations.

Thermal ecology, water ecology, body size, parasite burden, adaptation, *Podarcis bocagei*.

Consecuencias crípticas del cambio global: efectos sobre el estado de salud y la ecología sensorial del género *Iberolacerta*

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Aunque las lagartijas, debido a su termofilia y regulación activa de la temperatura, parecen relativamente invulnerables al calentamiento global, se han documentado extinciones en los cinco continentes como consecuencia de éste y se ha predicho la extinción de casi el 40% de sus poblaciones. A pesar de ser animales heliotermos, la actividad en condiciones demasiado cálidas puede elevar su temperatura corporal a máximos críticos que les obliguen a enfriarse en refugios. Una excesiva permanencia en éstos puede reducir el tiempo dedicado a la alimentación, búsqueda de pareja, defensa del territorio, etc., hasta el punto de afectar a su fitness.

Muchas consecuencias de un aumento de la temperatura a menudo resultan poco evidentes hasta que son estudiadas. Uno de los aspectos apenas considerados es el efecto que éste puede tener sobre la ecología sensorial de las especies; concretamente, sobre la comunicación sexual. De entre los distintos tipos de señales sexuales, las químicas (feromonas), que usan la mayor parte de las especies de lagartijas, son las que más evidentemente dependen de la temperatura. Las señales de una determinada especie o población estarán adaptadas a unas condiciones concretas de temperatura y humedad, maximizando su efectividad de transmisión. Un cambio de esas condiciones, motivado por el calentamiento global, hará que las señales pierdan efectividad, con consecuencias en los mecanismos de selección sexual.

Las especies montañas del género *Iberolacerta* presentan distribuciones discontinuas, con poblaciones aisladas en montañas a distinta altura. Este escenario resulta propicio para examinar las adaptaciones (o su ausencia) y los efectos de las distintas condiciones ambientales de cada población. Así, nos planteamos estudiar si el efecto de la temperatura sobre la eficacia de las señales era diferente entre especies y poblaciones situadas a distinta altitud, dentro del rango de cada especie. Para ello expusimos secreciones femorales de machos procedentes de distintas poblaciones de *I. cyreni*, *I. monticola*, *I. bonnali*, *I. aranica*, *I. aurelioi* y *Podarcis muralis* a distintas condiciones de temperatura y usamos el número de extrusiones linguales de otros machos coespecíficos como medida de su eficacia. Simultáneamente examinamos su respuesta inmune, condición corporal y grado de parasitación para determinar si existía una relación con la altitud de la población. Los resultados mostraron diferencias en la efectividad de las señales según el tratamiento sólo en algunas especies y/o poblaciones, así como en el estado de salud poblacional. Esto sugiere diferencias entre especies en la vulnerabilidad ante un aumento de las temperaturas.

Cryptic consequences of global change: effects on health status and sensory ecology of the genus *Iberolacerta*

Although lizards, due to its thermophilia and active temperature control, seem to be relatively invulnerable to global warming, extinctions have been documented in five continents as a result of it, and it has been predicted the extinction of nearly 40% of their populations. Despite being heliotherm animals, remaining active in too warm conditions can raise their body temperature to critical highs forcing them to cool in shelters. An excessive use of refuges may reduce lizard's time for feeding, mate searching, territorial defense, etc., and finally affect their fitness.

Many effects of temperature rising are often not evident until they are examined. One aspect scarcely considered is the effect that increased temperatures may have on the sensory ecology of species; specifically on sexual communication. Among different types of sexual signals, chemical ones (pheromones), which are used by most of the species of lizards, are those most clearly temperature dependent. The signals of a given species or population are adapted to specific conditions of temperature and humidity, maximizing their transmission effectiveness. A change of these conditions, due to global warming, will cause the signals lose effectiveness, with consequences on the mechanisms of sexual selection.

Species of the genus *Iberolacerta* have discontinuous distributions, with isolated populations in mountains with different heights. This is an appropriate system to examine the adaptations (or lack thereof) and the effects of different environmental conditions of each population. Thus, our goal is to study whether the effect of temperature on the effectiveness of the signals differed between species and populations located at different altitudes within the range of each species. To do this we exposed femoral secretions of male *I. cyreni*, *I. monticola*, *I. bonnali*, *I. aranica*, *I. aurelioi* and *Podarcis muralis* from different populations to different temperature treatments. Then we used tongue-flick rate of other conspecific males as effectiveness measure of chemical signals. We also examined their immune response, body condition and degree of parasitism to determine whether there was a relationship with the altitude of the population. The results showed differences in the

effectiveness of the signals according to the treatment only in some species and/or populations, as well as in the population health status. This suggests there are differences among species in vulnerability to increased temperatures.

Chemical signaling, global warming, sexual selection, immune response, lizards.

Compromiso entre ornamentación sexual y sistema inmune en *Psammodromus algirus*

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Los modelos que explican la evolución de los ornamentos sexuales proponen que se trata de rasgos costosos correlacionados con la calidad de los machos, que pueden ser utilizados por las hembras como señales honestas de los beneficios genéticos que esos machos pueden ofrecer. Los carotenoides presentan interacciones simultáneas con la coloración epigámica y con la respuesta inmune, por lo que constituyen un sistema idóneo para estudiar el compromiso entre ambos. En este trabajo, se han analizado las respuestas de la coloración sexual a la activación inmune en machos de lagartija colilarga (*Psammodromus algirus*) de dos poblaciones, una de montaña y otra de llanura, con diferencias ambientales que podrían afectar al sistema inmune (ectoparasitismo sólo en la población de montaña). Los individuos se mantuvieron en cautividad y fueron sometidos a un experimento que consistió en activar el sistema inmune inyectando LPS a los sujetos experimentales (vs. suero a los sujetos control) para analizar los efectos de la activación en la coloración sexual rojizo-anaranjada. Se encontraron diferencias entre ambas poblaciones en la situación de partida en cuanto a la extensión (superficie coloreada/superficie total) y la saturación de la coloración sexual: la población de montaña presentó mayor saturación y la de llanura mayor superficie de color. Además, fueron precisamente esos dos aspectos los que se vieron comprometidos por la activación del sistema inmune de forma diferencial en ambas poblaciones: en la población de montaña fue la saturación la que se vio afectada por la activación del sistema inmune, mientras que en la de llanura lo fue la superficie. Estas diferencias podrían revelar interacciones entre la situación basal del carácter y el desarrollo del mismo en diferentes escenarios ecoinmunológicos.

Trade-off between sexual ornaments and immune system in *Psammodromus algirus*

The models which explain the evolution of sexual ornaments propose that these traits are correlated with male quality, and can therefore be interpreted by females as honest signals of genetic quality. Because of their simultaneous effects on epigamic coloration and immunity, carotenoids constitute an ideal system to study the trade-off between both. We have analyzed the responses of sexual coloration and immune activation in male *Psammodromus algirus* lizards from two populations, a montane area and a lowland one, in relation to the environmental differences that could affect their immune system (ectoparasitism only occurs in the montane lizard population). Individuals were kept in captivity and they were subjected to an experiment in which we activated their immune system by injecting them with bacterial lipopolysaccharides (vs. saline solution in control males) to analyze the effects of activation on the reddish-orange sexual coloration. We found between-population differences in the proportion of coloured surface area in the head (colour surface / total surface) and in the saturation the red coloration: montane lizards showed higher saturation, whereas lowland ones had larger pigmented surface areas. Moreover, these two aspects were differently compromised by the different activation of their immune systems in both populations: in the montane population, saturation was reduced by the activation of their immune system, while in the lowland population the amount of coloured area decreased. These differences could reveal interactions between the baseline sexual character and its development in differential ecoimmunological scenarios.

Sexual selection, sexual ornaments, trade-offs, intraspecific variation, immune system.

La radiación ultravioleta no aumenta el estrés oxidativo en la lagartija *Psammodromus algirus* a lo largo de un gradiente altitudinal

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Los lacértidos, por su condición de ectotermos, pasan bastante tiempo expuestos al sol para termorregular. Como consecuencia de ello, se encuentran expuestos a la radiación ultravioleta (UV) ($\lambda = 280-400$ nm) procedente del sol, que por otro lado, es uno de los componentes más dañinos de la luz solar. La radiación UV causa numerosos daños en el organismo, desde daños a nivel molecular hasta daños a nivel de tejidos, incluso puede causar la muerte en algunos organismos. La fotooxidación de las biomoléculas produce radicales libres, que pueden oxidar otras moléculas del organismo. Cuando la maquinaria antioxidante no es capaz de combatir la acción de estos radicales libres, en el organismo se produce una situación de estrés oxidativo. Dado que la radiación UV aumenta en intensidad conforme ascendemos en altitud, nuestra hipótesis predice que los lacértidos procedentes de altitudes más elevadas deberían estar mejor adaptados a estas condiciones de alta radiación UV, que los lacértidos de altitudes bajas.

En este trabajo, comprobamos esta hipótesis en la lagartija colilarga (*Psammodromus algirus*) a lo largo de un gradiente altitudinal (6 localidades, desde los 300 hasta los 2500 m sobre el nivel del mar). Durante un experimento de laboratorio, las lagartijas procedentes de cada localidad estuvieron expuestas 5 horas a condiciones de radiación UV (grupo experimental, bombilla de luz UV) o de luz fotosintéticamente activa (grupo control, bombilla de luz blanca). 24 horas después de la exposición, se cogieron muestras de tejido de la cola. Como indicadores del estrés oxidativo, medimos el daño oxidativo (hidro-peroxidación de lípidos y proteínas) y la capacidad antioxidante (a nivel enzimático y no enzimático). Como resultado, no encontramos diferencias en los niveles de estrés oxidativo entre los tratamientos; sin embargo, encontramos que el daño oxidativo fue menor en los individuos procedentes de mayores altitudes. Con esto, concluimos que la radiación ultravioleta no es un agente oxidante para *P. algirus*. Coincidiendo con un trabajo previo, nuestros resultados sugieren que el ambiente de altitudes bajas es más oxidante para las lagartijas. Por lo tanto, *P. algirus* está bien adaptada a ocupar un amplio rango de altitudes, lo que podría favorecer a la especie en el caso de la necesidad de tener que ascender en altitud, en un escenario de calentamiento climático.

Ultraviolet radiation does not increase oxidative stress in the lizard *Psammodromus algirus* along an elevational gradient

Lizards, as ectotherms, spend much time thermoregulating. Consequently, they are subjected to ultraviolet (UV) radiation ($\lambda = 280-400$ nm) from the sun, which is the most harmful component of solar radiation spectrum. UV-radiation can cause several damages from molecular to tissue level, or even the death in several organisms. Photooxidation provoked by UV-radiation produces reactive oxidative species (ROS). When antioxidant machinery cannot combat ROS concentration, oxidative stress occurs in the organisms. Given that UV-radiation increases with elevation, we hypothesized that lizards from high elevations should be better adapted against UV-radiation than lizards from lower elevations.

In this work, we test this hypothesis in Large *Psammodromus* (*Psammodromus algirus*) along an elevation gradient (six localities, from 300 to 2500 m above sea level). We ran an experimental process in which lizards from each locality were exposed to 5-hour doses of UV-radiation (UV-light bulb, experimental group) or photosynthetically active radiation (white-light bulb, control group), and 24 h after the exposition we took tissue samples from the tail. We measured oxidative damage (lipid and protein peroxidation) and antioxidant capacity (enzymatic and non-enzymatic) as oxidative stress biomarkers. We did not find differences in oxidative stress between treatments. However, we found that oxidative damage was smaller in lizards from highlands, and thereby we conclude that UV-radiation is not a stressor agent for *P. algirus*. Matching with a previous work, our findings suggest that lowland environment is more oxidative to lizards. Therefore, *P. algirus* is well adapted to inhabit a wide elevation range, and this probably will favour the lizard in case it ascends as consequence of climate warming.

Elevational gradient, global warming, lacertids, oxidative stress, *Psammodromus algirus*.

Consequences of corticosterone-mediated sexual conflict in common lizards (*Zootoca vivipara*)

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Frequently, males and females show different optimal fitness strategies concerning the system and frequency of mating, giving rise to sexual conflict. The outcome of this sexual conflict is determined by the balance between the costs and benefits of mating for each sex and proves essential in the evolution of reproductive strategies that can lead to speciation. Stress is a common response during reproductive events. It affects a diverse range of behavioural interactions, including mating, and oftentimes it negatively affects female fecundity and offspring quality. Asymmetric (sex-specific) effects of stress may influence the balance of costs and benefits of mating and thereby alter sexual conflict resolution.

Common lizards (*Zootoca vivipara*) exhibit a polygynandrous mating system in which males possess higher potential rate of reproduction than females, their fitness (i.e. number of offspring) largely depending on the number of mates they can fertilize. Females' potential reproductive rate is lower and they exhibit stronger mate discrimination. Their offspring do not increase proportionally with the number of mates and their lifetime fitness may be affected by current reproductive investment. In a mating experiment, we used corticosterone, one of the main glucocorticoid hormones involved in the stress response of lizards, to investigate female mating behaviour under high blood corticosterone levels and thereby assess the effects of increased asymmetric costs of mating in sexual conflict resolution. Since multiple paternity is common in the wild, we presented females with at least two different partners (either known or novel). This allowed us to further explore the hypothesis that high corticosterone level females might use multiple mating as a mechanism to compensate their higher costs of mating through increased genetic diversity, viability or sexual attractiveness of their offspring (i.e. potential benefits). Results revealed that high corticosterone level females were more aggressive towards conspecifics, especially males and known partners. They showed reduced probability of copulation compared to control females and this response was size-dependent. In contrast, males showed very similar behaviour towards females from different corticosterone group. Regardless of male increased interest towards known females, the probability of copulation between different groups of partner novelty was unchanged. Furthermore, copulations were longer on average with novel females, implying an increased chance of fertilization. Our results are consistent with the hypothesis that females try to reduce (rather than compensate) the elevated costs of mating through stronger mate discrimination, favouring long-term reproductive success over current attempts. This suggests an important, potential impact of environmental sources of stress during breeding in the evolution of reproductive strategies.

Sexual conflict, female choice, stress, multiple mating, mating behaviour, lizard.

¿Por qué es caro el sexo? Un estudio ecológico en un sistema multiparasitado

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Durante la época de celo, los machos de los vertebrados afrontan una inmunomodulación negativa debido al incremento de los niveles de testosterona en plasma. Estos cambios hormonales conducen a un aumento en la susceptibilidad a enfermedades de tipo parasitario. En el año 2013, se cuantificó la presencia de ectoparásitos, parásitos sanguíneos e intestinales de 75 individuos en una población de media montaña de *Lacerta schreiberi*. La prevalencia e intensidad de estos tres grupos de parásitos se puede explicar por la biología y tipo de transmisión de estos parásitos y la biología sexual y la ecología del comportamiento del hospedador. Los machos adultos que habitaban más frecuentemente arbustos higrófilos presentaron un mayor número de ectoparásitos. A pesar de que no hemos encontrado una correlación entre el número de garrapatas y la condición física de los individuos, aquellos lagartos a los que se les quitaron experimentalmente las garrapatas se movieron más que los del grupo control. Además los individuos con mayor número de garrapatas, estaban más frecuentemente infectados por helmintos intestinales. Estos individuos también tuvieron una respuesta inflamatoria de la piel menor. Por otra parte, los machos adultos de mejor condición se movieron distancias más largas y soportaron infecciones por los tres tipos de parásitos estudiados, mientras que los machos con peor condición física se movieron menos y soportaron una diversidad de parásitos menor. Por último, en relación con los parásitos sanguíneos sólo aquellos machos de mejor condición física estaban infectados. Este resultado sugiere que la transmisión de los parásitos sanguíneos se lleva a cabo durante los contactos intra e intersexuales. Por tanto, los datos de nuestro estudio concuerdan con la idea de que el sexo es costoso en esta población de lagarto verdinegro, al menos en términos de infección por parásitos.

Why is sex costly? An ecological study of a multiparasite system

During the mating season, male vertebrates may face a negative immunomodulation due to the increase of their plasma testosterone levels. These hormonal changes may lead to an increase in the susceptibility to parasitic diseases. During the mating season of 2013, 75 individuals of *Lacerta schreiberi* in a population breeding at middle elevation were screened for blood parasites, intestinal worms and ectoparasites. The prevalence and intensity of these three types of parasites in the individuals of the study population can be explained by the biology and type of transmission of these parasites and the sexual and behavioral ecology of the host lizard species. Adult male lizards living in humid bushes showed the highest levels of infection by ectoparasites. Even though there was no correlation between tick load and body condition, lizard where ticks were experimentally removed moved longer distances than those from the control group where ticks remain untouched. Moreover, the individuals with more ticks tended to have more intestinal worms. Furthermore, adult males infected by intestinal worms had a worse inflammatory skin reaction than those males without intestinal worms. Otherwise, adult male lizards with better body condition moved longer distances in the forest and supported the highest diversity of parasites, while males with worse body condition moved less and had less parasite diversity. In relation with the hematic parasites, only adult individuals with better body condition were infected, suggesting a transmission of the parasite during intra and intersexual interactions. Therefore, these results agree with the idea that sex is costly in this population of the Iberian Schreiber's Green lizard, at least in terms of parasite infection.

Lacerta schreiberi, parasite, nematode, tick, hemoparasite, behavioral ecology.

Función de las manchas UV y de la coloración ventral en combates entre machos en una población policromática de *Podarcis muralis*

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Las señales cumplen un importante papel regulando la intensidad de los enfrentamientos intraespecíficos. En este trabajo se examina el papel de dos posibles señales cromáticas en combates entre machos de *Podarcis muralis* procedentes de una población con un marcado polimorfismo en la coloración ventral; distinguiéndose principalmente tres morfos alternativos (blanco, amarillo y naranja). Además, y como muchos otros lacértidos, *P. muralis* presenta una serie de manchas azules para el ojo humano en las escamas ventrales externas (outer ventral scales, OVS). Estas manchas presentan su pico máximo de reflectancia alrededor de los 370 nm, dentro del espectro ultravioleta (UV), y estudios recientes sugieren que podrían aportar información sobre la calidad individual y la condición física de los machos. El objetivo de este trabajo fue examinar si alguna de las variables cromáticas de los machos constituye un predictor fiable de la habilidad para la lucha. Para ello, capturamos un total de 60 lagartijas adultas (20 individuos por morfo; SVL>65 mm). En el laboratorio, cada lagartija se enfrentó a un competidor de cada uno de los morfos en un torneo equilibrado entre morfos, dando lugar a un total de 99 combates. Los resultados de esos combates se analizaron con un modelo de Bradley-Terry. La condición residente/intruso de ambos contendientes se igualó en todos los combates para evitar su demostrada influencia como determinante del resultado del combate. Discutimos los resultados obtenidos para la relación entre los morfos implicados, las medidas cromáticas de las manchas UV de las OVS y la calidad individual para el combate.

Role of alternative ventral coloration and UV ornaments in staged combats between males of a polychromatic population of common wall lizard (*Podarcis muralis*)

Signalling plays a key role regulating the intensity of animal contests. In this study, we evaluate the role of two potential chromatic signals in determining the outcome of male-male combats in *Podarcis muralis* lizards from a population showing polymorphic ventral coloration, with discrete white, yellow and orange morphs. As in other lacertids, males of this species display a row of ventrolateral UV/blue scales (outer ventral scales, OVS) with a reflectance peak around 370 nm, within the UV wave band. Previous studies suggest that chromatic variables of these patches may signal individual fighting ability and/or physical condition. In order to test this possibility, we staged combats between 60 adult lizards (20 individuals/morph; SVL>65 mm), where each lizard confronted rivals from each morph in a tournament with a balanced design. Intruder/resident condition was controlled in order to avoid its strong effect in determining contest outcome. A general dominance/fighting ability ranking was calculated using the Bradley-Terry model, and used to explore the general relationship between morphs, chromatic measurements of the UV patches, and fighting ability.

Communication, contest, polychromatism, visual signal, UV, lizard.

Respuesta inmune y comportamiento antidepredador de renacuajos de sapo partero en un gradiente de urbanización

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Grandes áreas de hábitat natural se han convertido en zonas urbanas en los últimos años. Estos cambios ambientales inducidos por el hombre suponen un desafío evolutivo: muchas especies nativas no pueden adaptarse al estrés urbano mientras otras colonizan y “explotan” con éxito este nuevo entorno. El éxito de éstas últimas puede deberse a características previas que les pre-adaptan a las nuevas condiciones del ambiente urbano (p. ej. altos niveles de contaminación y baja presión de depredación), o ser el resultado de un proceso reciente de adaptación fenotípica.

Los renacuajos de especies como el sapo partero común (*Alytes obstetricans*) se encuentran en zonas con diferentes niveles de urbanización y a menudo en parques urbanos. El largo período de tiempo que los renacuajos invernantes de esta especie pasan en hábitats acuáticos, expuestos a la contaminación del agua y a los depredadores acuáticos, los hace especialmente adecuados para estudiar si el nivel de urbanización ha afectado a alguna adaptación fenotípica.

Los cambios en el comportamiento son a menudo la respuesta inicial a las alteraciones ambientales provocadas por el hombre. El comportamiento antidepredatorio es crucial para la supervivencia, pero costoso, así que debe estar bien equilibrado en un contexto de asignación de recursos. La ausencia o escasez de depredadores en los hábitats urbanizados pueden alterar este equilibrio y promover cambios en la reacción de los renacuajos a las amenazas depredadoras. Por otro lado, el estrés, consecuencia de la presión de depredación o la contaminación urbana, menoscaba la condición o estado de salud.

Nuestro objetivo fue examinar si existen diferencias en la respuesta antidepredadora, en el estado de salud y en la relación entre ambas en función del grado de urbanización. Así, se capturaron renacuajos hibernantes en once estanques a lo largo de un gradiente de urbanización y se midieron rasgos morfológicos (longitud y altura relativa de la cola), condición física (residuos del peso sobre la longitud) y estado de salud mediante la prueba de la fitohemaglutinina, un indicador de la respuesta inmune celular. El comportamiento antidepredador se midió como la respuesta de los individuos a señales de alarma de conespecíficos. Se observaron diferencias entre poblaciones en casi todas las variables medidas, pero el grado de urbanización sólo explicaba las diferencias en la respuesta inmune (menor en las poblaciones más urbanizadas) y en la respuesta a las señales de alarma (menor en las poblaciones con un grado intermedio de urbanización), aunque la relación entre ambas respuestas no parece verse influida por éste.

Immune response and antipredatory behaviour of common midwife toad tadpoles in a gradient of urbanization

Large areas of natural habitats have been converted to urban areas in the last years. These environmental human-induced changes pose an evolutionary challenge: many native species may not adapt to urban stress while other species are able to successfully colonize and “exploit” this new environment. Success of urban species may be due to previous suitable characteristics that pre-adapted them to the new conditions (e.g. high levels of pollution and low predation pressure) or be the result of a recent phenotypic adaptation process.

Tadpoles of some species, such as the common midwife toad (*Alytes obstetricans*), can be found in areas with different levels of urbanization and often in urban parks. The long period of time that overwintering tadpoles spend in aquatic habitats, exposed to water pollution and aquatic predators, make them appropriate specimens for studying if urbanization level is related to some phenotypic adaptation.

Behavioural changes are usually the initial response to human-induced environmental alterations. Antipredatory behaviour is crucial for survival but costly, so it must be well balanced in a context of resource allocation. Absence or scarcity of predators in urbanized habitats may alter this balance and promote changes in tadpoles’ reaction to predatory threats. On the other hand, stress, consequence of predation pressure or urban pollution, may impair immune response.

Our goal was to examine if tadpoles present differences in health status, antipredatory response, and in the relation between both, related to urbanization. Thus, we collected hibernating tadpoles in eleven ponds along a gradient of urbanization and measured morphological traits (relative length and height of the tail), body condition (residuals of the regression of body mass against length) and health status by the phytohemagglutinin test, an indicator of the cellular immune response. Then, we studied the behaviour of individuals in the presence of conspecific alarm cues as a measure of antipredatory response. We found differences among populations in almost all measured variables, but urbanization only was related to the

immune response (lower in more urban populations) and antipredatory response (lower in intermediate populations). However, the relationship between antipredatory and immune response seems not to be influenced by urbanization.

Behaviour, urban habitats, immune response, predation, tadpoles.

Contributo para o estudo da capacidade de quimiorrecepção dos juvenis de tartaruga marinha comum (*Caretta caretta*, L.)

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Os juvenis da tartaruga marinha comum (*Caretta caretta*) são encontrados nas águas do arquipélago da Madeira. As tartarugas pelágicas encontram-se na principal fase de crescimento do seu ciclo de vida e, conseqüentemente, de maiores necessidades energéticas. No entanto, os conhecimentos sobre a ecologia das tartarugas marinhas comuns pelágicas ainda são rudimentares, nomeadamente sobre o modo como encontram o alimento numa área tão vasta como o oceano. Estudos efetuados com outras espécies pelágicas, nomeadamente com aves procelariiformes, revelaram que o sistema olfativo desempenha um papel importante na deteção de áreas alimentares através da deteção de picos de DMS (dimetilsulfido), composto aromático que existe naturalmente no ambiente marinho e que está relacionado com áreas de elevada produtividade. Partindo do pressuposto que as tartarugas marinhas comuns utilizam semelhante mecanismo, foram efetuadas experiências comportamentais de modo a avaliar a capacidade de quimiorrecepção do DMS (aérea e aquática). O objetivo era averiguar se as tartarugas marinhas comuns possuem sensibilidade ao DMS e se realmente utilizam este composto, em condições naturais, para a deteção de áreas alimentares. Foram testadas 4 tartarugas em cativeiro e três tartarugas selvagens em mar aberto. Os resultados das experiências de quimiorrecepção aérea, em cativeiro, revelaram que uma das tartarugas possuía claramente sensibilidade ao DMS e as experiências de mar confirmaram este resultado. Contudo, as experiências não foram conclusivas relativamente à questão se as tartarugas pelágicas usam efetivamente o DMS como pista aérea olfativa na deteção a longa distância das áreas alimentares. Nas experiências de quimiorrecepção aquática não foi observada sensibilidade ao DMS pelas três tartarugas testadas. As experiências de condicionamento clássico, em que o DMS e o alimento eram apresentados aproximadamente ao mesmo tempo, revelaram que, após um certo período de tempo, a tartaruga não associava o estímulo de DMS com uma possível recompensa alimentar. A principal causa de morte das tartarugas marinhas no arquipélago da Madeira é o bycatch pela pesca dirigida ao peixe-espada preto (*Aphanopus carbo*). Um dos iscos utilizados é a cavala (*Scomber japonicus*). Foram efetuadas experiências de quimiorrecepção aquática de modo a avaliar a atratividade que a cavala poderia ter para as tartarugas marinhas. Os resultados revelaram que, em 90% dos casos, eram atraídas pelo odor da cavala.

A contribution to the study of the chemoreception capacity of juvenile loggerhead sea turtles (*Caretta caretta*, L.)

Loggerhead sea turtle juveniles (*Caretta caretta*) are found in Madeira archipelago waters. Pelagic turtles are in the main growth phase of their life cycle and consequently have higher energy needs. However, knowledge about the ecology of pelagic loggerhead sea turtles is still quite rudimentary, mainly about the mechanisms that lead them to find food in the vast ocean. Studies with other pelagic species, such as procellariiform birds, revealed that the olfactory system plays an important role for the detection of feeding areas, through the detection of concentration peaks of DMS (dimethyl sulfide), a scent compound that naturally exists in the marine environment and is related to areas of high productivity. Based on the assumption that loggerhead sea turtles use a similar mechanism, behavioral experiments were conducted in order to analyze the chemoreception capacity for DMS (airborne and aquatic chemoreception). The first step was to observe if pelagic loggerheads demonstrate sensitivity to DMS and the second was to verify if they actually use the DMS, in natural conditions, as an airborne cue to find areas where food patches might be available. Four juveniles of loggerhead sea turtles were tested in captivity and three wild turtles in the open ocean. The results of airborne chemoreception experiments in captivity revealed that one turtle clearly demonstrated sensitivity to DMS and the sea experiments confirmed this result. However, the experiments were not conclusive about whether pelagic turtles actually use the DMS as an airborne cue to detect long distance food patches. In aquatic chemoreception experiments we did not observe sensitivity to DMS by the three sea turtles tested. In the classical conditioning experiment, where DMS and food were given nearly at the same time we found that, after a certain period of time, the sea turtle tested did not associate the DMS stimulus with a possible food reward. The main cause of mortality of loggerhead sea turtles in Madeira waters is bycatch by black-scabbardfish fishery (*Aphanopus carbo*). Chub mackerel (*Scomber japonicus*) is one of the baits used in this fishery. Aquatic chemoreception experiments were conducted in order to evaluate the attractiveness of this fish for sea turtles. For the three sea turtles tested, the results showed that, in 90% of the cases, they were extremely attracted by the underwater smell of chub mackerel.

Loggerhead sea turtle, chemoreception, dimethyl sulfide, feeding ecology, bycatch, Madeira.

Características demográficas de una población pedomórfica de tritón pirenaico (*Calotriton asper*) en un lago de elevada altitud

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En el estudio que se presenta se han analizado los caracteres demográficos del ciclo vital de una población pedomórfica de tritón pirenaico (*Calotriton asper*) que habita en el Ibón de Perramó, a 2270 m de altitud en la comarca de la Ribagorza (provincia de Huesca). La población está constituida a la vez por individuos metamórficos y pedomórficos. Se ha estimado la estructura de edad mediante técnicas esqueletocronológicas y el tamaño corporal de la población. Se han descrito las características pedomórficas de los individuos y si existe relación con el sexo (machos y hembras) o grupo poblacional (inmaduros y larvas). Los resultados muestran diferencias en la distribución de edades entre machos y hembras, hembras e inmaduros y entre machos y larvas. En cambio no hay diferencias en la distribución de edades dentro de cada sexo ni en los inmaduros cuando se comparan los individuos metamórficos y los pedomórficos. Se observan diferencias significativas en la longitud hocico-cloaca (SVL) en los individuos adultos, que muestran un dimorfismo sexual claramente marcado. Los individuos metamórficos y los pedomórficos agrupados por sexo y grupo poblacional no muestran diferencias significativas, siendo la SVL similar en los inmaduros y en los machos, y entre las hembras metamórficas y machos tanto los metamórficos como los pedomórficos. La longitud hocico-cloaca está positivamente correlacionada con la edad en las hembras e inmaduros pero no en los machos y larvas. En cambio cuando se analizan los individuos metamórficos y pedomórficos la SVL está significativamente correlacionada con la edad en las hembras pedomórficas y en los inmaduros metamórficos. Estos resultados describen una población de *C. asper* caracterizada por presentar caracteres vitales atípicos si se comparan con los conocidos hasta el momento para la especie.

Demographic traits of a paedomorphic population of Pyrenean newt (*Calotriton asper*) living in a high altitude lake

Demographic life history traits were studied in a paedomorphic population of Pyrenean newt (*Calotriton asper*) inhabiting lake Perramó, at 2270m a.s.l. in the region of Ribagorza (province of Huesca). The population includes metamorphic and paedomorphic individuals simultaneously. We estimated age structure using skeletochronology and body size data. We describe paedomorphic characteristics of individuals and investigate whether there is a relationship with sex (males and females) or population group (immature and larvae). The results show differences in the age distribution between males and females, females and immatures and between males and larvae. However no differences were found in the age distribution within each sex or immatures when paedomorphic and metamorphic individuals are compared. Significant differences in snout-vent lengths (SVL) of adult individuals were observed, showing a clearly marked sexual dimorphism. The metamorphic and paedomorphic individuals grouped by sex and population group show no significant differences, the SVL being similar in immatures and in males, and also between metamorphic females and males both for metamorphic and paedomorphic individuals. Snout-vent length is positively correlated with age in females and immatures but not in males and larvae. Instead, when metamorphic and paedomorphic individuals are analyzed, the SVL is significantly correlated with age in paedomorphic females and metamorphic immatures. These results describe a population of *C. asper* characterized by atypical life history traits when compared with those previously known in the species.

Calotriton asper, life history, paedomorphosis, skeletochronology, age structure, body size.

Estrategias tróficas del anfibio invasor *Discoglossus pictus*: ¿competencia u oportunismo trófico?

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Uno de los factores determinantes en el éxito de proliferación de especies invasoras y su efecto sobre las especies nativas es conocer cómo aprovechan los recursos tróficos disponibles en el medio que invaden. En general, las especies invasoras pueden seguir dos estrategias en relación al uso de los recursos: una estrategia competitiva, en la que la especie invasora explota los mismos recursos utilizados por las especies nativas, o una estrategia oportunista, en la que la especie invasora explota recursos infrautilizados por las especies nativas. En Europa, el sapillo pintado (*Discoglossus pictus*) es una de las pocas especies introducidas de anfibios que se encuentra actualmente en expansión. Esta especie presenta un elevado solapamiento en el hábitat de reproducción con una especie nativa, el sapo corredor (*Bufo calamita*). El objetivo del presente estudio fue determinar cómo estas dos especies (invasora y nativa) gestionan los recursos cuando coinciden en el mismo hábitat acuático y qué efectos sobre el nicho trófico tendría su co-ocurrencia para ambas. Basándonos en los resultados obtenidos en laboratorio, utilizamos el análisis de isótopos estables de nitrógeno y carbono en individuos recolectados en campo para comprobar y completar dichos resultados. Los resultados de laboratorio indicaban una mejor explotación de recursos por parte de la especie introducida y sugerían la existencia de una estrategia competitiva que podría conllevar al desplazamiento de la especie nativa. Estos resultados fueron corroborados en condiciones naturales a partir de los resultados isotópicos. A pesar de encontrar similitud en el nicho trófico entre ambas especies en hábitats donde no coexistían, encontramos que los hábitats donde ambas especies co-existían registraba una clara segregación en el nicho trófico entre ambas especies.

Trophic strategies of the invasive amphibian *Discoglossus pictus*: trophic competition or opportunism?

One of the critical factors explaining the success of invasive species and their effect on native species is to know how they use trophic resources. Two main trophic strategies have been proposed to explain resource acquisition by invasive species: invasive species can behave aggressively over the resources exploited by natives, displacing them from their niches (competition hypothesis), or they may exploit niche opportunities that most native species are unable to use efficiently (opportunism hypothesis). In Europe, painted frogs (*Discoglossus pictus*) are one of the few introduced amphibian species that are currently expanding. This species shows high overlap in the breeding habitat with one native species, the natterjack toad *Bufo calamita*. The aim of the present study was to determine how the two species use trophic resources when they coexist in the same habitat and the potential effects of the invasive on the native species. To corroborate previous findings obtained under laboratory conditions, we collected stable isotope data (nitrogen and carbon) in specimens of both species collected in field. The results of the experiment indicated a high rate of exploitation of trophic resources by the introduced species and suggested that *D. pictus* follows a competitive strategy which may involve the displacement of the native species to lower quality resources. These results were corroborated with the isotopic data. Despite the trophic similarities registered between both species in non-shared ponds, stable isotopes analyses allowed us to detect niche partitioning in shared ponds.

Invasive species, stable isotopes analyses, amphibians, opportunism, competition, trophic niche.

What influences the colonization of artificial arboreal refuges by *Hyla arborea*?

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With our study, we aim to understand how *Hyla arborea* uses trees and arboreal refuges near reproductive areas throughout the year, and what environmental variables influence refuge colonization by this species. We specifically aim to answer the following questions: (1) Are the refuges placed higher in trees more frequently colonized by *H. arborea*? (2) Are refuges installed on isolated trees more or less colonized than those on tree patches? (3) Does the temperature inside the refugia affect the colonization rate? (4) What environmental variables influence the refuge choice by *H. arborea*? We installed 70 PVC refuges on isolated trees and tree patches, at two different heights (breast and above-head), in an area dominated by rice fields and reed beds, in Baixo Vouga Lagunar (municipality of Estarreja, North-Western Portugal). Colonization of refuges by *H. arborea* was monitored from February 25th 2013 to April 24th 2014, fortnightly, for periods of four consecutive days. In each sampling period, we measured the top and bottom temperatures of each colonized refuge and of 20 randomly chosen non-colonized refuges. We also measured the temperature of the ventral surface and the snout-vent length of the individuals colonizing the refuges, as well as the temperature on its position in the tube. We registered the values of 20 variables related with microhabitat, time, season, meteorological conditions and handling. We recorded 2277 occupancy events on the 112 days of sampling, including 524 multiple colonization events, with a maximum of nine individuals in one tube. Our results reveal that refuges installed in tree patches were more frequently colonized, these differences being significant in Summer and Winter. Colonization was not significantly different in refuges installed higher or lower in the trees throughout the year. We found no differences on average temperatures between colonized and non-colonized refuges throughout the sampling period. Significant differences on colonization rates were also found among seasons, namely among Spring and all other seasons. The lowest colonization rate occurred in Spring, which can be related to the reproductive season. We also discuss the influence of the other measured variables on the colonization rate of artificial refuges. This study will help to fill the gaps on the knowledge of this species ecology. We encourage the use of this sampling method in future studies on the ecology of arboreal amphibians.

Hyla arborea, temperature, season, handling, microhabitat.

Habitat related differences in morphology, locomotion and bite force performance in the generalistic lizard *Podarcis bocagei*

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If morphological variation is tightly related with habitat use, then differences in morphology should affect fitness through their effect on performance within specific habitats. In this study, we investigated intraspecific ecomorphological variation in the generalistic lacertid *Podarcis bocagei* by examining two habitat types: agricultural wall vs. dunes where we expected to reflect saxicolous vs. ground-dwelling habits. We collected lizards from four populations of *P. bocagei*: two from wall habitats and two from dune habitats. In the laboratory, we recorded morphological traits, potentially relevant for locomotion and bite performance. We also quantified locomotor performance by measuring sprint speed, climbing capacity and maneuverability. Finally, we measured bite force for each animal. Based on these measurements, we used univariate and multivariate statistics to examine whether lizards inhabiting the two habitat types differed in morphology and whole-animal performance, while taking variation across populations into account. Our results indicated that lizards from walls and dunes differed in total body size, as well as relative head high, trunk length and hind limb length. Confirming previous observations we also observed highly significant differences between sexes in almost all morphological traits examined. Contrary to ecomorphological predictions, habitat-related differences in morphology were not reflected in locomotor performance, where we only found differences in sprint speed across populations, which were not related to habitat variation. Finally, bite force was significantly different both between habitats and sexes. Our results suggest that the differences between lizards of the two habitats derive from natural selection constraints involving refuge use. Further studies on habitat use preferences and behaviour should support this conclusion.

Habitat use, locomotion, morphometrics, bite force, ecomorphology, Lacertidae.

Respuesta compensatoria en la reproducción de la tortuga *Testudo graeca* frente a las perturbaciones del hábitat

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Los testudínidos son unas de las familias más amenazadas del planeta. El estado de riesgo o amenaza de las tortugas está estrechamente ligada a variaciones en sus rasgos de vida. Por eso, el estudio de los patrones reproductivos y las alteraciones que pueden sufrir son necesarios para el desarrollo de planes de conservación que resulten exitosos. Una de las principales causas que provoca el declive de las poblaciones de tortugas es la fragmentación y pérdida de calidad del hábitat. El objetivo de este trabajo es describir la reproducción de *Testudo graeca* en el sureste de España y analizar si las características del hábitat están influenciando en los patrones reproductivos de las hembras. En 6 años del periodo 2005 a 2014 realizamos 534 radiografías a hembras a lo largo de la época reproductiva (abril hasta junio) en 14 poblaciones con diferente estructura y calidad de hábitat, a lo largo del rango de distribución de la especie en el sureste ibérico (2600 km²).

En conjunto, el porcentaje de hembras grávidas fue de un 47% respecto al total de hembras radiografiadas, con un promedio de dos puestas durante la primavera, y con 3.16 huevos por puesta (entre 1 y 7 huevos). Los parámetros reproductivos dependieron de características del individuo, de variaciones climáticas interanuales y de variables de hábitat. Así, el tamaño de la hembra (longitud del espaldar) está directamente relacionado con el número de huevos. El número de hembras grávidas está afectado por las variaciones interanuales, concretamente en el año 2014, que fue extraordinariamente seco, el porcentaje de hembras grávidas alcanzó el 24.9% mientras que en el resto de los años la media fue de 75.2%. Respecto a los efectos del hábitat en la reproducción, paradójicamente encontramos que las poblaciones con una menor superficie de hábitat de calidad mostraron una tendencia a tener un número mayor de hembras grávidas y mayor número de huevos por puesta. A raíz de estos resultados, hipotetizamos que la tortuga mora en el sureste ibérico muestra una respuesta de compensación frente a alteraciones en la calidad del hábitat.

A compensatory response against habitat disturbances in the reproduction of the tortoise *Testudo graeca*

Testudinidae is one of the most world-wide endangered families. The threatened or endangered status of many tortoise species may be linked to variations in their life histories. The study of reproduction rates and their variation is necessary for the development of successful conservations programs. One of the most common causes of population declines are habitat loss and fragmentation. The aims of the present study were to describe the reproduction rates of *Testudo graeca* in the southeast of Spain and to analyze the effects of habitat structure on female reproductive patterns. Fieldwork was conducted along 6 years, between 2005 and 2014. We sampled 534 wild females using X-ray throughout the reproductive period (from April to June) in 14 wild populations along the distribution area in the Iberian southeast (2600 km²).

The overall percentage of gravid females was 47%, performing in average two clutches during spring of 3.16 eggs per clutch (between 1 and 7 eggs). Reproductive parameters were related to individual characteristics, interannual climate variations and habitat variables. Concretely, female size (carapace length) is positively related to the number of eggs. The number of gravid females in 2014, which was unusually dry, reached 24.9% while the rest of the years the average was 75.2%. Regarding the effects of habitat on reproduction, paradoxically we found that populations with a lower habitat quality tend to have higher rates of gravid females and a higher number of eggs per clutch. Following these results, we hypothesized that the spur-thighed tortoise in the Iberian southeast shows a compensatory response against habitat disturbances.

Reproduction, tortoise, hábitat, *Testudo graeca*, disturbance.

Revelando as regras para a co-existência de espécies (répteis) em zonas de transição entre ecoregiões

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As potenciais interações dos membros do mesmo grupo ecológico são muitas vezes realizadas através de segregação espacial ou de diferenças nas suas dietas. Estes padrões de segregação têm sido observados entre várias espécies o que indica que poderá haver evitamento de tais interações, contudo a total segregação é raramente vista em Squamata. Nós examinámos os padrões de co-existência da comunidade de répteis existente no vale do Rio Homem (Parque Nacional da Peneda-Gerês, norte de Portugal). O local de estudo encontra-se numa área em que se verifica a transição entre as ecoregiões Euro-siberiana e Mediterrânica e que, por isso, apresenta uma comunidade mista de répteis composta por espécies com diferentes afinidades biogeográficas, nomeadamente Atlânticas e Mediterrânicas. Os nossos objetivos são: 1) perceber se há uma segregação de fina escala a nível do habitat com paralelismo entre as respetivas afinidades biogeográficas (i.e. as suas distribuições na Península Ibérica); e 2) perceber se as espécies apresentam algum potencial de interações devido à similaridade de preferências dietárias e como as diferenças dietárias e a segregação espacial estão relacionadas. Em 2012-2014, conduzimos visitas a um transeto no vale do Rio Homem cujas observações de espécimes foram geo-referenciadas com um dispositivo GPS. A seleção do habitat pelos répteis no local de estudo foi obtida pela frequência em que cada espécie foi observada numa das quatro categorias de habitats por nós proposta (desde habitat totalmente aberto até habitat totalmente fechado) dependendo dos habitats disponíveis. Foram construídas Matrizes de Similaridade de espécies relativas ao habitat, dieta e distribuição Ibérica com comparações par-a-par, e posteriormente foi calculada a correlação entre as três matrizes através de testes parciais de Mantel, primeiro para toda a comunidade e depois separando lagartos e serpentes. No total, analisámos 136 pontos GPS de 9 espécies. As espécies Mistas e Mediterrânicas selecionaram categorias de habitat abertas, contrariamente, duas das quatro espécies Atlânticas evitaram totalmente áreas abertas, selecionando apenas habitat fechados. As restantes duas espécies Atlânticas mostraram diferenças sazonais na seleção do habitat. Os nossos resultados sugerem que todas as espécies de répteis presentes no vale do Rio Homem selecionam o seu habitat de acordo com os seus padrões geográficos de distribuição mais amplos (i.e. Ibéricos). Não encontramos nenhuma ligação entre as preferências pelo habitat e das dietas, significando que a competição indireta não é um factor limitante. Concluímos então que a co-existência de espécies Mediterrânicas e Atlânticas é possível através da seleção diferencial de micro-habitats independentemente das semelhanças dietárias.

Uncovering the rules of (reptile) species coexistence in transition zones between eco-regions

Potential interactions of member of the same ecological guild are often released through spatial segregation or differences in their diet. Spatial segregation patterns have been observed between several species pairs indicating potential avoidance of such interactions, but full segregation patterns are rare in squamates. We examined the patterns of co-existence of the reptile community present in the Rio Homem valley (Peneda-Gerês National Park, northern Portugal). The study site lays in the area that is a transition zone between the Euro-Siberian and Mediterranean Ecoregions and has a mixed reptile community composed by species with different biogeographic affinities namely Atlantic and Mediterranean. Our goals were to examine (1) if species segregate on a fine-scale habitat level paralleling their biogeographic affinities (i.e. their distribution pattern across Iberia), (2) if species have a potential to interact due to similar food preferences (diet similarity analysis), and how are diet differences and spatial segregation related. In 2012-2014, we conducted a number of visits along a single transect at Rio Homem valley, and reptile sights were georeferenced with a GPS device. Habitat selection of reptiles in the study site was characterized by the frequency in which it had been observed within four possible Habitat Categories (from fully open to fully covered) depending on the available habitats. Species Similarity Matrices of habitat, diet and Iberian distribution were built with all pairwise comparisons and the correlation among the three matrices examined using Partial Mantel tests for the whole community and then for lizards and snakes separately. We recorded 136 GPS points from 9 species. Mediterranean and Mixed species selected open-habitat categories whereas two out of four Atlantic ones extremely avoided open habitats. The remaining Atlantic species showed differences in seasonal habitat selection. Our results showed that essentially all reptile species' selects its optimum habitat according to their broader (Iberian) geographic distributional pattern. We found no link between habitat and dietary preferences, thus indirect competition was not a limiting factor, and therefore the co-existence of Mediterranean and Atlantic species is possible through a differential microhabitat selection regardless the similarity in diet.

Reptiles, habitat selection, spatial segregation, transition zone, Iberian Peninsula.

Competitive abilities of the native *Bufo calamita* and the invasive *Discoglossus pictus* near the origin of the introduction: how is their coexistence possible?

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After a century of coexistence between the Natterjack toad (*Bufo calamita*) and the introduced Mediterranean Painted Frog (*Discoglossus pictus*), neither population or abundance declines of the native *B. calamita* have been attributed to the presence of *D. pictus* despite several studies highlighting a great niche overlap. Here we gather data on two laboratory experiments trying to shed light on this topic. On one hand, we assessed the competition abilities (survival, behavior, growth, etc.) of both species together under different conditions of density and pond desiccation, using specimens from near the introduction origin area. *B. calamita* slightly diminished its survival when coexisting with *D. pictus*, but although the mean size of tadpoles was affected by stress factors like density and diminishing water levels, it was not affected by the presence of *D. pictus* itself. Both species were highly plastic morphologically, but *D. pictus* was much more plastic behaviorally. Linking this data with physiological abilities of both species (second experiment), we can argue that these species use two very different strategies: *Bufo calamita* is a species basing its growth on physiological efficiency and not on high consumption rates, and therefore has a poor ability to modulate its behavior in order to increase its feeding rates or quality. However this tactic might enable this species to cope with extremely high densities of tadpoles, or the introduction of a competitor which growth is extremely based on a high consumption, like *D. pictus*. On the other hand, as a consumer species (basing its growth on a high consumption) *D. pictus* is more negatively affected by any factor hindering its high intake. These “complementary” functioning might be a first explanation for the coexistence of both competitors without a long coevolutionary history.

Invasive species, tadpole community, physiological tactics, growth physiology.

***Psammodromus algirus* como reservatório de *Borrelia lusitaniae*, agente etiológico da borreliose de Lyme, em Portugal**

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As lagartixas são frequentemente infestadas por carraças e algumas espécies estão descritas como reservatórios de *Borrelia burgdorferi* s.l., o agente etiológico da borreliose de Lyme, especificamente de *B. lusitaniae*, a genoespécie de *Borrelia* mais prevalente em Portugal. Neste estudo avaliámos a importância de lagartixas como hospedeiros de *Ixodes ricinus*, o principal vector de borreliose de Lyme na Europa, e o seu papel como reservatório de *B. lusitaniae* num foco enzoótico. Para isso avaliámos a prevalência de infecção de *B. burgdorferi* s.l. em carraças recolhidas de lagartixas, bem como nos seus tecidos. A infecção por *B. burgdorferi* s.l. foi pesquisada através de um PCR nested que amplifica a região intergénica 5S-23S desta bactéria, e a identificação da genoespécie foi feita através da análise por sequenciação dos amplicões resultantes. *Borrelia lusitaniae* foi a genoespécie identificada como mais frequente nas carraças recolhidas da vegetação e foi detectada em carraças que se alimentavam em *Psammodromus algirus*, *Podarcis hispanica* e *Lacerta schreiberi*. Larvas de *I. ricinus* infectadas com *B. lusitaniae* foram encontradas apenas em *P. algirus*, e detectou-se também uma biopsia positiva recolhida da cauda de uma *P. hispanica*, sugerindo infecção sistémica. Estes resultados reforçam a importância das lagartixas como reservatório de *B. lusitaniae*, como já reportado anteriormente, sugerindo que *P. algirus*, especificamente, actua como um dos principais reservatórios em Portugal.

***Psammodromus algirus* as a reservoir for *Borrelia lusitaniae*, an etiologic agent of Lyme borreliosis, in Portugal**

Lizards are often infested by ticks and some species are considered reservoirs for *Borrelia burgdorferi* s.l., the etiologic agent of Lyme borreliosis, specifically *B. lusitaniae*, the most common *Borrelia* genospecies in Portugal. We assessed the importance of lizards as hosts for *I. ricinus*, the main vector of Lyme borreliosis in Europe, and as reservoirs for *B. lusitaniae* in an enzootic focus, by assessing infection prevalence in their ticks and tissues. *B. burgdorferi* s.l. infection was detected through a nested PCR targeting the 5S-23S rDNA intergenic spacer region and the genospecies was identified by sequence analysis of the resulting amplicons. *Borrelia lusitaniae* was the most prevalent genospecies in questing *I. ricinus* and was found in ticks feeding on *Psammodromus algirus*, *Podarcis hispanica* and *Lacerta schreiberi*. *P. algirus* was the only lizard species harbouring *I. ricinus* larvae infected with *B. lusitaniae*. One positive biopsy was retrieved from the tail of one *P. hispanica* which suggests a systemic infection. Our results reinforce the importance of lizards as reservoirs for *B. lusitaniae*, as previously reported, suggesting that *P. algirus*, in particular, acts as a main reservoir for *B. lusitaniae* in Portugal.

Psammodromus algirus, *Podarcis hispanica*, Lyme borreliosis, tick, *Borrelia lusitaniae*, *Ixodes ricinus*.

Prevalência de parasitas sanguíneos *Hepatozoon* em populações de *Podarcis*

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O filo Apicomplexa é um vasto e diverso grupo consistido por protistas unicelulares, maioritariamente parasitas obrigatórios, e é um dos grupos de animais menos estudados em termos de biodiversidade. O género *Hepatozoon* é o mais abundante em répteis e mamíferos, mas também um dos menos estudados do filo em geral. Como tal, é importante melhorar o nosso conhecimento sobre estes organismos. Compreender também como a distribuição de populações de parasitas é influenciada, por exemplo, por variáveis ambientais é crucial para responder variadas questões, tal como prever e antecipar futuras mudanças na distribuição destas populações. Esta informação é particularmente relevante considerando as mudanças climáticas globais que observamos nos dias de hoje. Os níveis de parasitemia foram avaliados em várias populações de *Podarcis* da Península Ibérica e vários factores, de variação ambiental a atributos populacionais, foram testados, de forma a compreender a sua influência na distribuição dos parasitas sanguíneos *Hepatozoon*. A Prevalência variou consideravelmente entre populações da mesma espécie, assim como entre diferentes espécies de *Podarcis*, mesmo ocorrendo em simpatria. Possíveis razões para esta variação serão discutidas.

Prevalence of *Hepatozoon* blood parasite in *Podarcis* populations

The Apicomplexa phylum is a large and diverse group of unicellular protists, most of which are obligate parasitic organisms, and one of the poorest-studied groups of animals in terms of biodiversity. *Hepatozoon* is the most abundant genus in reptiles and mammals but also one of the less studied of the phylum. As such, increasing our limited knowledge about such organisms is of considerable importance. Also, understanding how the distribution of parasite populations is affected by, for example, environmental variables is crucial to answering various questions, one of which being how to anticipate and predict future changes in the distribution of such populations. This is especially relevant in light of the current global climatic changes. Parasitaemia levels in multiple *Podarcis* populations of the Iberian Peninsula were assessed and several factors, from environmental variation to population attributes, were tested, in order to understand their influence on the distribution of *Hepatozoon* blood parasites. Prevalence varied considerably between populations of the same species and between different *Podarcis* species even when they occurred in sympatry. Possible reasons for this variation will be discussed.

Hepatozoon, parasite, *Podarcis*, prevalence.

Variação de níveis de parasitemia de parasitas sanguíneos em répteis endêmicos das Ilhas Canárias

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Membros do género *Hepatozoon* são os parasitas intracelulares mais comuns e amplamente distribuídos em répteis. A prevalência destes protozoários foi estudada em todas as Ilhas Canárias entre três géneros de répteis: *Gallotia*, *Tarentola* e *Chalcides*. Todos estavam infectados, embora algumas diferenças significantes tenham sido encontradas, sendo *Gallotia* o género com a prevalência mais alta. Os outros dois géneros, apesar de também infectados, demonstraram menor intensidade. A partir da sequenciação de parte do gene 18S rRNA, foi realizada uma avaliação molecular para determinar possíveis padrões de co-evolução dos parasitas com o seu hospedeiro vertebrado e para aferir a ocorrência de trocas de hospedeiro. Análises filogenéticas foram também efectuadas para ver como espécies de *Hepatozoon* destas ilhas estão relacionadas com outras linhagens de parasitas conhecidas.

Variation in parasitaemia levels of blood parasites in reptiles endemic to the Canary Islands

Members of the genus *Hepatozoon* are the most common and widely distributed intracellular parasites found in reptiles. The prevalence of these protozoans was studied in all the Canary Islands within three reptile genera: *Gallotia*, *Tarentola* and *Chalcides*. All of them were infected, but some significant differences were found, with *Gallotia* being the genus with the highest prevalence. The other two genera were also infected but with lower prevalence. Molecular assessment, by sequencing part of the 18S rRNA gene, was performed to determine possible patterns of co-evolution of the parasites with their vertebrate host and to assess host-switch events. Phylogenetic analysis was also performed to see how *Hepatozoon* species from these islands are related to other known parasite lineages.

Hepatozoon, co-evolution, host-switch, *Gallotia*, *Chalcides*, *Tarentola*.

Hiding deep in the blood: a survey of hemoparasites of wild endemic reptiles from Madagascar

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Madagascar is one of the world's top 12 "megadiversity" hot spots hosting unique and threatened flora and fauna. Parasites are a major component of biodiversity but remain largely uncharacterized in wildlife, despite the importance they might have in establishing conservation priorities. In this study we combine microscopic and molecular assessment of hemoparasites in endemic reptile species from Madagascar. We detected two distinct parasite groups: the apicomplexans *Hepatozoon* and *Sarcocystis*, and the filarial nematodes. Prevalence and intensity of the apicomplexans were low overall, while microfilaria infections in chameleons (*Furcifer* sp.) were relatively high. We detected mixed infections of two hemogregarine haplotypes in *Madagascarophis colubrinus*, and of hemogregarines and microfilaria in a chameleon. Molecular analyses of hemogregarines suggest occurrence of prey-predator transmission with identical sequences found in the snakes *M. colubrinus* and *Ithyocyphus oursi*, and their chameleon preys (*Furcifer* sp.). Based on previous studies on *Hepatozoon domerguei*'s lifecycle in these hosts and due to their morphological similarity, we identified the hemogregarine parasites as *H. domerguei*. However, future studies that include the examination of invertebrate hosts are needed to confirm this preliminary taxonomic identification. A distinct hemogregarine haplotype was found in the iguanid lizard *Oplurus* sp., which also displayed morphologically different gametocytes and should be further investigated. *Sarcocystis*, which is known to have severe effects in some of its hosts (e.g. wild rats), was identified in the blood of *Tracheloptychus petersi*, a lizard species listed as Vulnerable under IUCN red list criteria and with a decreasing population trend. This is the first report of this parasite in this lizard species and its implications need to be further studied. Lastly, by combining morphological and genetic information, *Foleyella furcata* filarial nematodes were identified in several *Furcifer* chameleons. This study provides insights on the distribution, diversity and host-parasite interactions of hemoparasites in wild reptile populations from Madagascar, which may be important not only for compiling the list of biological diversity of Madagascar but also for future risk assessments and conservation measures.

Apicomplexa, *Hepatozoon*, hemogregarine, *Sarcocystis*, nematode, arthropod borne diseases.

Niveles bajos de contaminación química antropogénica pueden amenazar a los anfibios al afectar al reconocimiento de los depredadores

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La teoría actual sugiere que la mortalidad directa y los efectos fisiológicos causados por los contaminantes son factores principales implicados en el declive global de anfibios. Sin embargo, se ha demostrado que concentraciones no letales de contaminantes pueden actuar de forma sinérgica con otros factores como la depredación causando elevadas tasas de mortalidad en las larvas de anfibios. En este estudio mostramos cómo concentraciones no letales de contaminantes suponen una amenaza para los anfibios, al alterar sus sistemas de reconocimiento de depredadores, y en consecuencia, incrementar el riesgo de depredación. Este efecto es incluso más importante, dado que cantidades pequeñas de contaminantes se encuentran omnipresentes en el medio. La alteración de la naturaleza química del medio en sistemas acuáticos podría ser una importante causa en los declives sufridos por las poblaciones de anfibios. Analizamos los efectos potenciales de dos contaminantes (i.e. ácido húmico y nitrato de amonio) en la capacidad de los renacuajos de sapo de espuelas (*Pelobates cultripes*) para reconocer las pistas químicas de un depredador habitual, ninfas de la libélula *Anax imperator*. Los renacuajos de muchas especies de anfibios se basan en la detección de pistas químicas de depredadores disueltas en el agua para evaluar el riesgo de depredación, al cual responden reduciendo sus niveles de actividad. Así pues, comparamos la actividad natatoria de los renacuajos en presencia y ausencia de pistas químicas de libélula disueltas en el agua, a diferentes concentraciones de ácido húmico y nitrato de amonio. Observamos que estos contaminantes interfirieron en el reconocimiento de las pistas de los depredadores. Los renacuajos redujeron su actividad natatoria en respuesta a las pistas de los depredadores en ausencia de contaminantes, pero no respondieron a estos estímulos cuando el agua contenía ácido húmico o nitrato de amonio, incluso en bajas concentraciones. Es más, los cambios en la actividad de los renacuajos asociados a los contaminantes *per se* no fueron significativos, indicando que no se produjeron efectos de toxicidad.

Low levels of chemical anthropogenic pollution may threaten amphibians by impairing predator recognition

Current theory suggests that direct mortality and physiological effects caused by pollutants are major factors explaining global amphibian decline. However, it has been shown that sublethal concentrations of pollutants can act synergistically with other factors such as predators to cause high mortalities in amphibian larvae. Here we show how even sublethal concentrations of pollutants can constitute a dangerous hazard to amphibians by disrupting their predator recognition systems, and hence increasing the risk of predation. This effect is indeed much more important since very low amounts of pollutants are ubiquitous. Altering the natural chemical environment in aquatic systems may be an important cause for declines in amphibian populations. We analyzed the effects of two common contaminants (humic acid and ammonium nitrate) on the ability of tadpoles of the western spadefoot toad (*Pelobates cultripes*) to recognize chemical cues from a common predator, nymphs of the dragonfly *Anax imperator*. Tadpoles of many amphibian species rely on detection of water-borne predator cues to assess the risk of predation, to which they respond by reducing their activity level. Therefore, we compared tadpoles' swimming activity in the presence and absence of water-borne chemical cues from dragonflies, at different concentrations of humic acid and ammonium nitrate. We found that these pollutants interfered the recognition of predator cues. Tadpoles reduced swimming activity in response to predator cues in the absence of pollutants, whereas they remained unresponsive to these cues when either humic acid or ammonium nitrate were added to the water, even at low concentrations. Moreover, changes in tadpole activity associated to the pollutants themselves were not detected, indicating that there was no clear toxic effect.

Amphibians, ammonium nitrate, chemical cues, humic acid, predator recognition, water pollutants.

Rãs verdes residentes numa mina de urânio desativada: como é possível? O caso de estudo da Cunha BaixaSÉRGIO M. MARQUES^{1,2}, FERNANDO GONÇALVES^{1,2}, RUTH PEREIRA³¹Departamento de Biologia da Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.²CESAM (Centro de Estudos do Ambiente e do Mar), Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.³Departamento de Biologia, Faculdade de Ciências & CIIMAR - Interdisciplinary Centre of Marine and Environmental Research. Universidade do Porto, Porto, Portugal.

Apesar da preocupação generalizada com a persistência de anfíbios na Terra, devido à sua conhecida sensibilidade a diferentes tipos de stressores ambientais, encontraram-se em Portugal rãs verdes aparentemente saudáveis a habitar e a reproduzir-se dentro de um complexo mineiro de extração de urânio. Na sua globalidade, as várias lagoas de efluente no complexo são ricas em metais e radionuclídeos tendo, contudo, uma acidez distinta que modula a sua toxicidade. Independentemente das condições extremas presentes no complexo mineiro os organismos adultos parecem beneficiar da baixa pressão predatória existente, uma vez que são abundantes. Contudo este benefício implica alguns custos a nível de saúde dos animais que mostraram estar efetivamente expostos a metais devido aos níveis elevados de alguns destes como o Al, Fe, Cu, Se e U bioacumulados no fígado e rim, assim como com alterações histopatológicas concomitantes nestes órgãos, bem como nos pulmões. De modo a lidar com esta exposição, vários mecanismos pareceram estar ativos, nomeadamente i) a sobreprodução de melanomacrófagos, resultando num elevado número de grandes centros melanomacrófagos nos tecidos. Tal está provavelmente relacionado com o papel antioxidante da melanina, que é capaz de complexar metais, prevenindo que estes exerçam os seus efeitos tóxicos; ii) maior expressão de genes codificantes para proteínas (hemoglobina, albumina e fibrinogénio) que podem desempenhar um papel fundamental na desintoxicação de metais e atuar como antioxidantes, conferindo uma elevada proteção basal contra a toxicidade dos metais. Para ter uma nova perceção sobre a capacidade dos anfíbios em lidar com um tipo de contaminação, que inclui uma exposição tanto química como radiológica, este trabalho integra diferentes estudos (histopatológicos, genotóxicos, enzimáticos e genómicos) realizados com rã-verde residente num habitat improvável.

Green frogs inhabiting a deactivated uranium mine: how is it possible? The Cunha Baixa's case study

Despite the widespread concerns with the persistence of amphibians on earth due to their well known sensitivity to different kinds of environmental stressors, green frogs were found, apparently healthy, living and breeding in a uranium mine complex in Portugal. The various effluent ponds in the complex are globally metal and radionuclides-rich, having however distinct acidity that modulates their toxicity. Regardless of the extreme conditions present in the mine complex, the adults seem to benefit from the reduced predation pressure offered, being abundant there. Nevertheless this benefit implies some costs in terms of health of the animals, which have shown to be effectively exposed to metals due to the high levels of some of them like Al, Fe, Cu, Se and U bioaccumulated in the liver and kidney with concomitant histopathological changes in these organs as well as in lungs. To cope with this exposure several mechanisms seemed to be activated, namely i) the overproduction of melanomacrophages, resulting in high numbers of large melanomacrophagic centers in the tissues. This is probably related to the antioxidant role of melanin, which is able to complex metals, preventing them from exerting their toxic effects; ii) higher expression of genes coding for proteins (haemoglobin, albumin and fibrinogen) that can play a fundamental role in metal detoxification and act as antioxidants, providing high basal protection against metal toxicity. To provide new insights about the ability of amphibians to cope with a particular kind of contamination, which includes both a chemical and a radiological exposure, this work integrates different studies (histopathological, genotoxic, enzymatic and genomic) carried out with Iberian green frogs occupying an improbable habitat

Amphibians, *Pelophylax perezi*, contamination resistance mechanisms, uranium mine, extreme habitat.

Metal tolerance inheritance may unveil possible patterns of microevolution in amphibian populations

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Chemical contamination may lead to the occurrence of genetic erosion in natural populations, and, consequently, may impair their viability under future environmental perturbations. This is of particular concern if the loss of genetic variability is irreversible due, for example, to the contaminant-driven elimination of alleles when chemical tolerance is a fully or incomplete recessive (or incompletely dominant) trait – the recessive tolerance inheritance hypothesis. Accordingly, this work aimed at investigating metal tolerance inheritance in natural populations of the frog species *Pelophylax perezi*. Twenty-one egg masses, at Gosner stage 8-10, were collected from a population inhabiting a metal free pond and exposed to a metal rich acid mine effluent or to a control (FETAX medium). Time to death was registered for each egg, with observations following a logarithmic time scale: 12h00min, 16h57min, 23h57min, 33h49min, 47h46min, 67h29min and 95h19min. For each egg mass, the median lethal time (LT50) and respective quartiles (LT25 and LT75) were computed. Subsequently, critically sensitive egg masses (categorized as those with an LT75 similar or below the average of the set of LT50 for all egg masses) were identified. The within egg mass variability in time to death responses was evaluated through its relative spread: the difference between the lower and upper quartiles relatively to the median, $[(LT75-LT25)/LT50]$. If metal tolerance corresponded hypothesis were true we should expect that: 1) no egg mass should present the full range of tolerance (not observed); 2) intermediate tolerant egg masses should present larger relative spreads (not observed). On the contrary, we observed that 1) most of the masses presented the full range of tolerance, 2) median tolerance and relative spread positively correlated when excluding egg mass J (the only one with a bimodal distribution). These results could be explained by (fully or nearly) co-dominance mechanism but a convex pattern of relative spreads would be expected. Additional possible explanations are: 1) differential tolerance could be linked to differential phenotypic plasticity, 2) differential tolerance could be linked to different genetically determined physiological mechanisms. In conclusion, if tolerance is indeed (fully or nearly) co-dominant, then a strong enough contamination pulse would eradicate sensitive and intermediately tolerant genotypes, eventually leading to allele's loss.

Recessive tolerance inheritance hypothesis, acid mine drainage, genetic erosion, *Pelophylax perezi*.

Avaliação da diversidade genética de populações de anfíbios em habitats aquáticos contaminados

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A contínua contaminação de água e solos pode contribuir para um decréscimo da diversidade genética das populações naturais, reduzindo a sua capacidade de lidar com condições menos favoráveis ou ambientes em mudança. Em particular, os anfíbios são um grupo muito sensível, graças à permeabilidade da sua pele que permite uma respiração cutânea, embora a presença de protecção física também permita a difusão química dos agentes presentes no meio ambiente. Assim, as populações mundiais de anfíbios têm vindo a diminuir, devido a contaminações químicas, alterações climáticas, perda de habitats e presença de espécies invasoras.

Com este trabalho, pretendemos caracterizar geneticamente quatro populações naturais de *Pelophylax perezi* expostas a diferentes tipos de contaminação, usando dezasseis adultos (8 fêmeas e 8 machos) de cada um dos sítios de colheita. Para tal, usámos diferentes tipos de marcadores moleculares. Pesquisámos SNPs em dois genes mitocondriais, o gene RNA ribossomal 12S e o gene da subunidade 2 da NADH desidrogenase, bem como em duas sequências de DNA autossómico, o intrão 1 da sérum albumina e o gene da rodopsina. Analisámos também 10 loci de STRs. Com os dados obtidos, foram feitas análises de diversidade e subestruturação, usando software como Arlequin, Structure e Network. Os resultados dos marcadores autossómicos e mitocondriais mostraram, na generalidade, uma baixa diversidade genética e uma não diferenciação de populações, embora cada população possuísse genótipos únicos. Apenas o gene da rodopsina apresentou indivíduos heterozigóticos e o gene codificador da proteína ND2 foi o marcador mais polimórfico com 22 SNPs. Oito SNPs são responsáveis por mutações missense e três destas são previstas como deletérias. Os resultados podem indicar uma perda de genótipos. As análises de STRs estão a ser conduzidas e os resultados preliminares mostram também uma baixa diversidade e ausência de diferenciação das populações. Em conclusão, este trabalho mostra que a contaminação de habitat em algumas populações naturais de anfíbios pode não afectar as suas características genéticas, uma vez que todas as populações apresentaram baixa diversidade e ausência de diferenciação. É necessário então a continuação deste trabalho, por exemplo, com genes codificadores de enzimas anti-oxidantes (como catalase, superóxido dismutase ou glutathione peroxidase), ou outros marcadores selectivos que possam reflectir melhor as respostas dos organismos às contaminações dos habitats.

Assessment of the genetic diversity in amphibian populations from water-contaminated habitats

Chronic contamination of waters and soils may contribute to a decreased genetic diversity in natural populations, reducing their ability to cope with less favorable conditions or changing environments. In particular, amphibians are a very sensitive group of species due to their permeable skin that allows cutaneous respiration, although the absence of physical protection also allows the diffusion of chemical agents present in the environment. Thus, worldwide natural populations of amphibians are declining, due to chemical contamination, climate changes, habitat loss, and also presence of invasive species.

With this work, we aim to genetically characterize four natural populations of the common green frog *Pelophylax perezi* exposed to different types of contamination, using sixteen adult organisms (8 females plus 8 males) from each collection site. Analyses were performed using different types of molecular markers. We searched for SNPs in two mitochondrial DNA genes, the 12S ribosomal RNA gene and the NADH dehydrogenase subunit 2 (ND2), and two autosomal DNA sequences from serum albumin intron 1 (SAI-1) and rhodopsin genes. We also analyzed 10 STR loci. Diversity and structure analyses were computed using Arlequin, Structure and Network software. Results from mitochondrial and autosomal markers showed, in general, a low genetic diversity and no differentiation among the different populations, although there were some unique genotypes in each population. Heterozygous individuals were found only for the rhodopsin gene and the ND2 protein-coding gene was the most polymorphic marker, with 22 SNPs. Eight SNPs originated missense mutations, and three of these were predicted as deleterious. The results may indicate loss of genotypes in these populations. STR analyses are in progress and preliminary data show also evidence of low genetic diversity and absence of population differentiation. Altogether, this work showed that habitat contamination in some natural populations of amphibians may not affect their genetic make-up, as all populations presented a low genetic diversity and no differentiation was found

between populations. Further work will be needed, for instances, with genes coding for anti-oxidant enzymes (e.g., catalase, superoxide dismutase and glutathione peroxidase), or other selectable markers that can better reflect the response to habitat contamination.

Pelophylax perezi, habitat contamination, genetic diversity.

Lípidos, carotenoides y POPs en huevos de la Tortuga laúd (*Dermochelys coriacea*): interacciones e influencia en el éxito reproductor

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Se aportan datos de referencia sobre los perfiles de ácidos grasos, carotenoides y las concentraciones de contaminantes orgánicos persistentes (POP) en huevos de la tortuga laúd, *Dermochelys coriacea*. Además se han investigado las asociaciones entre estos compuestos y se ha evaluado como pueden afectar el éxito reproductivo en esta especie. Se evaluaron 18 nidos durante junio y agosto de 2008 en la Reserva Natural Pacuare, Costa Rica. Se midió para cada nido la longitud curva del caparazón y el ancho curvo del caparazón de la madre, el tamaño de puesta y el número de falsos huevos (SAG, estructuras de albumen con cáscara, sin embrión ni vitelo, característicos de esta especie) en el momento de la ovoposición. De uno a tres huevos viables se obtuvieron de cada nido (47 huevos en total). Se calculó la viabilidad, fecundidad y las tasas de eclosión de cada nido. Los lípidos, carotenoides y los POPs se analizaron en todos los huevos. Se calculó la media \pm SD para cada compuesto y para cada nido. Los niveles de POPs fueron similares a los observados en poblaciones de la Guayana Francesa y ligeramente más bajos que los asociados a las poblaciones de Florida. Todos los huevos se caracterizaron por poseer niveles bajos de carotenoides, y se encontró una relación positiva entre la concentración de carotenoides y la proporción de huevos viables. Encontramos una correlación positiva e intensa entre POPs y PUFAs (ácidos grasos insaturados), lo que sugiere interacciones moleculares. El efecto negativo de los POPs sobre los embriones estaría enmascarado por el efecto positivo de los ácidos grasos. Los PUFAs parecen aumentar la tasa de fecundidad y la longitud de las crías; mientras que los éteres difenilos polibromados (PBDE), un tipo particular de POPs, mostraron una correlación negativa con el éxito de eclosión. Todos los compuestos estudiados estaban relacionados con alguno de los parámetros reproductivos. Este estudio proporciona evidencias potenciales de que los PUFA mejoraran la fertilidad y el desarrollo embrionario, mientras que el nivel de carotenoides limita la vitelogenénesis, y la concentración de POPs influye en el éxito de eclosión.

Lipids, carotenoids and POP concentrations in eggs of the Leatherback turtle (*Dermochelys coriacea*): Potential influences on reproduction

The aims of this study were to provide additional baseline data on fatty acid profiles, carotenoids and persistent organic pollutant (POP) concentrations in eggs of the Leatherback sea turtle, *Dermochelys coriacea*, to investigate any associations among these compounds and to clear up how any of these compounds may affect reproductive success in this species. Eighteen clutches were evaluated during June and August 2008 at Reserva Pacuare Beach, Costa Rica. Curved carapace length, curved carapace width, clutch size and number of shelled albumen globules (SAGs) were determined at the time of oviposition. One to three viable eggs were collected from each nest (47 eggs total). Viability, fertility and hatching rates were calculated for each nest. Lipids, carotenoids and POPs were analysed on each egg. Mean \pm SD values were calculated for each compound and for each clutch. Correlations were performed searching for interactions among different compounds and for potential effects on reproductive parameters. POP levels were similar to those reported in French Guiana populations and slightly lower than those associated with Florida populations. Low carotenoid levels characterized all eggs of this species, and a positive relationship between carotenoid concentrations and the viability rate was found. POPs and polyunsaturated fatty acids (PUFAs) were positively and strongly correlated, suggesting molecular interactions. PUFAs appeared to increase fertility rate and hatchling length, whereas polybrominated diphenyl ethers (PBDEs), a particular kind of POP, showed a negative correlation to the hatching success. All the studied compounds were related to any of the reproductive parameters. This study provides potential evidences of PUFA enhancing fertility and hatchling size, of carotenoids limiting vitellogenesis and of PBDEs influencing hatching success. A positive correlation found between POPs and PUFAs may indicate that harmful effects of these contaminants on the reproduction of Leatherback turtles may be masked.

Pollution, ecophysiology, eggs, Leatherback turtle, carotenoids, lipids.

¿Tenemos en cuenta todas las vías relevantes al exponer larvas de anfibios a plaguicidas?

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La legislación de la UE sobre fitosanitarios permite vender plaguicidas sin analizar su toxicidad sobre las fases acuáticas de anfibios, las cuales estarían cubiertas por los datos obtenidos de peces. Aparte de las diferencias de sensibilidad entre taxones, los escenarios usados en peces no son totalmente representativos de los hábitats de anfibios. Por ejemplo, dado que la ingesta de plaguicidas es poco relevante para los peces, la absorción dérmica es la única ruta de exposición considerada. Nuestra hipótesis es que otras matrices también pueden ser importantes en la exposición de anfibios a plaguicidas. Presentamos tres experimentos realizados con plaguicidas formulados cuyos ingredientes activos son un herbicida (glufosinato amónico, GA), un fungicida (folpet) o un insecticida (fenpiroximato). Expusimos larvas de *Rana temporaria* a concentraciones ambientales estimadas durante 21 días (procedimiento aprobado por la autoridad en bienestar animal de Renania-Palatinado). Establecimos tres rutas de exposición aplicando los plaguicidas en el agua, sedimento o alimento, además de un tratamiento combinando las tres rutas. Independientemente de la ruta, los tres plaguicidas retrasaron el desarrollo ($p \leq 0,001$), especialmente en larvas expuestas a folpet en el alimento o a GA en el sedimento. La exposición a través del alimento afectó al crecimiento; las larvas tratadas con cualquiera de los compuestos mostraron menor masa que los controles (GA: 748 ± 17 ; folpet: 780 ± 8 ; fenpiroximato: 766 ± 10 mg control: 828 ± 6 ; $p \leq 0,053$), y las tratadas con GA o folpet mostraron también menor longitud cabeza-cloaca (LCC) (GA: $14,1 \pm 0,4$; folpet: $14,0 \pm 0,2$; control: $15,3 \pm 0,5$ mm; $p \leq 0,049$). La exposición a fenpiroximato combinando las tres rutas incrementó el efecto negativo sobre la LCC y la masa (4,0-6,4 y 5,8-13,4%, respectivamente, inferiores que en exposiciones mediante una sola ruta; $p \leq 0,091$). Igualmente, la condición corporal de las larvas expuestas a folpet a través de la combinación de las tres rutas fue un 9,9-10,6% inferior a la de las lavas expuestas mediante una única vía ($p \leq 0,038$). También observamos un aumento leve pero significativo del porcentaje de larvas anómalas ($7,5 \pm 3,5\%$; $p < 0,001$) entre las expuestas a folpet en el sedimento. Estos datos muestran la importancia de las matrices de exposición a plaguicidas distintas del agua para larvas de *R. temporaria*. Dado que la legislación de la UE considera seguras las condiciones de exposición empleadas, nuestros resultados apuntan a la necesidad de incluir en el análisis de riesgos escenarios de exposición a plaguicidas específicos de anfibios europeos. Financiado por el 7º Programa Marco (IEF-People) de la Comisión Europea.

Are all relevant routes taken into account when exposing amphibian tadpoles to pesticides?

The EU legislation on plant protection products allows pesticides to be placed on the market without testing their toxicity on aquatic amphibian stages, being these covered by fish-derived data. Besides differences between taxa in sensitivity, the exposure scenarios used for fish are not fully representative of amphibian environments. For instance, because pesticide ingestion has little relevance in fish, dermal uptake from the water is considered as the only exposure route. We hypothesize that other exposure matrices can also be important for pesticide uptake in amphibian tadpoles. We present three experiments conducted with pesticide formulations containing as active ingredients an herbicide (glufosinate ammonium, GA), a fungicide (folpet) or an insecticide (fenpyroximate). We exposed *Rana temporaria* tadpoles to predicted environmental concentrations during 21 days (procedures approved by the Rhineland-Palatinate authority on animal welfare). We established three exposure routes applying pesticides to either water, sediment or food, plus an additional treatment with the three routes simultaneously. Regardless of the exposure route, the three pesticides delayed development (all $p \leq 0.001$), being this effect especially severe on tadpoles exposed to folpet via food, or to GA via sediment. The exposure through food affected growth rate; tadpoles treated with either compound showed lower body mass than controls (GA: 748 ± 17 ; folpet: 780 ± 8 ; fenpyroximate: 766 ± 10 ; control: 828 ± 6 mg; $p \leq 0.053$), and those treated with GA or folpet had also shorter snout-vent length (SVL) (GA: 14.1 ± 0.4 ; folpet: 14.0 ± 0.2 ; control: 15.3 ± 0.5 mm; $p \leq 0.049$). When compared to single-route exposures, the three combined routes increased the negative effect of fenpyroximate on SVL and mass (4.0-6.4 and 5.8-13.4% lower in the three-route treatment, respectively; $p \leq 0.091$). Likewise, tadpoles exposed to folpet through the three routes showed a body condition 9.9 to 10.6% lower than tadpoles exposed through one route only ($p \leq 0.038$). We also found a slight but significant occurrence of abnormalities ($7.5 \pm 3.5\%$; $p < 0.001$) in tadpoles exposed to folpet via sediment. These data highlight the importance of pesticide exposure matrices other than water in *R. temporaria* tadpoles. Because the exposure conditions used in this study are considered as safe by the EU legislation, our results support the necessity of including European amphibian-specific exposure scenarios in pesticide risk assessment. Financed by the European Commission through the 7th Framework Program (IEF-People).

Risk assessment, plant protection products, predicted environmental concentrations, *Rana temporaria*, sediment, food intake.

***Podarcis sicula*: um colonizador de sucesso, um invasor perigoso**

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O fenómeno de invasões biológicas é um problema crescente e preocupante no que toca a conservação da biodiversidade, devido à ameaça para as espécies nativas. A lagartixa italiana, *Podarcis sicula* é um dos répteis que está a ser introduzido em todo o mundo. A sua distribuição nativa compreende a Península Italiana, a Sicília e a Costa Norte do Adriático, e é considerada introduzida nas Ilhas Tyrrhenians, Córsega e Sardenha, Menorca nas Ilhas Baleares e nas ilhas e costa a este do Adriático. Para além destas regiões, populações isoladas podem ser encontradas na Península Ibérica, Sul de França, Suíça, Turquia, Grécia, Reino Unido, Norte de África e Estados Unidos da América. Esta lagartixa pode ser encontrada numa variedade de habitats, desde habitats naturais com vegetação Mediterrânica até agro-ambientes e áreas urbanas onde há uma maior probabilidade de ser transportada acidentalmente. Para além disso, estudos prévios indicam hibridização e competição entre as populações introduzidas e espécies de *Podarcis* nativas, reforçando o carácter invasor desta lagartixa. Assim, a determinação das vias e fontes da invasão é essencial para que melhores medidas de conservação sejam colocadas em prática. Se as populações tiverem uma origem comum, as medidas deverão focar-se nessa área. No entanto, se as origens forem múltiplas, a gestão deverá ser orientada à espécie e não à área. Neste trabalho, integramos os dados genéticos de todas as populações exóticas já publicados e os gerados por nós, com a informação filogeográfica da espécie da sua área nativa. A análise filogenética, realizada com base no gene mitocondrial Cyt-b, sugere eventos múltiplos e independentes de colonização originários de vários pontos da sua distribuição nativa. Este facto é mais um suporte ao carácter invasor da espécie. No que toca a medidas de conservação, a erradicação só é possível se as populações forem detectadas cedo, antes da sua expansão ocorrer. A prevenção, nomeadamente tendo em conta os recentes meios pelos quais esta espécie está a ser introduzida, comércio de animais, carga e comércio de plantas, deverá ser priorizada.

***Podarcis sicula*: a successful coloniser, a hazardous invader**

Biological invasions are nowadays a major concern to biodiversity conservation due to the threat to native biota. The Italian wall lizard, *Podarcis sicula* is one reptile species that is being introduced worldwide. From its native distribution in the Italian Peninsula, Sicily and north Adriatic coast, this species is considered introduced in the Tyrrhenian Islands, Corsica and Sardinia, Menorca in the Balearics and in the islands and coastal areas of the eastern Adriatic Sea. Besides these regions, scattered populations are now known from Iberian Peninsula, Southern France, Switzerland, Turkey, Greece, Britain, North Africa and United States. This lizard is able to inhabit a variety of habitats, from natural Mediterranean vegetation to agroenvironments and urban areas which likely make it prone to be passively transported. Moreover, the reports of hybridisation and competition of introduced populations with native *Podarcis* suggest an invasive character. At this point, determining the invasion pathways is crucial in conservation terms. If populations had a common origin, conservation measures should be addressed to the putative source area. However, if origins were multiple, management should be species-oriented. Here, we join newly generated and published genetic data on all alien populations known to date together with the phylogeographic information of the species in its native range. Phylogenetic reconstruction based on the mitochondrial gene Cyt-b indicates multiple independent colonisation events originated from source areas throughout most of the species range. This, hence, supports that the invasive character extends to the whole species. While eradication is only feasible for those populations in early stages of invasion, urgent management measures are to be concentrated in prevention, namely, regarding the recent invasion pathways as pet trade, cargo and nursery trade.

Biological invasions, Italian wall lizard, origin, cytochrome b.

Transmisión de parásitos desde tortugas alóctonas: una nueva amenaza para las tortugas autóctonasRAÚL IGLESIAS¹, JOSÉ M. GARCÍA-ESTÉVEZ¹, CESAR AYRES², ANTONIO ACUÑA³, ADOLFO CORDERO-RIVERA⁴¹Laboratorio de Parasitología, Facultad de Biología, Universidad de Vigo. Campus Lagoas-Marcosende, 36310 Vigo, Spain.²AHE (Asociación Herpetológica Española). Apartado de correos 191, 28911 Leganés, Madrid, Spain.³Veterinary surgeon, OAM Parque das Ciencias Vigozoo. A Madroa, Teis, 36316 Vigo, Spain.⁴Evolutionary Ecology Group, Dept. Ecology and Animal Biology, EUE Forestal, University of Vigo. Campus Universitario A Xunqueira s/n, 36005 Pontevedra, Spain.

Durante el invierno de 2012-2013 se detectó un episodio de mortalidad en una población de *Emys orbicularis* que habita un conjunto de lagunas en Galicia. La mayoría de los ejemplares afectados mostraban una pérdida de movilidad en las extremidades, y la mayoría fallecieron a los pocos días de ser detectados. En la necropsia de los ejemplares se detectó la presencia de trematodos vasculares pertenecientes a la especie *Spirorchis elegans*, así como huevos atrapados en la mayoría de los órganos internos. Se realizó un control sanitario de las tres poblaciones naturales (Gándaras de Budiño, Arnoia y Avia) y de la población creada artificialmente en el Parque Natural de Corrubedo. Se detectaron ejemplares afectados tanto en la población de Gándaras de Budiño como en la población asociada al río Avia. El origen norteamericano de *Spirorchis elegans*, la ausencia de epizootias similares anteriores en el área del brote, y la presencia habitual de su huésped tipo, la altamente invasiva tortuga de orejas rojas, en esta área sugiere un nuevo caso de contagio de parásitos que resulta en una severa enfermedad emergente.

Parasite spill-over from alien turtles: a new threat for autochthonous turtles

During the winter of 2012-2013 an episode of mortality was detected in a population of *Emys orbicularis* inhabiting a set of clay pits in Galicia (NW Spain). Most of the affected individuals showed a loss of mobility in the limbs, and most died within a few days of being detected. At necropsy, the presence of vascular trematodes belonging to the species *Spirorchis elegans* were detected, as well as eggs trapped in most of the internal organs. A health inspection was held at the three natural populations (Gándaras de Budiño, Arnoia and Avia) and at the population artificially created in the Natural Park of Corrubedo. Affected individuals were detected in Gándaras de Budiño and in the population associated with the river Avia. The North American origin of *Spirorchis elegans*, the absence of prior similar epizootics in the outbreak area, and the habitual presence of its type host, the highly invasive red-eared slider, in this area suggest a new case of parasite spill-over resulting in a severe emerging disease.

Parasite, spill-over, *Trachemys*, *Emys*, mortality, NW Spain.

Control de la especie invasora, *Lampropeltis californiae* en la isla de Gran Canaria - LIFE10NAT/ES/000565

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La culebra real de California (*Lampropeltis californiae*) fue detectada por primera vez en 1998 en el Barranco Real de Telde (La Solana) en el este de Gran Canaria. En la primavera y verano de 2007 se contabilizaron cientos de observaciones en La Solana, confirmándose ese año su naturalización. En septiembre de 2011 se inició el proyecto LIFE+LAMPROPELTIS, financiado por la Unión Europea, va a permitir el desarrollo de nuevas técnicas de captura, el mejor conocimiento de la actividad biológica, su interacción con las variables ambientales del entorno, y el aumento del papel de colaboración de la población en la lucha contra la invasión. El trabajo del equipo de operarios del proyecto, junto con la inestimable colaboración ciudadana a través del Sistema de Alerta Temprana, ha permitido capturar desde el inicio del proyecto en septiembre de 2011 hasta junio de 2014, 1416 ejemplares, de los cuales 781 lo han sido por colaboración ciudadana, 189 mediante el uso de distintos tipos de trampas y el resto por captura directa del personal del proyecto mediante acecho. Desde el año 2013 se están utilizando perros y busardos de Harris (*Parabuteo unicinctus*) para localizar serpientes, aportando hasta la fecha 6 y 10 capturas respectivamente. Las capturas se realizan principalmente durante los meses de marzo a junio, aunque con variaciones entre un año y otro, y disminuyendo en verano debido a las altas temperaturas y a la falta de humedad. El mayor éxito de capturas está asociado al período de apareamiento de la especie. En los primeros momentos de mayor actividad, las capturas se corresponden principalmente con individuos adultos con un mayor porcentaje de machos. A partir de marzo, aparecen hembras preñadas y el número de hembras capturadas supera al de machos. Los datos biométricos muestran que todos los individuos responden al fenotipo de culebra real de California. Esta culebra muestra cuatro diferentes patrones de color: normal lineal, normal anillado, albino lineal y albino anillado. Ambas poblaciones muestran diferencias significativas en el patrón de color. En Telde-Valsequillo prevalecen las culebras albinas, mientras que en Gáldar la mayoría de las culebras son normales lineales. Todos los datos anteriores muestran la sobresaliente adaptación de la culebra al medio natural en Gran Canaria, lo que convierte a esta especie en un serio problema medioambiental, especialmente para las poblaciones endémicas de reptiles, el lagarto de Gran Canaria (*Gallotia stehlini*), la lisa de Gran Canaria (*Chalcides sexlineatus*) y el perenquén (*Tarentola boettgeri*). El caso del lagarto de Gran Canaria, parece especialmente preocupante, ya que en el estudio realizado dentro del proyecto LIFE, se ha podido constatar la reducción de ejemplares jóvenes y de talla media en las zonas con presencia de culebra real de California, con densidades 10 veces inferiores a zonas sin presencia de culebra.

Control of the invasive alien species *Lampropeltis californiae* on the island of Gran Canaria – LIFE10NAT/ES/000565

In 1998, the California Kingsnake was first found in El Barranco Real de Telde (La Solana) in the East of Gran Canaria. In spring and summer 2007, hundreds of sightings were recorded in La Solana. The Life project began in September 2011, funded by the European Union. This project will develop new actions in order to improve the results obtained to control the California kingsnake, minimizing its impact on native biodiversity, the implementation of proven techniques for the detection and capture of exotic invasive snakes introduced onto other islands, and raise awareness and encourage involvement of all pertinent community sectors. The technical work of the project field, together with the invaluable cooperation of citizens through the Early Warning System has enabled capturing from the start of the LIFE + LAMPROPELTIS in September 2011 until June of 2014, 1416 snakes, of which 781 have been through citizen cooperation, 189 by using different types of traps and the rest by direct capture by team work. Since 2013 they are using dogs to locate snakes and Harris's Hawk (*Parabuteo unicinctus*), with 6 and 10 captures respectively. The captures have been done mainly from March to June, decreasing in summer due to the high temperatures and in the absence of moisture. The capture success is closely related to the snakes' activity and mating. Most captured individuals were adult and up until May the rate of male snakes was higher than female. From March onwards, pregnant snakes were found and in May the rate changed; catching more female snakes. The biometric data and the foliosis show that all the individuals caught in Gran Canaria belong to the Californian kingsnake. These snakes showed four different color patterns: banded, striped, banded albino and striped albino. Both populations show significant differences in the color pattern. In Telde-Valsequillo, the albino snakes prevail, whereas in Gáldar most snakes are striped and the number of albinos is around 4%. This data suggests that the population in Gáldar comes from a new introduction. Genetic analyses in this project confirm this hypothesis. All previous data shows that the California Kingsnake has a high ability of adaptation and acclimatization, so that the spread

all over the island is more than likely, which could pose a serious risk to critically endangered species, especially for endemic populations of reptiles, the lizard of Gran Canaria (*Gallotia stehlini*), smooth Gran Canaria (*Chalcides sexlineatus*) and Canary Island Wall Gecko (*Tarentola boettgeri*). The case of Gran Canaria lizard seems particularly worrying, since in the study within the project LIFE has been able to confirm the reduction of juveniles and of average height in areas where California Kingsnake is naturalized.

Kingsnake, *Lampropeltis*, aliens, reptiles, Canary Islands.

¿Se equivocó Plinio el Viejo? Primeros resultados de la lucha contra los ofidios introducidos en Ibiza

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Hace 2000 años, Plinio el Viejo y otros autores dejaron escrito “Ebusi, terra serpentes fugat”; sin embargo, la presencia de serpientes introducidas en la isla de Ibiza esta constatada desde 2003. Este año, el Consell Insular de Ibiza ha iniciado un programa piloto para evaluar su distribución, el estado de sus poblaciones y aspectos sobre su biología que nos indiquen las líneas a seguir para evitar que su expansión continúe. Este proyecto está siendo ejecutado por la Asociación Herpetológica Española.

Se han realizado mapas de distribución a partir de 146 observaciones en el año 2014, y se han tomado datos básicos de biometría, dieta, depredación, reproducción y estructura de poblaciones en las tres especies inicialmente introducidas: *Hemorrhois hippocrepis*, *Malpolon monspessulanus* y *Rhinechis scalaris*. Para ello se han instalado 93 trampas, sin cebar y con seis tipos distintos de cebo, y se ha contado con una unidad canina entrenada en la búsqueda activa de ejemplares en el medio natural.

Los datos preliminares indican que dos especies introducidas en la isla de Ibiza están naturalizadas y son abundantes en el entorno de Santa Eularia, Sant Llorenç y Santa Gertrudis, predominando *H. hippocrepis*, y siendo más escasa *R. scalaris*; sin embargo, no hay datos de *M. monspessulanus* desde hace cinco años. Las dos especies presentes en la actualidad realizan cortejos y cópulas en el mismo periodo que en las regiones donde son nativas. Al menos *H. hippocrepis* realiza puestas con huevos viables. La presencia de neonatos en fechas de eclosión parece constatar la reproducción efectiva (ejemplares de *R. scalaris* de 16 g de masa corporal y 29 cm de longitud total observados el 05/10/2013; ejemplares de *H. hippocrepis* de 9 g y 38 cm observados el 24/10/2013). Al menos en *H. hippocrepis* se ha constatado la depredación sobre el único reptil nativo y endémico de las islas Pitiusas, *Podarcis pityusensis*, y otros datos apuntan a la depredación sobre polladas de aves nativas, algunas endémicas. En el breve plazo desde la introducción, *H. hippocrepis* ha mostrado una respuesta funcional hacia el gigantismo, con ejemplares mostrando el 213% de la biomasa máxima registrada en ejemplares en su área nativa de la península Ibérica.

La utilización de trampas parece efectiva siempre que su número sea elevado. La elección del tipo de trampa, cebo y lugar de colocación condiciona los resultados obtenidos. Sin embargo, la continuidad de este estudio a lo largo del año 2015 se hace imprescindible para aumentar el tamaño de muestra. El próximo año se pretende la utilización de nuevos métodos de captura, como redes de intercepción y enmalle, adecuación de las trampas de doble embudo, utilización de hembras en celo como cebo, y especialmente la colocación de mayor número de trampas de cajón.

Was Pliny the Elder wrong? First results on the control of the invasive alien snakes in Ibiza

Two thousand years ago, Pliny and other authors wrote “Ebusi, terra serpentes fugat”; however, the presence of snakes introduced to the island of Ibiza has been observed since 2003. This year, the Consell Insular de Ibiza has initiated a pilot project to assess their distribution, population status, and aspects of their biology, to establish the guidelines to be followed in order to prevent their expansion. This project is being executed by the Spanish Herpetological Association.

Distribution maps from 146 observations in 2014 have been elaborated, and data on basic biometry, diet, predation, reproduction, and population structure of the three species originally introduced, *Hemorrhois hippocrepis*, *Malpolon monspessulanus*, and *Rhinechis scalaris*, have been retrieved. For this purpose, 93 traps were installed, without bait and with six different types of bait, and we have counted also of a canine unit trained in the active search of specimens in the wild. Preliminary data indicate that two introduced species on the island of Ibiza are naturalized and are abundant in the vicinity of Santa Eulalia, Santa Gertrudis and Sant Llorenç, being *H. hippocrepis* predominant, while *R. scalaris* is still scarce. However, there are no data from *M. monspessulanus* in the last five years. The two species currently present in the island perform courtship and mating in the same period as in the regions where they are native. At least *H. hippocrepis* lays viable eggs. The presence of hatchlings seems to confirm successful reproduction (*R. scalaris* with 16 g of body mass and 29 cm of total length observed on October 5th 2013; *H. hippocrepis* with 9 g and 38 cm observed on October 24th 2013). At least in *H. hippocrepis*, predation on the only native, and endemic reptile in the Pityusic Islands, *Podarcis pityusensis*, has been observed. Other data suggest predation on broods of native birds, some of them endemic to the Islands. In the short time

since its introduction, *H. hippocrepis* has shown a trend towards the gigantism, with individuals exhibiting up to 213% of the maximum biomass recorded in their native area in the Iberian Peninsula.

The use of traps seems effective if their number is high. The type of trap, bait and placement site affect the results. However, the continuation of this study throughout 2015 is essential to increase the sample size. Next year we intend to use new methods of capture, such as interception and gill nets, match double-funnel traps, use estrous females as baits, and especially place more box drawer traps.

Invasive, introduced snakes, Ibiza, control program.

Gestión de anfibios en Cataluña: control de especies alóctonas en los micro-hábitats acuáticos en las Terres de l'Ebre

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La introducción de especies exóticas en el medio natural está considerada como una de las causas más importantes de la pérdida de biodiversidad a escala global. Las especies exóticas pueden representar una amenaza para las especies autóctonas, entre otros motivos, depredando las especies autóctonas o su descendencia, compitiendo por el alimento u otros recursos o por transmisión de enfermedades. En el caso de los anfibios ya se han descrito agresiones de especies exóticas hacia este grupo. Desde 2007, en el marco del "Plan de Conservación de Gallipato en Cataluña", se está realizando un trabajo de seguimiento de la especie con el objetivo de conocer el estado actual de sus poblaciones así como su distribución, e identificar las principales amenazas a las que se enfrenta. Para ello, se han catalogado las especies exóticas presentes en diferentes balsas identificadas y se han realizado acciones encaminadas a erradicar o controlar sus poblaciones. Este trabajo se está llevando a cabo en las comarcas del Sur de Cataluña, en las Terres de l'Ebre. A fecha de hoy se han prospectado 130 balsas para comprobar la presencia de gallipato y catalogar el resto de especies de anfibios (mediante manguero) y/o comprobar la presencia de especies exóticas. Se han identificado siete especies exóticas invasoras en 19 de las 130 balsas prospectadas, pertenecientes a los distintos grupos (anfibios: una especie, reptiles: una especie, peces: cuatro especies e invertebrados: una especie). En función de las características particulares de cada microhábitat acuático y las especies encontradas en cada uno de ellos, se han utilizado diferentes técnicas de erradicación y control, algunas más agresivas que otras contra las especies exóticas. El plan de actuación se ha temporalizado de forma específica para minimizar sus efectos sobre la fauna autóctona. La aplicación de los diferentes métodos de control, dependiendo de las características de la balsa y de las especies invasoras, ha reducido el número de balsas infectadas con alguna o varias especies alóctonas invasoras de 19 en 2007 a únicamente tres en la actualidad. Los resultados de estas actuaciones han sido muy positivos para la fauna autóctona, especialmente para los anfibios.

Amphibian management in Catalonia: Control of non-native species in aquatic micro habitats in Terres de l'Ebre

Release of alien species in the wild is considered one of the most important issues in global biodiversity loss. Alien species may be a threat to native species populations by, amongst other things, preying on native species or their offspring, out-competing them for food or other resources, and/or causing or carrying disease. In the case of amphibians, attacks of alien species on this group have already been described. Since 2007, in the framework of the "Iberian ribbed newt Conservation Plan in Catalonia", we have monitored the species in order to assess the current status of its populations and distribution, and to identify the main threats for its populations. Non-native species occurring in rafts are catalogued and efforts are made to eradicate or control their populations. This work is being carried out in the regions of southern Catalonia, in Terres de l'Ebre. A total of 130 rafts have been prospected so far, both to check the presence of Iberian ribbed newt and list other amphibian species, and/or to catalogue the presence of exotic species. Seven invasive alien species have been found in 19 out of the 130 rafts, belonging to different groups (amphibians: one species, reptiles: one species, fish: four species and invertebrates: one species). According to the particular characteristics of each aquatic microhabitat, and the species found in each of them, we have used different techniques for eradication and control of exotic species, some more aggressive than others. This action plan was specifically scheduled to minimize its effects on native fauna. The use of different techniques of control, depending on the characteristics of the raft and invasive species, has reduced the number of infected rafts with one or more invasive alien species from 19 in 2007 to only three at the present. The results of these actions have been very positive for the native wildlife, especially amphibians.

Aquatic microhabitat, eradication, control, non-native species.

The discovery of *Batrachochytrium salamandrivorans* in Belgium: personal observations and preliminary European risk assessment

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Emerging infectious diseases are currently causing biodiversity loss on a global scale. The greatest disease-driven loss of biodiversity ever documented is being caused by amphibian chytridiomycosis. A new case of amphibian chytridiomycosis was recently discovered in the Netherlands, in the form of *Batrachochytrium salamandrivorans*.

During December 2013, the author performed two monitoring rounds in a hitherto abundant Belgian population of *Salamandra salamandra*, which led to the discovery of an individual displaying skin erosions, deep ulcerations across the entire body and a highly damaged tail. Presence of *B. salamandrivorans* was confirmed by Ghent University (Belgium). As this record represented the first individual found alive in a progressed state of *B. salamandrivorans* infection, an overview is given here of how *B. salamandrivorans* impacts individuals in-situ. Based on subsequent discoveries of this fungal threat in other sites, a preliminary geographical risk analysis for Europe is additionally provided using spatial data on the occurrence of susceptible species and an average speed of 15 km/year into account. In absence of dispersal barriers, *B. salamandrivorans* is in risk of spreading throughout most of Europe due to its current occurrence in an area characterized by continuous populations of *S. salamandra* reaching widely into Germany and France.

Batrachochytrium salamandrivorans, chytridiomycosis, *Salamandra salamandra*, fungus, Belgium.

Patrones espaciales y temporales de la culebra de escalera (*Rhinechis scalaris*) en los alrededores de torres eléctricas: Resultados de radioseguimiento

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La culebra de escalera (*Rhinechis scalaris*) es una culebra mediterránea con una amplia distribución en la Península Ibérica. Su comportamiento reservado ha imposibilitado el profundo conocimiento de sus patrones ecológicos, estudiados básicamente por radioseguimiento en épocas de máxima actividad (e.g. primavera). Desde 1998, Iberdrola ha recogido datos de incidencias eléctricas dentro de su red en las líneas de media tensión (MVPT) directamente provocadas por la culebra de escalera. La mayoría de las incidencias están localizadas en dos áreas concretas de España (centro y este) durante primavera y otoño. Experimentos desarrollados en cautividad señalaron que las culebras se sentían atraídas a los MVPTs debido a estímulos olfativos procedentes de sus presas. El uso del espacio y sus tasas de movimiento en relación con las zonas de MVPTs puede aportar una valiosa información para el diseño de estrategias que impidan las electrocuciones además de conocer más profundamente los patrones ecológicos de la especie.

Analizamos los patrones espaciales y temporales de la culebra de escalera mediante radioseguimiento de cuatro especímenes (dos machos, dos hembras), que ocupaban un área de alta densidad de incidencias (Castellón, España). Los análisis combinaron sistemas de información geográfica con regresiones lineales para analizar 990 localizaciones obtenidas a lo largo de un ciclo anual de actividad (2012-2013). Los micro hábitats seleccionados por los cuatro especímenes eran principalmente formados por vegetación herbácea en los alrededores de muros de piedra. Además cada individuo usó diferentes micro hábitats en los diferentes periodos del año (log-lineal; $p=0.98$). Las culebras mostraron un comportamiento muy reservado con un periodo anual de actividad de febrero a noviembre y un periodo de total inactividad de diciembre a enero, siendo el periodo de mayor actividad de marzo a abril. La media de movimiento diario (10.85 ± 3.80 m) y el área cubierta por los individuos (2.247 ± 1.088 ha) fue substancialmente diferente a la información previamente mostrada para la especie en otras regiones de la Península Ibérica. La localizaciones más cercanas de las culebras a las MVPTs (± 50 m) ocurrieron en otoño, coincidiendo con uno de los dos grandes picos de incidencias recogidas para la especie por Iberdrola. Ninguna culebra fue observada trepando a las torres cercanas. Nuestros resultados, a pesar del bajo número de individuos estudiados, sugieren un uso esporádico de estas infraestructuras por las culebras.

Spatial and temporal patterns of the ladder snake (*Rhinechis scalaris*) in electrical towers surroundings: results from a radio-telemetry monitoring

The ladder snake (*Rhinechis scalaris*) is a Mediterranean snake, widely distributed in the Iberian Peninsula. Its secretive behaviour hampered a good knowledge of population ecological patterns, inferred using radio-telemetry monitoring but for limited periods over the annual cycle of activity (e. g. in spring). Since 1998, Iberdrola has been recording electrical power outages directly produced by the ladder snake in medium voltage power towers (MVPT) within its network. Fatality records were mostly located in two areas within Spain (Central and Eastern), during spring and autumn. Experiments developed in captivity pinpointed that snakes were attracted to MVPTs by prey scents. Information on space use and activity rates of snakes in relation to areas with MVPTs could give valuable insights into management strategies for avoiding snakes' fatalities, as well as, to increase the knowledge on species ecological patterns.

Here we present findings on spatial and temporal patterns of the ladder snake obtained through radio-tracking monitoring of four specimens (two males, two females) inhabiting an area with high occurrence of fatalities (Castellón, Spain). Analyses combined Geographical Information Systems and linear regressions over 990 records acquired throughout an annual cycle of activity (2012–2013). Selected micro-habitats for the four individuals were mostly composed by herbaceous vegetation in the surroundings of stone walls, but each individual used different micro-habitats according to the period of year (log-lineal; $p=0.98$). Snakes showed a highly secretive behaviour with a period of activity from February to November and an inactivity period from December to January, being the period with higher activity in March and April. Average daily displacement (10.85 ± 3.80 m) and area covered by individuals (2.247 ± 1.088 ha) were substantially different to previous information reported for the species in other regions of Iberia. The closest snakes' locations to MVPTs (± 50 m) occurred in autumn which agree with one of the two major peaks of fatalities recorded for the snake by Iberdrola. However, no snakes were observed climbing on MVPTs. Our results, in spite of low number of individuals monitored, suggest a sporadic use of these infrastructures by snakes.

Snakes, telemetry, ecology, electrical outages, home range.

Toads on roads. Study of the effects of roads in amphibians

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Roads have multiple effects on wildlife, such as animal mortality, habitat and population fragmentation, and modification of animal reproductive behavior. Amphibians in particular, due to their activity patterns, population structure, and preferred habitats, are strongly affected by traffic intensity and road density. Studies on the effects of road-kills and associated conservation measures usually focus on highways, but amphibians also die massively on secondary roads, where conservation measures are not applied. Many countries (e.g. Portugal) do not have national programs for monitoring road-kills, a common practice in other European countries (e.g. UK, The Netherlands), but in order to implement effective conservation measures it is necessary to identify hotspots of road-kills. However, monitoring road-kills is expensive and time consuming, and depends mainly on volunteers. Therefore, cheap, easy to implement, and automatic methods for detecting road-kills over larger areas (broad monitoring) and extended in time (continuous monitoring) are necessary. We present here the preliminary results from a research project which aims to build a cheap and efficient system for detecting amphibian road-kills using computer-vision techniques from robotics. We propose two different solutions: 1) a Mobile Mapping System to detect automatically amphibian road-kills in roads, and 2) a Fixed Detection System to monitor automatically road-kills in a particular road section in the long term. The first methodology will detect and locate road-kills through the automatic classification of road surface images taken from a car with a digital camera, linked to a GPS. Road kill casualties will be detected automatically in the image through a classification algorithm developed specifically for this purpose. The second methodology will detect amphibians crossing a particular road point, and determine if they survive or not. Both Fixed and Mobile systems will use similar programs. The algorithm is trained with collected data. For now, we can present some results only about the Mobile Mapping System. We are performing different tests with different cameras, namely a lineal camera, used in different industrial solutions of quality control, and an outdoor Go-pro camera. Our results proved that we can detect different road-killed and live animals at a high spatial resolution while driving at reasonable speed. Both systems will provide the capacity to detect automatically the casualties of road-kills. With these data, it will be possible to analyse the distribution of road-kills and hotspots, to identify the main migration routes, to count the total number of amphibians crossing a road, to determine how many of those individuals are effectively road-killed, and to define where conservation measures should be implemented. All these objectives will be achieved more easily and with lower cost in funds, time, and personal resources.

Amphibians, roads, road-kills, mobile mapping system, fixed mapping system, Portugal.

Aljibes como trampas mortales para anfibios y reptiles en ambientes áridos

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Las regiones áridas se están viendo afectadas cada vez más por la pérdida y transformación de hábitats. La creciente red de vías de comunicación en el noroeste del Sahara facilita el uso de los hábitats adyacentes a los caminos. En aquellas regiones donde la ganadería es el recurso económico tradicional y principal, la población local está actualmente construyendo numerosos aljibes para abreviar al ganado, cubriendo de esta manera un área mayor para el pastoreo. Los aljibes pueden atraer a vertebrados desérticos y actuar como trampas mortales para especies con poblaciones ya escasas en estas áridas regiones. Este estudio es el primero en analizar el impacto de los aljibes como trampas mortales para los anfibios y reptiles en el Sáhara, mediante el muestreo de 823 aljibes en el suroeste de Marruecos para identificar y cuantificar las especies afectadas. Anfibios y/o reptiles se encontraron atrapados en el 25,2% de los aljibes. Se encontraron diferencias en la proporción de aljibes con anfibios y reptiles en relación a la edad de los aljibes (más frecuentes en aljibes antiguos), forma (más frecuentes en los aljibes rectangulares), tamaño (más frecuente en los aljibes grandes), regiones (más frecuentes en el Anti Atlas e Ifni) y hábitat (más frecuente en matorral macaronésico y menos en suelo pedregoso desnudo). Cuatro especies de anfibios y 35 de reptiles estaban atrapadas en los aljibes, siendo *Bufo boulengeri* la especie más común entre los anfibios y *Agama impalearis* la más común entre los reptiles. Algunas están catalogadas como amenazadas a escala nacional (*Dasypeltis sahelensis*, *Naja haje*). Al menos 459.017 individuos de anfibios y reptiles quedan atrapados anualmente en el área de estudio. La baja productividad y baja densidad de vertebrados terrestres en esta árida región sugieren que los aljibes tienen un impacto sustancial sobre los anfibios y reptiles. Como la construcción de aljibes se está incrementando, se precisan acciones de manejo para mitigar este impacto sobre la comunidad herpetológica.

Water cisterns as death traps for amphibians and reptiles in arid environments

Arid regions are increasingly being anthropogenically altered. In the north-western Sahara, a growing road network facilitates the use of habitats adjacent to roads. In regions where livestock is the traditional and main economic resource, local people are currently building numerous water cisterns for watering livestock, leading to an increase in the extent of pasturing of domestic livestock. Cisterns may attract desert vertebrates and act as death traps for species with already sparse populations in these arid areas. This study is the first to examine the impact of cisterns as lethal traps for amphibians and reptiles in the Sahara, using a survey of 823 cisterns in south-western Morocco to identify and quantify affected species. Differences were detected in the proportion of cisterns with amphibians and reptiles according to cistern age (more frequent in old cisterns), shape (more frequent in rectangular cisterns), size (more frequent in large cisterns), regions (more frequent in the Anti-Atlas and Ifni), and habitats (more frequent in non-Macaronesian scrubland and less frequent in bare stony soil). Four amphibian and 35 reptile species were trapped in cisterns, being *Bufo boulengeri* the more frequent among amphibians and *Agama impalearis* the more frequent among reptiles. Some are listed as threatened at the national scale of Morocco (*Dasypeltis sahelensis*, *Naja haje*). At least 459 017 individual amphibians and reptiles were trapped annually within the study area. The low productivity and low population densities of terrestrial vertebrates in this arid region suggest that cisterns have a substantial impact upon amphibian and reptile species. As cistern construction is increasing, management actions are required to mitigate this impact on the herpetological community.

Amphibians, conservation, Morocco, reptiles, Sahara, water cisterns.

Impacto de diferentes regimes de fogos em comunidades de répteis do Parque Nacional da Peneda-Gerês

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O fogo é um perturbação frequente em muitos ecossistemas do planeta com efeitos profundos na diversidade e abundância das comunidades. É a perturbação com maiores efeitos na Península Ibérica, geralmente dominados por ecossistemas propensos e adaptados a ela, resultado da sua história evolutiva associada ao fogo. No entanto, organismos resilientes ao fogo não são adaptados ao fogo por si, mas sim a um específico regime de fogos, sendo na verdade, as alterações na frequência e extensão dos fogos “a perturbação”. O fogo abre a estrutura do habitat favorecendo animais adaptados a estas condições. No entanto a ocorrência de fogos repetidos irá reter a vegetação nos seus estados primordiais, não permitindo o aparecimento de espécies que necessitam de uma estrutura madura da sucessão para se desenvolverem. Os répteis mostram uma grande ligação a este desenvolvimento da sucessão ecológica. Examinámos o efeito de diferentes padrões de fogos nas comunidades de répteis do Parque Nacional da Peneda Gerês, uma reserva natural com um longo historial de fogos. Sondamos os répteis em cinco locais que sofreram fogos em diferentes períodos e diferentes frequências e comparamos com a respectiva parte não ardida de cada uma. Cada uma foi visitada oito vezes, tendo sido observados 602 indivíduos de 10 espécies diferentes. No geral, transectos não ardidos obtiveram uma maior riqueza específica e mais baixa abundância.

The impact of repeated-fire regimes on the reptile community at the Peneda-Gerês National Park

Fire is a common disturbance in many ecosystems around the world with profound effects on diversity and composition of communities. Fire is the most important disturbance in Iberian landscapes, often dominated by fire-prone ecosystems resulting from a long evolutionary association with fire. Although, resilient organisms to fire are not adapted to fire per se, but rather to a particular fire regime, being the increasing in fire frequency and extension observed presently the true disturbances. In this scenario, conservation of biodiversity depends on understanding responses to fire in animal communities. Fire opens the habitat, what will benefit animals adapted to post-fire open terrain conditions. However, frequent repeated fires would retain the vegetation structure in early successional stages and animal species that depend on mature vegetation structure for provision of fit habitat will not develop in those conditions. Reptiles appear to show this strong response to habitat structure. We examined the effect of repeated-fire regimes on the reptile community at the Peneda-Gêres National Park, an area with a long history of fires. We surveyed reptiles in five sites affected by fires occurred in different periods and frequencies and compared with respective unburnt part in the edge of each one. Each transect was surveyed eight times, and we observed 602 reptile specimens from 10 species. In general, unburned transects had higher species richness and lower total abundance than their respective burned pairs. However, some transect pairs did not follow this general rule, suggesting than the fire history (number of fires and time since last fire) as well as the habitat structure previous to the last fire can also drive the response of communities to fire disturbances.

Disturbance, repeated-fire regimes, habitat structure, diversity, conservation.

Respuesta de la comunidad de reptiles a tratamientos de aclareo de repoblaciones forestales en Sierra Nevada (SE ibérico)

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La pérdida y transformación del hábitat suponen la mayor amenaza a la diversidad biológica mundial. Comprender la respuesta de los seres vivos a estas alteraciones producidas por el ser humano en los paisajes es esencial para la conservación de las especies. Las repoblaciones forestales realizadas en España hasta la segunda mitad del siglo XX se han caracterizado por el empleo masivo de especies del género *Pinus*, en densidades de pies arbóreos muy por encima de las que se desarrollan en bosques naturales. Estas plantaciones densas, coetáneas y monoespecíficas, generan una serie de problemas en los sistemas naturales, como la disminución de la diversidad vegetal y animal, aumento de la incidencia de plagas, deterioro de las características edáficas, y aumento de la frecuencia e intensidad de incendios forestales. Identificadas estas afecciones derivadas de las plantaciones de coníferas, la gestión forestal se ha visto obligada a realizar medidas silviculturales para reducir la densidad de pies por hectárea, lo que se conoce como aclareo forestal. Hemos aprovechado tareas de aclareo forestal realizadas en pinares de repoblación de Sierra Nevada, y se ha analizado, por primera vez en la Península Ibérica, la respuesta de la comunidad de reptiles a esta gestión forestal. Se ha analizado la riqueza, abundancia y diversidad ecológica de la comunidad de reptiles en cuatro grados de aclareo, con cuatro réplicas por tratamiento, y cuatro muestreos por réplica. Siendo los reptiles organismos ectotermos, nuestra hipótesis ha sido que estos parámetros de la comunidad aumentarán con la reducción de la densidad de pinos, debido al acceso de la luz hasta el sustrato, donde viven la mayoría de los reptiles Mediterráneos.

Los resultados confirman una respuesta positiva (mayor riqueza, abundancia y diversidad ecológica) en aquellas masas de coníferas aclaradas, con una mayor respuesta a mayor grado de aclareo. Se pretende que los resultados obtenidos en este estudio sirvan de modelo en gestión forestal, aportando una razón más para sostener la gestión, mediante aclareo, de las antiguas repoblaciones forestales con coníferas en el ámbito Mediterráneo.

Reptile community response to thinning in plantations of Sierra Nevada (SE Iberian Peninsula)

Habitat loss and degradation is the greatest threat to global biodiversity. Understanding the response of living organisms to human-induced landscape changes is crucial for the conservation of species. Plantations, carried out in Spain to the second half XX Century, have been characterized by the use of *Pinus* species in tree densities above values observed in natural forests. These dense, even-aged and monospecific plantations generate a number of problems in natural systems such as a loss in plant and animal diversity, increased incidence of pests, deterioration of soil characteristics, and increased frequency and intensity of wildfires. Once recognized the problems generated by pine plantations, Spanish forestry managers performed silvicultural measures to reduce the density of trees per hectare, which is known as forest thinning. We conducted a pioneer study in the Iberian Peninsula aimed at studying the response of the reptile community to different levels of pine forest thinning in Sierra Nevada. We analyzed species richness, abundance and diversity of the reptile community to four levels of thinning, with four replicates per treatment, and four visits per replicate. Being reptiles ectotherm organisms, our hypothesis was that the parameters of the community would increase in parallel to a tree density reduction, because of more light reach the soil, where most of Mediterranean reptiles dwell.

Our results confirmed a positive response (greater species richness, abundance and ecological diversity) was registered in clearer conifer plots, with a greater response to higher thinning management. It is intended that the results of this study serve as a model for forest management, providing one more reason to support management by the thinning of old conifer afforestation in the Mediterranean region.

Thinning management, reptiles, community, species richness, density, ecological diversity.

Cambio climático y tamaño corporal en ofidios del sureste ibérico

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Se denomina Cambio Climático a la variación global del clima de la Tierra, debido a causas tanto naturales como antropogénicas producidas en muy diversas escalas de tiempo. Desde hace unos años hasta la actualidad, debido a actividades humanas, se viene observando un intenso y acelerado cambio del cual derivan una serie de impactos medioambientales como es el calentamiento global. El estudio de sus efectos sobre los seres vivos constituye un campo de investigación en emergencia. Este trabajo se centra en uno de los efectos quizá menos perceptibles a corto plazo: la variación del tamaño corporal de los seres vivos con respecto al tiempo. Este fenómeno se ha demostrado en diversas especies de endotermos, sin embargo, existe un mayor desconocimiento en ectotermos. En este estudio se analizan tres especies de ofidios (*Rhinechis scalaris*, *Hemorrhois hippocrepis* y *Malpolon monspessulanus*) del sureste ibérico que podrían estar respondiendo al calentamiento global modificando su medida corporal en un período de 35 años (1980-2014). Debido a diferencias filogenéticas (origen evolutivo y tipo de dieta), se podría esperar distintas respuestas entre las especies de estudio. Se analizó la tendencia, a nivel temporal, de variación del tamaño corporal. Para ello se utilizó como medida morfométrica, la longitud hocico-cloaca (LHC) de ejemplares preservados en la colección del departamento de Zoología de la Universidad de Granada (unos 2000 individuos durante el período de estudio). Adicionalmente, se hizo un análisis de la evolución temporal en el cociente juveniles/adultos y en la razón de sexos (hembras/machos), como índices informativos sobre el estado de conservación de estas poblaciones. Los resultados rechazaron la existencia de un cambio en el tamaño corporal de las mismas. Tampoco fue hallado cambio para el cociente juveniles/adultos ni la razón de sexos. Por tanto, se concluye que los ofidios analizados no están respondiendo fenotípicamente al cambio climático mediante una variación en su tamaño corporal. Este trabajo pone en evidencia que grupos animales ectotermos como son los ofidios (reptiles), no han sufrido cambio en sus medidas corporales durante las últimas décadas como consecuencia del calentamiento global. No obstante, las especies analizadas son las más comunes y menos amenazadas de la Península Ibérica, por ello se recomienda que este estudio se lleve a cabo con otras especies más vulnerables frente a cambios ambientales.

Climate change and body size in snakes from the SE Iberian Peninsula

Climate change is defined as the variation of the Earth's climate in a global scale, due to natural or anthropic causes. Recent human activities have led to an intense and accelerated climate change in our planet. In this scenario, the study of the effects of climate change on organisms is an emergent research field. Here, we focus on one of the responses which have received less attention: the variation of the body size. This fact has been proved in some endothermic species, however there is a great lack of knowledge on the response of ectothermic ones. We aim to test if three snake species (*Rhinechis scalaris*, *Hemorrhois hippocrepis* and *Malpolon monspessulanus*) from the south-eastern Iberian Peninsula could be reacting against climate change by varying their body size, during a 35 years period (1980-2014). We expect different responses among the species under study, as a result of phylogenetic differences (evolutionary origin and diet). It was analyzed the temporal trend in variation of body size. As morphometric measure, was used the snout-to-vent-length of the vouchers preserved in the scientific collection of the Zoology Department of Granada University (2000 individuals for study period). Besides it was analyzed the evolution in the juvenile/adult and female/male ratios, as indexes about the conservation status of the studied populations. Our results rejected any temporal trend in the body size of the studied species. Any change in the juvenile/adult neither in the female/male ratios was found. We conclude that the analyzed snakes do not respond phenotypically to global warming by varying body size in the study area. This paper shows how a terrestrial ectothermic group (ophidians, reptiles), have not changed the body size as a response to global warming in the last decades. The studied species are the most common within the Iberian snake community and consequently the least threatened; therefore, this study could be repeated with other species more vulnerable to environmental changes.

Climate change, body size, snakes.

Amphibian conservation in Mediterranean farmland landscape: the role of temporary ponds

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In a farmland landscape, non-productive interstitial landscape elements, such as hedgerows, windbreaks, grassy margins or vernal ponds, play vital role in enhancing the biodiversity, serving as refuge, corridors, feeding grounds, etc. With the intensification process these elements tend to be replaced by others more production-related. As such, important breeding habitats like temporary ponds are usually regarded as a nuisance by land owners and are being converted to farm ponds (permanent) or to arable land. Can Mediterranean amphibian species offset this destruction by occupying these and other artificial habitats? Or will these habitats only provide conditions to few wide spread generalist species?

We assessed these alternative views by documenting the decline of temporary ponds in a Mediterranean farmland landscape (Southwest Portugal) between 1991 and 2009, and by estimating pond survival probabilities using a Cox proportional hazards regression model. To model the occupancy of natural (temporary ponds and streams) and artificial water bodies (farm ponds, irrigation channels and drainage ditches) by amphibians across a breeding season (February-June 2010) we developed a hierarchical Bayesian dynamic multi-species occupancy model that allowed us to control for differences in detectability across species, sampling occasions and habitat types.

This landscape lost 56% of its temporary ponds in less than two decades. Mainly the temporary ponds were lost due to agricultural-related activities such as cultivation, conversion to permanent farm ponds or drainage (89.3%). Ponds situated inside an irrigation perimeter had the lowest survival rate and the natural park did not provide a positive effect in the survival probability of temporary ponds. Temporary ponds had an estimated species richness at least twice as high as other habitats. Seven of the 10 species recorded had the highest occupancy probability in temporary ponds and never, or rarely, occurred in artificial habitats. Only one species was widely spread in the permanent habitats (farm ponds and irrigation channels).

These results suggest that artificial habitats are unlikely to provide adequate breeding habitat for most species in a Mediterranean intensive farmland. In such landscapes amphibian conservation should be focused in the conservation of remaining temporary ponds, and at restoring or creating new temporary ponds where these have been lost.

Amphibians, artificial habitats, farmland, occupancy models, temporary ponds, imperfect detectability.

Distribución actual, estima poblacional y estado de conservación de la rana pirenaica, un endemismo en peligro crítico de extinción

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La rana pirenaica (*Rana pyrenaica*) es una especie de rana parda endémica del Pirineo, que se distribuye por parte de la cordillera Pirenaica en Navarra y Aragón (España) y en una pequeña zona en la vertiente francesa. Actualmente está catalogada como En Peligro de Extinción por la UICN, principalmente por ocupar un área de distribución muy pequeña y altamente fragmentado. Hasta el presente momento no existen datos acerca del estado de las poblaciones y las actuales amenazas para la especie. En este trabajo presentamos los resultados de un programa de monitoreo de cuatro años. Entre los años de 2010 y 2013, re-visitamos prácticamente todas las localidades conocidas de *R. pyrenaica*, así como otras localidades potenciales, identificadas mediante un modelo de distribución potencial. Cada localidad fue revisitada muchas veces a fin de confirmar la presencia o ausencia de la especie, apuntando los tipos de microhábitat, y llevando a cabo una estima del número de individuos reproductores. Nuestros resultados muestran que la mayoría de las poblaciones son pequeñas y muy fragmentadas. Los modelos de conectividad bajo diferentes escenarios sugieren una baja conexión entre las principales zonas de distribución, principalmente debido a las barreras naturales (ej. montañas) y las inducidas por el hombre (ej. introducción de peces en las cabeceras de los ríos). La mortalidad de los renacuajos en las poblaciones controladas está influida por los eventos climáticos extremos. Un análisis molecular de muestras de la piel y boca de 666 adultos y renacuajos de 108 localidades ha detectado la presencia del hongo quítrido (Bd) en la mayor parte de las poblaciones. Los quítridos fueron detectados en 48 localidades dentro del rango de distribución de la especie, con 184 individuos detectados como Bd positivo (27.6%). Nuestros datos sugieren que la presencia del Bd en esta especie es elevada, con la mayoría de las poblaciones afectadas, aunque no sabemos si se producen eventos de mortalidad masiva. Esta situación de poblaciones con baja densidad, un alto grado de fragmentación del hábitat, cambios climáticos y la presencia de enfermedades infecciosas como el Bd, sugieren que esta especie merece catalogarse como En Peligro Crítico de Extinción y necesita un seguimiento continuado y medidas de conservación *in situ*, quizás también *ex-situ*, para asegurar su supervivencia.

Current distribution, population estimates and conservation threats of the critically endangered Pyrenean frog

The Pyrenean frog (*Rana pyrenaica*) is an endemic brown frog species of the Pyrenean Mountains, mainly distributed in Navarra and Aragón (Spain) with a few populations in France. It is currently categorized as Endangered by the IUCN, mainly because it occurs in a small range with highly fragmented habitat. There are no current data on the population status, potential population trends and current threats for the species. Here, we report the results of a four year monitoring program on this species. Between 2010 and 2013, we re-visited almost all known localities of *R. pyrenaica*, as well as other potential sites identified by potential distribution modeling. Each locality was visited several times to confirm absence or presence of the species, record microhabitat types, as well as perform a crude estimate of the number of breeding individuals. Our results show that most populations are very small and very fragmented. Connectivity models under different scenarios suggest low connectivity between main distribution areas, mainly by natural barriers (e.g. mountains) and human induced ones (e.g. introduction of fish in headwaters). Mortality of tadpoles in some controlled populations is influenced by extreme climatic events. A molecular analysis of skin and mouth samples from 666 adults and tadpoles from 108 localities detected the presence of the chytrid fungus (Bd) in most populations. The chytrid was detected in 46 localities across the species range, with 184 individuals testing Bd positive (27.6%). Our data suggest that the presence of Bd in this species is high, with most populations affected, although we do not know if it is producing massive mortalities or not. The current situation with low population densities, high habitat fragmentation, climate change and the presence of infectious diseases such as Bd, suggest that this species may deserve the category of Critically Endangered and needs a continuous monitoring and *in situ* conservation measures, maybe also *ex situ* too, to ensure its persistence.

Conservation, endangered species, connectivity, global change, chytridiomycosis, fragmented area.

Recuperación de un anfibio norteafricano en peligro de extinción: el proyecto de la salamandra de los Beni Snassen

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La salamandra de los Beni Snassen *Salamandra algira spelaea* es uno de los anfibios con una distribución más restringida del norte de África. Su presencia está limitada a una zona muy confinada del macizo de los Beni Snassen, en el nordeste de Marruecos. Este macizo montañoso muestra unas condiciones climáticas subhúmedas a semiáridas que pueden considerarse límites para una especie métrica de anfibio. Este factor, junto a la degradación del hábitat, por el uso agrícola del suelo y por el sobrepastoreo, así como la construcción de canteras y la canalización de las fuentes puede desencadenar la extinción definitiva de esta población única. En el presente trabajo se exponen las directrices del plan de recuperación de esta subespecie. En la primavera del 2014 se han construido tres balsas que pueden favorecer las poblaciones de esta especie en la periferia de su distribución. Estas balsas tienen unas dimensiones 2 m x 1,5 m y una profundidad máxima de 40-50 cm., similares a las balsas utilizadas de forma natural por la especie. La segunda parte del proyecto consiste en la captura de larvas y su mantenimiento en cautividad, con el objetivo de generar un futuro stock reproductivo en cautividad, que puede prevenir la extinción de la especie en el caso de un colapso de las poblaciones salvajes o servir para iniciar futuras repoblaciones en zonas que muestren las condiciones ambientales adecuadas para ello. Ambas medidas siguen las recomendaciones de manejo intensivo que se han propuesto desde la UICN para especies de anfibios críticamente amenazadas.

Recovering a North African endangered amphibian: guidelines for the Beni Snassen's fire salamander

The Beni Snassen's Salamander *Salamandra algira spelaea* is one of the amphibians with a more restricted distribution in North Africa. Its presence is confined to a very small area in the Beni Snassen massif in northeastern Morocco. This massif shows sub-humid to semi-arid climatic conditions that are possibly close to the eco-physiological limits of mesic amphibians. This factor, together with habitat degradation, related to the impacts of agriculture and overgrazing, quarrying, and canalization of water sources can trigger the extinction of this unique population. In the present work guidelines for the recovery plan for this subspecies are presented. First in the spring of 2014 three new ponds were built that can favor the recovery of some marginal populations of this species. These artificial ponds have dimensions of 2 m x 1.5 m and a maximum depth of 40-50 cm, similar to the natural ponds used by the species. The second part of the project involves the capture of larvae and their maintenance in captivity, with the aim of generating a future captive breeding stock, which can prevent the extinction of the species in the possible event of a collapse of wild populations or serve to repopulate areas with suitable environmental conditions. Both measures follow the intensive management recommendations proposed by the IUCN for critically endangered amphibian species.

Conservation, *Salamandra algira spelaea*, Béni Snassen, North Africa.

Once años de estudios intensivos para la conservación de la población natural de tortuga mediterránea *Testudo hermanni hermanni* en la Sierra de la Albera

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En la actualidad, la última población natural de tortuga mediterránea de toda la Península Ibérica se localiza en el extremo este de los Pirineos, en la sierra de la Albera, dentro del Espacio Natural protegido. Esta población se encuentra en estado crítico (1 a 2 individuos/ha) debido principalmente a actividades humanas. Con el objetivo de proteger, conservar y/o mejorar el hábitat de la tortuga mediterránea decidimos crear la Red de Custódia de la Albera (XCA). Mediante acuerdos de custodia entre nuestra entidad y propietarios podemos mejorar el estado de conservación de la población de la Albera. Los Planes Técnicos de Gestión de Custodia (herramienta técnica de gestión) que se redactan para cada finca adherida a la XCA permiten establecer y planificar medidas de gestión agroforestales favorables a la especie. En estos momentos ya son más de 350 ha en custodia donde se han realizado una serie de actuaciones. Entre estas actuaciones destacamos i) la creación de 60 zonas de nidificación mediante desbroces selectivos en los que se han detectado cuatro nidos dos años después de su creación, ii) la construcción de 50 majanos de colonización y cinco majanos experimentales de fundación para mejorar el estado de conservación del conejo como indicador biológico, iii) la recuperación de actividades tradicionales agropecuarias como el cultivo tradicional del viñedo y olivar o la recuperación de rebaños de cabra de la Albera (raza en peligro de extinción), y iv) la liberación de 388 ejemplares de tortuga mediterránea con una supervivencia anual del 80%.

Eleven years of intensive conservation studies on native population of the western Hermann's tortoise *Testudo hermanni hermanni* in Albera Range

At present, the last native population of Western Hermann's Tortoise in the Iberian Peninsula is located in north-west of Catalonia, within the Albera Natural Park. This tortoise population is critically endangered, with just 1 to 2 individuals/ha as a result of multiple extinction drivers. Aiming to protect, conserve and/or improve the Western Hermann's tortoise habitat we decide to create the Albera Stewardship Network (XCA). We then improve the state of conservation of Albera population through custody agreements between the group and landowners. The Technical Management Plans of Custody (technical tool of management), that are written per each land registered in the XCA, establish and plan agro forestry managements positive measures for tortoise conservation. Nowadays, 350 ha conformed the XCA where were conducted some actions. These actions included i) the creation of 60 nesting areas with selective brush clearance where four nests were detected after two years of their creation, ii) the building of 50 burrows of colonization and five experimental burrows of foundation to improve the state of conservation of the common rabbit as biological indicator, iii) recovery of farming traditional activities of vineyard and olive grove or the recovery of the herd of Albera goats (endangered race), and iv) the release of 388 tortoises with a survival rate of 80%.

Tortoise, management, conservation, land custody, habitat, custo-labelling.

Tartarugas marinhas em águas ibéricas: registos de Portugal e da Galiza

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Vários estudos têm tornado evidente a importância de alguns habitats e locais específicos de alimentação na Península Ibérica tanto para a tartaruga-comum das populações do Mediterrâneo e do Atlântico norte, como para a tartaruga-de-couro da população do Atlântico oeste. O presente estudo avalia os padrões espaciais e temporais registados para as tartarugas marinhas em Portugal Continental e na Galiza, entre 1990 e 2013, obtidos a partir de observações, arrojamentos, e registos de captura acidental/resgates. Neste período, foram registadas 1239 tartarugas marinhas pelas redes de arrojamentos portuguesa e da Galiza. Os registos incluem tartarugas-comuns (54%), tartarugas-de-couro (44.4%), tartarugas-verdes (1.4%), tartarugas-de-Kemp (0.16%) e tartaruga-de-escamas (0.04%). As tartarugas comuns e as de couro foram as mais frequentemente observadas, estando presentes em toda a costa Ibérica ao longo de todo o ano, com padrões espaciais e temporais diferentes para cada espécie. A tartaruga-comum é frequentemente mais observada a latitudes mais baixas (na zona sul de Portugal), enquanto as tartarugas de couro são frequentemente mais detetadas a norte, nas costas ocidental portuguesa e galega. Na península Ibérica, as pescas representam uma potencial ameaça à conservação das tartarugas marinhas uma vez que 12.2% das tartarugas-comuns registadas e 16.4% das tartarugas-de-couro foram capturadas acidentalmente. A maré negra provocada pelo derrame do “Prestige” foi uma outra ameaça antropogénica relevante, tendo sido responsável pelo petroleamento de 37.7% das tartarugas comuns registadas na Galiza. Nos seis meses posteriores ao derrame, 100 tartarugas petroleadas foram registadas, possivelmente constituindo o primeiro registo de arrojamento massivo de tartarugas marinhas na Europa. Uma vez que as tartarugas marinhas são migradores de longa distância, os dados recolhidos em várias zonas da sua rota migratória são necessários para avaliar as ameaças e os impactos nas suas populações a um nível global. A integração dos registos de Portugal e da Galiza permite estimar padrões espaciais e sazonais das tartarugas marinhas, e avaliar as causas de morte mais importantes ao longo de uma área geográfica mais alargada. Por sua vez, em conjunto com dados ecológicos destas várias áreas, estes registos servirão de base para a aplicação de medidas de conservação para as tartarugas marinhas na costa Atlântica da Península Ibérica.

Sea turtles in Iberian waters: records from continental Portugal and Galicia

Several studies have shown the importance of specific habitats and foraging grounds in the Iberian Peninsula for loggerheads from the Mediterranean and North Atlantic populations and for leatherbacks from the Western Atlantic population. The present study assesses spatial and temporal trends of sea turtle records in Continental Portugal and Galicia between 1990 and 2013 including sightings, strandings and incidental capture/rescues. Between 1990 and 2013, 1239 sea turtles were recorded by the Portuguese and Galician stranding networks. Records include loggerheads (54%), leatherbacks (44.4%), green turtles (1.4%), Kemp’s ridley (0.16%) and hawksbill turtles (0.04%). Loggerheads and leatherbacks were the most frequently sighted species, being present off the Iberian coast throughout the year, showing clear spatial and seasonal differences. Loggerheads were more frequently recorded at lower latitudes (southern Portugal), while leatherbacks were more frequently recorded towards the North of the peninsula (Galicia and West Portugal). In the Iberian Peninsula, fisheries may represent an important anthropogenic threat to sea turtle conservation since 12.2% of the recovered leatherbacks and 16.4% of loggerheads were victims of incidental capture. As a result of another major anthropogenic threat, in Galicia 37.7% of the recorded loggerheads were victims of the Prestige oil spill and 100 oiled stranded turtles were recorded in the 6-month period after the spill, possibly constituting the first massive sea turtle stranding event in Europe. Since sea turtles are long-range migratory animals, wide-scale data are necessary to evaluate the global threats and impacts on their populations. Combining data from Portugal and Galicia provided results on sea turtle occurrence, spatial and seasonal patterns and it also provided insights on causes of death while covering a wider area. The integration of the present results with local ecological knowledge is the cornerstone for future sea turtle conservation in the area.

Iberian Peninsula, sea turtles, strandings.

Proyecto SOS GALÁPAGOS, más de 10 años de estudio sobre galápagos acuáticos en el País Vasco

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El proyecto SOS GALÁPAGOS se puso en marcha en el año 2001. Inicialmente, fue un proyecto pionero de extracción de ejemplares exóticos en el humedal de Bolue, localizado en Getxo (Bizkaia), que posteriormente se ha extendido a los Territorios Históricos de Araba y Bizkaia. Los objetivos de este proyecto giran en torno a tres ejes fundamentales: a) controlar la población de galápagos exóticos, reduciendo su tamaño a una dimensión, lo menor posible, que pueda ser asimilable por el medio; b) conocer y evaluar el estado de las poblaciones de galápagos autóctonos y c) desarrollar una campaña de concienciación sobre la problemática de las sueltas de especies exóticas en un medio natural. De esta manera, durante estos años se han extraído cerca de un millar de ejemplares exóticos de las diferentes zonas húmedas y se han obtenido datos sobre su capacidad invasora en el área de estudio: aspectos reproductores, latencia invernal, zonas de suelta y problemática, experimentación con las diferentes técnicas de captura y vigilancia, respuestas a los agentes meteorológicos propios de la cornisa cantábrica, etc. Respecto a los taxones autóctonos, *Emys orbicularis* y *Mauremys leprosa*, se han encontrado nuevas poblaciones y se han actualizado los mapas de distribución de las mismas por el área de estudio. Además, se está llevando a cabo el seguimiento de algunas de estas poblaciones mediante metodologías diversas, como: radioseguimiento o captura-marcaje-recaptura, permitiéndonos obtener conclusiones interesantes e inéditas respecto a la propia biología de estas especies en el área de estudio. El proyecto SOS GALÁPAGOS, en principio, sigue adelante y nos va permitiendo estrechar el círculo en torno al conocimiento de los galápagos en el País Vasco.

The SOS Galápagos project, more than 10 years of study of pond turtles in the Basque Country

The SOS GALÁPAGOS project was set up in 2001. It was a pioneering project aiming at extracting exotic terrapins from the Bolue wetland, located in Getxo (Bizkaia), but was later extended to embrace the whole area of Araba and Bizkaia. The objectives of this project focus on three basic axes: a) to control the populations of exotic terrapins, reducing their size to a minimum dimension where the environment can cope with these species; b) to know and assess the status of the native terrapin populations; and c) to develop a public communication campaign on the risks of releasing exotic species into the wild. In this way, during all these years, we have extracted around a thousand individuals of exotic terrapins from different wetlands and gathered useful data on their invasive potential in the study area: reproduction biology, winter dormancy, main release areas and associated problems, trial of different capture and surveillance techniques, response to climatic variables, etc. With respect to native taxa, *Emys orbicularis* and *Mauremys leprosa*, we have found new populations and have hence updated their distribution maps for the study areas. Additionally, we are monitoring a series of populations by a diverse array of methodologies that include radiotracking and capture-mark-recapture, allowing us to extract important conclusions, some of which reveal new aspects of the biology of these species in the study area. The SOS GALÁPAGOS project is still ongoing and improving our knowledge of the biology of pond turtles in the Basque Country.

SOS GALAPAGOS project, *Emys orbicularis*, *Mauremys leprosa*, exotic terrapins, Basque Country.

Conservación y manejo de la comunidad de anfibios de La Alhambra y el Generalife, Granada

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El conjunto monumental de la Alhambra y Generalife alberga la población reproductora más importante de *Bufo spinosus* del área metropolitana de Granada, además de *Pelophylax perezi*. En este entorno se han contabilizado 22 hábitats para anfibios (17 albercas y 5 tramos de acequias), en 16 de los cuales ha sido confirmada la reproducción de al menos una especie de anfibio. La consulta de textos históricos, encuestas a trabajadores y prospección de campo, indican además la presencia histórica en este entorno monumental y actual en el próximo valle del río Darro de *Pleurodeles waltl* y *Alytes dickhilleni*, por lo que se han iniciado actuaciones de reintroducción de estas dos especies en la Alhambra y Generalife a partir de larvas de las poblaciones más próximas, dentro de la misma cuenca hidrográfica. Además, durante el periodo 2013-2014, se han realizado mejoras en los hábitats. Se han controlado (nueve puntos) o erradicado (ocho puntos) los elementos ícticos ornamentales, en el primer caso hasta densidades compatibles con la reproducción de los anfibios. Completan estos manejos la instalación de rampas de acceso y filtros en sumideros para evitar vaciado de larvas, la construcción de hábitats subacuáticos en 9 puntos, tres plataformas flotantes, una charca terrera, y la instalación de 160 refugios terrestres para postmetamórficos. Como resultado, durante el primer año, se ha visto incrementada la reproducción de anfibios en 10 de los puntos; los albercones del Generalife han pasado de no albergar larvas a tener más de 5.000 postmetamórficos de *B. spinosus*; las larvas de *P. waltl* sobreviven; y vuelven a observarse postmetamórficos de *A. dickhilleni* en este entorno monumental. Se ha obtenido un incremento paralelo de la biodiversidad de otras comunidades de seres vivos, como los macroinvertebrados acuáticos y aves, en especial con la retirada de los peces y un aumento natural de los macrófitos acuáticos.

Conservation and management of the amphibian community in the Alhambra and Generalife, Granada

The monumental ensemble of the Alhambra and the Generalife houses the most important *Bufo spinosus* population in the Granada metropolitan area, in addition to *Pelophylax perezi*. In this environment we have recorded 22 habitats for amphibians (17 pools and 5 irrigation ditches), in 16 of which at least one amphibian species reproduces. The consultation of historical references, surveys for workers of the monument, and field searches, further indicate the former presence within this monument and the current presence in the next Darro valley of *Pleurodeles waltl* and *Alytes dickhilleni*, so actions have been initiated to reintroduce these two species in the Alhambra and Generalife from the closest populations in the same watershed. Furthermore, during the period 2013-2014, amphibian breeding habitats within the monument were managed. We controlled (9 points) or eradicated (8 points) ornamental fish, in the first case to densities allowing amphibian reproduction, installed ramps in pools and filters in sinks to prevent larvae loss during draining, built underwater habitats in 9 points, floating platforms and a new pond, and installed 160 artificial terrestrial refugia for postmetamorphics. As a result, during the first year, the number of breeding amphibians increased in 10 sites; the Generalife Albercones, where no reproduction was previously observed, now host more than 5,000 postmetamorphic *B. spinosus*; reintroduced *P. waltl* larvae survive; and postmetamorphic *A. dickhilleni* have been observed again in this monumental environment. In parallel to the removal of fishes and the natural development of aquatic macrophytes, an increase in the biodiversity of other communities, such as aquatic macroinvertebrates and birds, was also observed.

Amphibians, terrestrial refugia, Alhambra, conservation actions, management.

Monitorização de anfíbios no Parque Natural da Serra de S. Mamede: contributo de novos resultados para o conhecimento do estado de conservação das populações

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A elaboração de estudos a longo prazo para a aquisição de informação sobre a evolução da distribuição das populações é ainda pouco comum, apesar destes projetos serem essenciais para a distinção do que podem ser declínios de populações ou flutuações naturais. A obtenção deste conhecimento condiciona a identificação e a aplicação de estratégias de gestão e conservação que devem estar ajustadas aos cenários de alterações ambientais cada vez mais rápidas e complexas que ocorrem, particularmente, na região mediterrânica. Nesta comunicação revêm-se os padrões de distribuição de anfíbios, na área do PN Serra de S. Mamede, obtidos nos últimos 30 anos e comparam-se os resultados preliminares de uma nova série de amostragens iniciadas em 2014. Pretende-se, deste modo, realizar uma primeira abordagem ao estado atual das populações, de forma a analisar se a tendência para a regressão e deslocação de populações, identificada em 2005, se mantém. Embora preliminares, os resultados deste estudo suportam a hipótese de um cenário crítico de declínio nas populações de anfíbios. À semelhança do observado no período de 2004-2005, as espécies de anfíbios, normalmente associadas a áreas com menor altitude, parecem sofrer uma regressão mais acentuada. Por outro lado, o registo daquelas espécies em locais cada vez mais próximos do cume da serra de S. Mamede (1025 m) indicia o movimento de populações para zonas com maior altitude, como resposta às condições mais adversas da penepianície. Estes resultados reforçam a necessidade de um processo dinâmico de gestão e de conservação com base em dados de distribuição das espécies atualizados.

Monitoring amphibians in PN Serra de S. Mamede: contribution of new results to the conservation status of populations

The development of long-term studies to acquire information on the evolution of the distribution of populations is still scarce, although these types of projects are essential to distinguish between what can be population declines or natural fluctuations. Obtaining this knowledge affects the identification and implementation of management and conservation strategies that should be adjusted to the scenarios of rapid and complex environmental changes occurring in the Mediterranean region. This communication reviews the distribution patterns of amphibians in the PN Serra de S. Mamede obtained in the last 30 years and compares the preliminary results of a new sampling series initiated in 2014. In this way, it is intended to perform a first approach to the current population status in order to examine whether the trend towards population regression and movements, identified in 2005, is still ongoing. Although preliminary, the results of this study support the hypothesis of a critical scenario of decline in amphibian populations. As observed in 2004-2005, amphibian species associated to lower lands seem to suffer a more marked decline. Moreover, the occurrence of these species in areas increasingly closer to the Serra de S. Mamede summit (1025 m) reveals the movement of populations to higher altitudes, in response to adverse conditions in lowlands. These results highlight the need for dynamic management and conservation processes based on up to date species distribution data.

Amphibians, conservation, monitoring, population declines, Mediterranean region.



POSTERS

(P01) Relaciones filogenéticas entre las especies de *Daboia* del norte de África

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Se ha señalado que distintos eventos paleogeográficos y climáticos ocurridos a partir del Mioceno han afectado los patrones de distribución y la estructura genética de la biota de la región mediterránea. Muchos estudios se han centrado en la comprensión de los procesos evolutivos en las especies europeas. Sin embargo, las especies del norte de África continúan poco estudiadas. El norte de África alberga dos especies de grandes víboras estrechamente relacionadas (recientemente elevadas a especie en base a rasgos morfológicos e inmunológicos) incluidas dentro del género *Daboia*: *D. mauritanica* (Duméril y Bibron, 1848) y *D. deserti* (Anderson, 1892). Aparentemente, muestran distribuciones parapátricas, con *D. mauritanica* apareciendo a lo largo de las zonas subhúmedas y semiáridas de la región norte del Magreb (incluyendo Marruecos, el Sáhara Occidental y el norte de Argelia y Túnez), y *D. deserti* restringida a las zonas semi-áridas del este de Argelia, el centro de Túnez y el noroeste de Libia. En el sur de Marruecos se han registrado ejemplares pálidos de *D. mauritanica*, similares a *D. deserti*, y recientes estudios filogenéticos basados en unos pocos ejemplares han señalado una baja diferenciación genética entre las dos especies. En este trabajo, se pretende obtener información sobre la historia evolutiva de las *Daboia* del norte de África mediante la evaluación de sus relaciones filogenéticas en base a dos marcadores de ADN mitocondrial (cytb y ND4) secuenciados para 40 ejemplares. El árbol filogenético y la red de haplotipos muestran altos niveles de diversidad genética y estructuración en la región del Magreb occidental, diferenciando tres clados en Marruecos: (1) Rif, y Medio y Alto Atlas, (2) Montañas del Anti-Atlas, y (3) montañas del norte del valle del Draa. Un cuarto clado incluye especímenes procedentes del este de Marruecos, Argelia, Túnez y Libia. La invalidez de *D. deserti* como especie queda pues reconocida. Procesos de contracción del rango de distribución en varias áreas del oeste de Marruecos y expansión poblacional durante las oscilaciones climáticas del Pleistoceno podrían explicar estos patrones filogenéticos. Futuros estudios usando datos paleoclimáticos deben ser dirigidos a testar estas hipótesis.

Phylogenetic relationships within North African *Daboia* species

Palaeogeographical and climatic events occurred since the Miocene are suggested to affect the distribution patterns and genetic structure of biota in the Mediterranean region. Many studies have focused on the understanding of evolutionary processes in European species, yet North African species remain largely unstudied. North Africa harbors two closely related species (recently elevated to species on the basis of morphological and immunological traits) included within the genus *Daboia*: *D. mauritanica* (Duméril & Bibron, 1848) and *D. deserti* (Anderson, 1892). They apparently show parapatric distributions, with *D. mauritanica* occurring along the sub-humid and semi-arid zones of northern Maghreb region (including Morocco, Western Sahara, and northern Algeria and Tunisia), and *D. deserti* restricted to the semi-arid areas of eastern Algeria, central Tunisia and northwestern Libya. Pale specimens of *D. mauritanica*, which are similar to *D. deserti* specimens, have been recorded in southern Morocco, and recent phylogenetic studies based on a few specimens found low genetic differentiation among the two species. In this work we aim to give insights into the evolutionary history of North African *Daboia* species by assessing their phylogenetic relationships based on two mitochondrial DNA markers (cytb and ND4) sequenced for 40 specimens across their distributional ranges. Phylogenetic tree and haplotype network show high levels of genetic diversity and structuring in the western Maghreb region, differentiating three clades in Morocco: (1) Rif, and Middle and High Atlas Mountains, (2) Anti-Atlas Mountains, and (3) northern mountains of the Draa valley. A fourth clade includes specimens from eastern Morocco, Algeria, Tunisia and Libya. The invalidity of *D. deserti* as species is thus recognized. Range contractions in several areas of western Morocco and population expansions during Pleistocene climatic oscillations might explain these phylogenetic patterns. Further studies using palaeoclimatic data should be directed to test this hypothesis.

Maghreb, phylogenetics, Pleistocene, Viperidae.

(P02) **Filogeografía e historia poblacional del sapo de espuelas (*Pelobates cultripes*) inferida a partir de marcadores mitocondriales y microsatélites**

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El sapo de espuelas, *Pelobates cultripes* (Cuvier, 1829), se distribuye por la mayor parte de la Península Ibérica, por la costa mediterránea francesa y en algunas áreas disyuntas de la costa atlántica francesa. Sus poblaciones están en declive debido a la pérdida de hábitat y al impacto negativo de especies invasoras. Estudios previos han encontrado poca estructura genética entre sus poblaciones. En el presente estudio, se han muestreado 524 individuos pertenecientes a 54 poblaciones a lo largo de la distribución de la especie para inferir su historia evolutiva a partir de datos de un locus mitocondrial (ND4, 872 pb) y 14 loci polimórficos de tipo microsatélite. Los resultados muestran bajos niveles de diversidad genética y poca divergencia genética entre poblaciones, aunque pueden delimitarse tres grandes grupos poblacionales. El grupo “Noroeste” se distribuye por la meseta norte, el grupo “Sur” se extiende a través de la mitad sur peninsular y por último un grupo “Noreste” ocupa desde el este ibérico hasta Francia. Las poblaciones mediterráneas y atlánticas francesas muestran poca diversidad genética y bajos niveles de divergencia genética entre ellas. Diferentes parámetros de diversidad genética muestran una tendencia decreciente desde el sur hacia el norte de la Península Ibérica. Se han elaborado modelos de distribución de especies (SDMs) para inferir la idoneidad ambiental actual y proyectarla al último periodo interglacial (LIG) y a dos hipotéticos escenarios climáticos para el último máximo glacial (LGM): MIROC y CCSM. Los resultados indican una favorabilidad similar entre LIG, LGM-MIROC (pero no con LGM-CCSM) y la distribución actual, con correlaciones significativas entre la favorabilidad del hábitat y las diferentes medidas de diversidad genética (diversidad nucleotídica y riqueza alélica), lo que sugiere estabilidad demográfica a lo largo del tiempo, con recientes expansiones en el extremo norte de la distribución de la especie.

Phylogeography and population history of the Western Spadefoot toad (*Pelobates cultripes*) inferred from mitochondrial and microsatellite markers

The Western Spadefoot toad, *Pelobates cultripes* (Cuvier, 1829) is distributed throughout most of the Iberian Peninsula, along the Mediterranean coast of France and in some disjunct areas in the French Atlantic coast. Its populations are declining range-wide due to habitat loss and the negative impact of invasive species. Previous studies have found little genetic structure across populations. In this study, a total of 524 individuals from 54 populations across the species' range were sampled to infer the evolutionary history of the species based on data from one mitochondrial locus (ND4, 872 bp) and 14 polymorphic microsatellite loci. The results show low levels of genetic diversity and little genetic divergence between populations, although three major population groups can be delineated. A “North-Western” group is distributed across the northern Iberian plateau, a “Southern” group extends throughout most of the southern half of the Iberian Peninsula, and a “North-Eastern” group ranges from eastern Iberia to France, where populations in the Mediterranean and Atlantic coasts show little diversity and low levels of divergence from each other. Different estimates of genetic diversity show a trend of decreasing values from south to north, suggesting persistence of higher historical population sizes in the south of the Iberian Peninsula. Species distribution models (SDMs) were used to infer present environmental favourability and projected to the Last Inter Glacial (LIG) and to two hypothetical scenarios in the Last Glacial Maximum (LGM): MIROC and CCSM. The results show similar favourability between LIG, LGM-MIROC (but not LGM-CCSM) and the current distribution, with significant correlations between habitat favourability and different measures of genetic diversity (nucleotide diversity, allelic richness), suggesting long-term demographic stability, with moderate recent expansions at the northern end of the range of the species.

Phylogeography, demographic history, species distribution models, microsatellites, amphibian.

(P03) **Preliminary multilocus phylogeography of the lacertid lizard genus *Omanosaura* from the Hajar Mountains in Northern Oman**

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A total of 2729 base pairs from three mitochondrial (12S, ND4 and CYTB) and two nuclear (C-MOS, MC1R) genes were used to assess the molecular diversity and phylogenetic structure of the two *Omanosaura* lacertid lizards endemic to the Hajar Mountains in Northern Oman. Sequence data from fifteen samples of *O. jayakari* and five of *O. cyanura* were analyzed using Maximum likelihood and Bayesian inference phylogenetic methods, haplotype networks, and molecular dating. Our divergence time estimates suggest that the two species diverged around 8 million years ago (Mya). Mitochondrial sequence data show that the genetic diversity within *O. jayakari* is very low. Phylogenetic analyses support a partition into two clades without a clear geographic association across the Hajar Mountains, their split dating back to approximately 0.31 Mya. On the other hand, *O. cyanura* presents three relatively deep lineages associated with three geographic regions of the mountains: lineage 1, restricted to the Musandam Peninsula in the extreme north of the mountain range; lineage 2, including only one sample located in the Western Hajars; and lineage 3, including two samples from the Jebel Akhdar, the highest part of the Mountains, situated between the Western and Eastern Hajars. According to our time estimates, lineage 1 diverged from the other two lineages around 3.95 Mya, and lineages 2 and 3 would have diverged approximately 2.44 Mya. Nuclear data confirm the genetic homogeneity of *O. jayakari* and the high genetic diversity within *O. cyanura*. More samples of *O. cyanura* from undersampled areas will be needed to confirm the consistency of this pattern.

Omanosaura, Lacertidae, Oman, phylogeography.

(P04) Colour evolution in insular ecosystems. Chromatic variation in *Podarcis liolepis*

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Evolution in islands and islets often leads to changes in chromatic characters of animals. In many cases, the result of these changes is an extravagant pattern, the loss of some chromatic character and/or the loss of its function, or the fixation of rare phenotypes. These changes are often explained by historical events as well as by several selective processes. The coloration of *Podarcis* lizards provides many examples of insularity driving changes in coloration. *Podarcis liolepis atrata*, an insular clade that inhabits the Columbretes archipelago, (a group of small volcanic islets facing the Iberian Mediterranean coast) shows some differences respect to the general pattern found in continental *P. liolepis* populations. However, no study has addressed these differences objectively (i.e. using reflectance spectrophotometry techniques). Here, we compare the coloration of *P. liolepis atrata* with those shown by a continental population of *P. liolepis liolepis* (Penyagolosa massif; Iberian System), focusing on male coloration.

Although data from other populations may be necessary for understanding the evolution of *P. liolepis* coloration, our spectral analyses reveal chromatic differences in most lizard body surfaces: the dorsal cryptic coloration, the long-wavelength based belly and throat, and the ultraviolet (UV) patches found on the outer ventral scales (OVS). Overall, *P. liolepis atrata* (mainly dorsal and ventral surfaces) are darker than *P. liolepis liolepis*. This is probably due to a general increase in melanin deposition, a common phenomenon in insular evolution. Also dorsal and ventral coloration are more chromatically pure and short-wavelength biased in *P. liolepis liolepis* than in *P. liolepis atrata*. The colour of throat and belly varies in the same way between populations. Although the UV patches found in OVS are the less variable colour character in this species (probably because they evolve under a strong social selection), we found a highly significant difference in hue, the OVS of male *P. liolepis liolepis* being more short-wavelength biased than those of *P. liolepis atrata*. The available evidence suggests that the hue of UV patches is related to individual quality signalling. Thus, differences in this variable may indicate differences in the intrasexual selection strength.

Coloration, evolution, insularity, lizards.

(P05) Mecanismos asociados con la especiación: descripción de isótopos estables en los reptiles de Socotra

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Como sistemas aislados, las islas oceánicas son ejemplos únicos en la naturaleza para los biólogos evolutivos, y han contribuido considerablemente a nuestra comprensión de los procesos de especiación. En las islas, las especies colonizadoras encuentran diferentes condiciones ambientales a las del continente y pueden adaptarse rápidamente nuevos diseños corporales y fisiológicos. Socotra es una isla situada en el Océano Índico que se separó del continente africano hace unos 18 millones de años. En este estudio nos hemos fijado en seis especies de reptiles pertenecientes a dos familias: cuatro especies de Gekkonidae y dos de Phyllodactylidae. Estudios filogenéticos demostraron que ambos grupos son monofiléticos con un ancestro común y posterior especiación dentro de la isla. Estas especies muestran rasgos contrastados como son las diferencias morfológicas en el tamaño corporal y el tipo de hábitat ocupado. Los isótopos estables son una poderosa herramienta para el análisis de la dieta y el origen de los recursos alimenticios de los organismos. Se estudiaron muestras de cola de 87 ejemplares de las seis especies de reptiles y se analizaron las proporciones de los isótopos $^{13}\text{C}/^{12}\text{C}$ y $^{15}\text{N}/^{14}\text{N}$. Se examinaron las diferencias interespecíficas, así como las diferencias relacionadas con el tamaño corporal, el microhábitat y la distribución geográfica. Se observaron diferencias significativas en *H. granti* tanto para el carbono como para el nitrógeno, probablemente debido a su segregación altitudinal. También se observó correlación en las proporciones de $^{13}\text{C}/^{12}\text{C}$ con el tamaño corporal. Con respecto a los valores de $^{15}\text{N}/^{14}\text{N}$, se detectaron diferencias entre microhábitats (suelo, acantilados y vegetación). Las diferencias interespecíficas relacionadas tanto con el tamaño corporal como con el microhábitat se ajustan a las diferencias observadas en la dieta de las seis especies. Nuestros resultados sugieren que las diferencias isotópicas reflejan la selección de microhábitat y en paralelo cambios en la dieta entre las especies. Los análisis isotópicos pueden contribuir a desentrañar los factores que impulsan la especiación ecológica en los reptiles de Socotra.

Mechanisms associated with speciation: Description of stable isotopes of Socotran reptiles

As isolated systems, oceanic islands are unique examples in nature for evolutionary biologists that have contributed considerably to our understanding of speciation processes. On islands, new colonizers found different environmental conditions to those of the continent and may rapidly adapt to new body and physiological designs. Socotra is an island located in the Indian Ocean that separated from the African continent about 18 million years ago. It is a perfect setting to study this phenomenon. We focused this study on six reptile species from two families of geckos (four Gekkonidae and two Phyllodactylidae). Phylogenetic studies demonstrated that both groups are monophyletic with further intra-island speciation. These species show contrasted traits such as morphological differences in body size and type of microhabitat occupied. Stable isotopes have become a powerful tool for analyzing the diet and the origin of food resources of wildlife. We have examined tail samples of 87 individuals and analyzed the $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ ratios in order to examine interspecific differences, as well as differences associated to body size, microhabitat and geographical position. We observed significant differences in *H. granti* for both carbon and nitrogen isotopic values in respect to the rest of the species, probably due to its altitudinal segregation. We have also observed correlations of in $^{13}\text{C}/^{12}\text{C}$ ratios with body size. With respect to $^{15}\text{N}/^{14}\text{N}$ ratios, we detected differences between geckos' microhabitats (soil, cliff and vegetation). The interspecific differences related to both body size and microhabitat are in accordance to dietary variation among reptile species. Our results suggest that isotopic differences reflect microhabitat selection and parallel dietary segregation among species. Isotopic analyses can contribute to disentangle the factors that drive ecological speciation in Socotra reptiles.

Stable isotopes, reptiles, speciation, Socotra, island.

(P06) **Favourable areas for the co-occurrence of species with mostly parapatric distributions: the case of Iberian tree frogs and midwife toads**

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The study of areas of sympatry of species with predominantly parapatric distributions may provide valuable insights about the factors shaping species occurrence. This information is key in biogeography, evolutionary biology and conservation planning. In this study we model the distributions of two pairs of partially co-occurring amphibian species in the Iberian Peninsula: tree frogs (*Hyla molleri* and *H. meridionalis*), with 11% of the occupied area shared by both species; and midwife toads (*Alytes obstetricans* and *A. cisternasii*), which co-occur in 4% of the total occupied area. We obtained distribution data from the herpetological atlases of Portugal and Spain, consisting of presences and absences on UTM 10x10-km grid cells. We built generalized linear models of presence / absence for each species and for the co-occurrence of each species pair, using 24 potential predictor variables representative of Iberian physiography, climate and human activity. Variables were selected for each model using information and significance criteria. The models were rated using calibration and discrimination measures and were compared using non-parametric correlation analysis after accounting for spatial autocorrelation. Preliminary results show a negative correlation between environmental favourability for *A. obstetricans* and *A. cisternasii*, indicating environmental segregation of the two species. Among the tree frogs, there was a strong positive correlation between favourability for *H. meridionalis* and for its overlap with *H. molleri* – i.e., sympatry occurs mainly in areas favourable for *H. meridionalis*. These results provide clues to understand the evolutionary history of these species, including the evolution of reproductive isolation, and suggest that conservation efforts for tree frogs may be focused on the areas that are favourable for both species, whereas midwife toads will require specific measures tailored for each species.

Alytes, biogeography, distribution modelling, *Hyla*, parapatry, sympatry.

(P07) Modelando el nicho realizado del galápago europeo (*Emys orbicularis*) en España peninsular con varios algoritmos y a varias distancias de separación entre puntos de presencia

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Durante los estudios de distribución del galápago europeo (*Emys orbicularis*) realizados durante las últimas décadas, se han obtenidos numerosos datos que pueden utilizarse para elaborar modelos de nicho ecológico (ENMs). Uno de los problemas comunes de los ENMs es la autocorrelación espacial de los datos, que infla artificialmente los estadísticos de evaluación por pseudorreplicación de los puntos de entrenamiento y test. Uno de los métodos más empleados para corregirla es descartar puntos de entrenamiento cercanos entre sí. De esta manera, cuanto mayor sea esta distancia mínima entre puntos, la autocorrelación espacial será menor, pero también se reduce el tamaño muestral. Esta contradicción metodológica es el objeto de estudio de este trabajo, mediante la modelización del nicho realizado del galápago europeo. Se han realizado 1560 modelos a partir de 6 grupos de datos de presencia a diferentes distancias de separación entre puntos: 200 metros (n=474), 1 km (n= 336), 5 km (n=206), 10 km (n=146), 20 km (n=94) y 50 km (n=41) utilizando el 60 % de los datos como entrenamiento y el 40 % restante como test, remuestreando estos puntos en cada una de las 10 réplicas de los 26 algoritmos utilizados. Los modelos fueron evaluados mediante el True Skill Statistic (TSS), igualando a 0 el TSS si el algoritmo no logró una solución. El dataset de 50 Km de separación ofreció resultados muy pobres, siendo Maxent sin “hinge features” ni “threshold features” el que ofreció unos resultados más estables y con relativamente buena capacidad de predicción. En el 65,38% de los algoritmos se observa que los modelos a 200 m y 1 km de separación son similares entre sí, mientras que difieren de los algoritmos a 5 km, 10 km y 20 km de separación, que a su vez son similares entre sí. Este patrón puede explicarse por la elevada autocorrelación espacial que existe entre los puntos de presencia a distancias de hasta 1 km, lo que se evidencia al calcular la I de Moran de las variables a diferentes resoluciones espaciales (200m y 1, 5, 10, 20 y 50 km), siendo en todas las variables superior a 0.5 hasta los 5 km de separación entre puntos. Así, los modelos deberían hacerse con el dataset a 5 km de resolución, siendo los Support Vector Machines y los Generalized Boosted Models los modelos mejores y más estables. En futuros trabajos, trataremos de buscar la distancia óptima que permita modelar manteniendo el mayor poder predictivo posible, pero evitando una autocorrelación espacial excesiva.

Modelling the realized niche of the European pond turtle (*Emys orbicularis*) in peninsular Spain with various algorithms and different minimal distances among presence points

Research on the European pond turtle (*Emys orbicularis*), performed during the last decades, provided a large amount of data, which can be used to build ecological niche models (ENMs). One of the commonest problems to make ENMs is the spatial autocorrelation, which artificially inflates measures of accuracy by training-test data pseudo-replication. A possible method to deal with this problem is to reduce the dataset selecting data with a minimum distance among points. By this way spatial autocorrelation will be smaller, but also the sampling size is smaller. Our aim is to explore this trade-off by the European pond turtle's realized niche modelling. We built 1560 models from 6 datasets registering presence sites separated among them 200 meters (n=474), 1 km (n=336), 5 km (n=206), 10 km (n=146), 20 km (n=94) and 50 km (n=41) respectively, using 60% of points as training and 40% as test in each of the 10 replicates of the 26 algorithms used. Models' accuracy was evaluated by the True Skill Statistic (TSS), which was set to 0 when the algorithm could not find a solution. 50 km dataset offered very poor results, being Maxent (with no “hinge features” nor “threshold features”) the model with more stable results and with relatively good prediction skills. 65.8% of the algorithms showed very similar “200 m” and “1 km” models, but different to “5 km”, “10 km” and “20km”, which were similar among them. This pattern can be explained by the high spatial autocorrelation among presence locations at distances up to 1 km, showed by the Moran's I, which was higher than 0.5 in all the covariates below 5 km resolution. Therefore, in our case, models should be built with the 5 km dataset. At this distance among points the better and more stable models were Support Vector Machines and Generalized Boosted Models. In future researches, we will try to find the optimal distance among presence points, at which models maintain a good predictive power, but avoiding an excessive spatial autocorrelation.

Modelling, distribution, *Emys orbicularis*, spatial autocorrelation, sample size.

(P08) **Distribución y estructura poblacional del galápago leproso (*Mauremys leprosa*) en las comarcas del Camp de Tarragona, Cataluña**

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En 2013 y 2014 la Generalitat de Cataluña ha llevado a cabo una diagnosis del estado de las poblaciones de galápago leproso (*Mauremys leprosa*) en las cinco comarcas del Camp de Tarragona, al Sur de Cataluña. Se ha localizado la especie en nueve cuadrículas UTM 10x10 más de las que se tenía constancia en los diferentes muestreos hechos entre diciembre de 1995 y 2002. Posiblemente este aumento no se debe a la recuperación y expansión de la especie, sino a una falta de muestreo en diferentes zonas, como son los tramos medios de los ríos Siurana y Francolí. Se ha capturado y marcado un total de 324 ejemplares, de los que 203 eran juveniles, lo que supone un 62,65% del total. Por otro lado, de los 121 adultos, 47 eran machos y 74 hembras, 38,84% y 61,16% respectivamente. El número de hembras duplica prácticamente al de los machos, lo que garantiza la viabilidad de las poblaciones. Esto es especialmente significativo en las poblaciones del río Siurana, donde un 73% de la población adulta (n=47 de 64) eran hembras y en la Sèquia Major con un 60% (n=12 de 20) de hembras. En el lado opuesto está la población de los humedales del Pla de Santa María, donde ocho de los nueve ejemplares adultos eran machos (88,8%).

Distribution and population structure of Mediterranean Turtle (*Mauremys leprosa*) in the counties of Tarragona, Catalonia

In 2013 and 2014 the Government of Catalonia has carried out a diagnosis of the status of Mediterranean turtle (*Mauremys leprosa*) populations in five counties of Camp de Tarragona, south of Catalonia. The species was found in 9 new 10x10 UTM grids, compared to data collected in samplings made between December 1995 and 2002. However, this increase may not come from the recovery and expansion of the species, but from a lack of sampling in some areas, such as the middle reaches of the rivers Siurana and Francolí. A total of 324 animals were captured and tagged, of which 203 were juveniles, which represents 62.65% of the total. On the other hand, of the 121 were adults, 47 were males and 74 females, a percentage of 38.84% and 61.16% respectively. The number of females is almost twice that of males, ensuring the viability of populations. This is especially significant in populations of Siurana river, were 73% of the adult population (n=47 out of 64) were females, and Sèquia Major with a 60% (n=12 out of 20) of female population. On the opposite side, in the wetlands of Pla de Santa Maria, eight out of nine adult specimens found were males (88.8%).

Mediterranean turtle, tagged, population structure.

(P09) **A dieta do Sacarrabos (*Herpestes ichneumon*) em Portugal como um contributo para novas localizações de répteis**

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O Sacarrabos (*Herpestes ichneumon*) é um mamífero da família Herpestidae, que ocorre em cerca de 87% do território de Portugal continental. Nas últimas décadas tem vindo a alargar a distribuição geográfica de sul para norte e do interior para o litoral. Este carnívoro pode atingir 3 kg e 1 metro de comprimento, tratando-se de uma espécie cinegética de caça menor, com o estatuto de conservação “Pouco preocupante”.

O acesso a 678 amostras de animais, em resultado de atropelamentos ou de ações de correção de densidades de predadores, ocorridos em 13 distritos permitiu aprofundar o conhecimento da ecologia desta espécie em território nacional. Concretamente, a análise dos conteúdos estomacais possibilitou o estudo da dieta, através da identificação e do registo dos organismos ingeridos. Os dados obtidos revelam que cerca de 16% dos itens consumidos correspondem a animais da classe dos répteis, em que é frequente a cobra-rateira (*Malpolon monspessulanus*), a cobra-de-escada (*Rhinechis scalaris*), a lagartixa-do-mato (*Psammmodromus algirus*) ou a cobra-de-pernas-tridáctila (*Chalcides striatus*). Durante este estudo foram detetadas novas espécies de répteis ainda não descritas na dieta de sacarrabos, como o licranço (*Anguis fragilis*), a cobra-lisa-meridional (*Coronella girondica*) e a cobra-de-ferradura (*Hemorrhoids hippocrepis*). Estes animais foram identificados através da morfologia externa, por comparação com coleções de referência. Tendo em conta que é conhecida a localização da recolha de cada sacarrabos, a data da morte e, que estes animais vivem em territórios com cerca de 3 km², percorrendo 4 a 5 km por dia (fêmeas e machos, respetivamente), constatou-se que quatro indivíduos neste estudo continham nos seus estômagos três espécies de répteis recolhidos em áreas geográficas cuja localização ainda não se encontra assinalada nos mapas do Atlas dos Anfíbios e Répteis de Portugal. Assim, este estudo confirma a presença de licranço (*Anguis fragilis*) em Castelo de Vide (Portalegre), na quadrícula PD36; da cobra-de-pernas-tridáctila (*Chalcides striatus*) no Ciborro (Montemor-o-Novo), na quadrícula NC69, em Nossa Senhora da Graça do Divôr (Évora), na quadrícula NC87; e da cobra-lisa-meridional (*Coronella girondica*) em Vila Nova de Paiva (Viseu), na quadrícula PF02. Concluindo, este estudo reporta três novas espécies de répteis na dieta de sacarrabos, bem como quatro novas localizações de três espécies de répteis em Portugal.

The diet of Egyptian Mongoose (*Herpestes ichneumon*) in Portugal as a contribute to new reptile locations

The Egyptian Mongoose (*Herpestes ichneumon*) is a mammal from the family Herpestidae, that occurs in 87% of the mainland territory of Portugal. In the last decades this species has expanded its geographical distribution from south to north and inland to the coast. This carnivore can reach 3 kg and 1 meter in length, and is a species of small game, with the conservation status “Least Concern”.

The access to samples of 678 animals, obtained from road-kills or corrective actions of predator densities, from 13 districts allowed an understanding of the ecology of this species in national territory. Specifically, the analysis of stomach contents permitted the study of its diet through the identification of ingested organisms. The data show that about 16% of the consumed items are reptiles, where Montpellier Snake (*Malpolon monspessulanus*), Ladder Snake (*Rhinechis scalaris*), Large Psammmodromus (*Psammmodromus algirus*), or Three-toed Skink (*Chalcides striatus*) are common occurrences. During this study, new species of reptiles were found to be described in the diet of Egyptian Mongoose, such as the Slow Worm (*Anguis fragilis*), Southern Smooth Snake (*Coronella girondica*) and Horseshoe Whip Snake (*Hemorrhoids hippocrepis*). These animals were identified based on external morphology by comparison with reference collections. Given the knowledge of the location where each Egyptian Mongoose was collected, as well as the date of death, and that these animals live in territories with approximately 3 km², travelling 4 to 5 km per day (females and males, respectively), it was inferred that four specimens in this study contained in their stomachs three species of reptiles collected in geographical areas whose location has not yet been recorded on the distribution maps of the Atlas dos Anfíbios e Répteis de Portugal. Thus, this study confirms the presence of Slow Worm (*Anguis fragilis*) in Castelo de Vide (Portalegre), in the grid PD36; Three-toed Skink (*Chalcides striatus*) in Ciborro (Montemor-o-Novo), in the grid NC69, in Nossa Senhora da Graça do Divôr (Évora), in the grid NC87; and Southern Smooth Snake (*Coronella girondica*) in Vila Nova de Paiva (Viseu), in the grid PF02. In conclusion, this study report three new reptile species in the diet of Egyptian Mongoose, as well as four new locations for three species of reptiles in Portugal.

Egyptian mongoose, *Herpestes ichneumon*, carnivore, diet, reptiles.

(P10) **Habitat selection may explain microgeographic distribution of two close related species of skinks at Chafarinas Islands in North Africa**

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Among the species of *Chalcides* scincid lizards inhabiting the Mediterranean region, The Ocellated skink (*Chalcides ocellatus*) is the most widespread species, while the Chafarinas skink (*Chalcides parallelus*) has been found only along a narrow coastal strip of approximately 250 km between Nador in northeastern Morocco and Cape Carbón in northwestern Algeria. One interesting aspect of the biogeography of these two skinks is that both species occur at Chafarinas archipelago but while *Ch. ocellatus* is spread in the three islands of the archipelago, *Ch. parallelus* is present only in one island. To explain the microgeographic distribution in the Chafarinas archipelago of these close related skinks, we conducted a study to determine what, if any, differences existed in microhabitat use characteristics between species and populations. We examined the microhabitat selection of both species, by testing the null hypothesis of no differences between habitat factors associated with the species and a set of locations chosen at random from the general habitat. We then tested for differences in microhabitat selection between species and islands where they occur. Preliminary results show that both species do not use islands habitat at random and that when both species occur together in the same island, they select different microhabitats. The suitable habitat for *Ch. parallelus* is more abundant in the only island where the species is present and it may explain the distribution of the species within the archipelago.

Habitat selection, species distribution, Chafarinas Islands, lizards, North Africa.

(P11) Tolerancia térmica en ciclos de vida complejos: efectos ontogenéticos

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La capacidad para tolerar el aumento de la temperatura durante un periodo limitado de tiempo es una característica crítica para evaluar la vulnerabilidad en ectotermos ante el cambio climático. En especies con ciclo de vida complejo, cada etapa del ciclo de vida puede experimentar diferentes ambientes térmicos (sobre todo si las fases no se solapan en el tiempo o en el espacio) y esto puede ser relevante para las predicciones de respuestas evolutivas y las evaluaciones de la vulnerabilidad ante el cambio climático. Los anfibios son considerados como el taxón más amenazado entre los vertebrados y presentan un ciclo de vida complejo. Ambientes térmicos acuáticos de larvas y metamórficos limitan la posibilidad de realizar comportamientos de selección para mitigar los efectos del estrés por calor o por frío, mientras que los juveniles terrestres tienen más opciones de termorregulación mediante la selección activa de microhábitats térmicos. En este estudio examinamos los límites térmicos críticos de tres estadios de vida de anfibios (renacuajos acuáticos, individuos metamórficos y juveniles terrestres) de cinco especies de anfibios de una comunidad mediterránea, para evaluar las diferencias en tolerancias térmicas de calentamiento y enfriamiento. Se obtuvieron los límites térmicos críticos superior e inferior (CT_{max} y CT_{min} , respectivamente) mediante el empleo de un protocolo dinámico de incremento / descenso de temperatura a una velocidad de rampa constante de $0,25^{\circ}C \text{ min}^{-1}$, después de un mínimo de tres días de aclimatación en laboratorio a $20^{\circ}C$. Obtenemos resultados significativos para todas las especies en el análisis de CT_{max} . El estadio larval es el más tolerante, mientras que los estadios de metamórfico y juvenil tienen una disminución de $2,15^{\circ}C$ de media en el CT_{max} , lo que significa que en estos estadios disminuye la tolerancia al calentamiento. En el análisis de CT_{min} obtenemos resultados significativos para cuatro de las especies analizadas. Para tres especies, el estadio juvenil es el más tolerante, mientras que en los estadios larval y metamórfico el CT_{min} disminuye $1,55^{\circ}C$ en promedio, lo que refleja una pérdida de tolerancia al enfriamiento. Estas disminuciones en los límites de tolerancia térmica pueden resultar determinantes a la hora de evaluar la vulnerabilidad de estas especies a sufrir estrés térmico agudo, ya que esta transición de historia de vida coincide con la temperatura más alta registrada en los estanques. Por lo tanto, es importante tener en cuenta la tolerancia térmica de cada etapa para predecir el impacto del cambio climático en ectotermos con ciclo de vida complejo.

Thermal tolerance in complex life cycles: ontogenetic effects

The ability to tolerate increased temperatures for a limited period of time is a critical trait for assessing vulnerability to climate change in ectotherms. In species with complex life cycle, each life-cycle stage may experience different thermal environments (especially if stages do not overlap in time or space) and this may be relevant to predictions of both evolutionary responses and assessments of vulnerability to global warming. Amphibians are considered the most endangered class of vertebrates and exhibit a complex life cycle. Aquatic thermal environments of larvae and metamorphs limit the possibility of behavioural selection to mitigate the impacts of heat or cold stress, whereas terrestrial juveniles have more thermoregulation options by active selection of thermal microhabitats. In this research we examine critical thermal limits to evaluate differences across the three amphibian life stages (aquatic tadpoles, metamorphic individuals and terrestrial juveniles) of five amphibian species from a Mediterranean community, to assess differences in thermal tolerances of warming and cooling. We obtained upper and lower critical thermal limits (CT_{max} and CT_{min} , respectively) by employing a dynamic protocol of temperature increase / decrease at a constant ramping rate of $0.25^{\circ}C \text{ min}^{-1}$, after a minimum of three days of lab acclimation to $20^{\circ}C$. We obtain significant results for all species in the analysis of CT_{max} . The larval stage is the most tolerant, while the metamorph and juvenile stages have a decrease of $2.15^{\circ}C$ on average in CT_{max} , which means that the tolerance to warming decreases in these stages. In the analysis of CT_{min} , we obtained significant results for four of the species analyzed. For three species, juvenile stage is the most tolerant, while the larval and metamorphic stages have an average decreased of $1.55^{\circ}C$ in CT_{min} , which reflects a lower tolerance to cooling. These decreases in critical thermal limits may result determinant when assessing the vulnerability of these species to suffer acute thermal stress since this life history transition coincides with the highest temperature recorded in the ponds. Thus, it is important to consider the thermal tolerance of each stage to predict the impact of climate change in ectotherms with complex life cycles.

Amphibians, ontogeny, thermal tolerance limits, thermal tolerance, climate change.

(P12) Efeitos de uma onda de calor nos girinos de *Discoglossus galganoi* – estará esta espécie preparada?VANESSA PINTO¹, NUNO GONÇALVES¹, RICARDO RODRIGUES¹, BRUNO CARREIRA^{1,2}, RUI REBELO¹¹Centro de Biologia Ambiental / Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa, 1749-016 Lisboa, Portugal.²Animal Ecology, Department of Ecology and Genetics, Uppsala University. 752 36 Uppsala, Sweden.

Na Península Ibérica, os atuais registos e as projeções das alterações climáticas apontam para o aumento da frequência e da intensidade de fenómenos climáticos extremos. Neste trabalho estudou-se o efeito de uma onda de calor no desenvolvimento de girinos da rã-de-focinho-pontiagudo, *Discoglossus galganoi* (Capula, Nascetti, Lanza, Bulli e Crespo, 1985). Em outras espécies de anuros as ondas de calor podem antecipar a metamorfose, mesmo que isso signifique um menor tamanho (ou seja, uma menor fitness) dos recém-metamorfoseados.

Neste trabalho, os girinos de *D. galganoi* foram sujeitos a quatro tratamentos de temperatura: primavera fria (15°C); primavera quente (25°C); primavera normal (elevação gradual de 15°C a 25°C) e primavera normal com uma onda de calor (elevação da temperatura a 25°C durante uma semana). Os girinos foram também sujeitos a dietas ricas ou pobres em proteínas, uma vez que a regulação da dieta pode constituir uma resposta eficiente às alterações metabólicas induzidas pela temperatura. As duas dietas testadas foram larvas de quironómideo (Diptera, Insecta) (dieta rica) e a combinação de macrófita *Juncus heterophyllus* com larvas de quironómideo (dieta pobre).

O efeito da onda de calor dependeu da dieta a que os girinos foram submetidos: os indivíduos alimentados com uma dieta animal (rica em proteínas) não sofreram efeitos significativos, ao passo que os indivíduos alimentados com uma dieta menos rica em proteínas atingiram a metamorfose com menores dimensões, mas sem antecipar a metamorfose. Assim, a diminuição do tamanho de chegada à metamorfose dos girinos de *D. galganoi* não foi acompanhada por uma redução do período larvar, pelo que esta espécie terá pouca capacidade de resposta a ondas de calor, sofrendo até uma diminuição na fitness dos recém-metamorfoseados.

Heat wave effects in *Discoglossus galganoi* tadpoles – is this species prepared?

Recent data and future climate change projections show an increase of extreme climatic phenomena in the Iberian Peninsula, both in frequency and intensity. This project assesses the effect of a heat wave in Painted-frogs *Discoglossus galganoi* (Capula, Nascetti, Lanza, Bulli e Crespo, 1985) tadpoles. Heat waves can anticipate metamorphosis in other anuran species, usually with a cost: a smaller body length (i.e., lower fitness) of froglets.

Tadpole groups were exposed to four different temperature treatments: cold spring (15°C), normal spring (gradual increase from 15°C to 25°C), normal spring with a heat wave (temperature increase to 25°C during one week) and warm spring (25°C). Because their diets may influence the efficacy of the responses, due to temperature induced metabolic changes, different tadpole groups were fed protein-rich (chironomid larvae (Diptera, Insecta)) or protein-poor (a mix of the macrophyte *Juncus heterophyllus* and chironomid larvae) diets.

The effects of the heat wave were influenced by the tadpoles' diet. The protein-rich fed tadpoles were not significantly affected, whereas the protein-poor fed tadpoles metamorphosed with a shorter body length, without anticipating metamorphosis. As the decrease in size at metamorphosis was not followed by a reduction in the larval period, this species will probably show a poor ability to respond to heat waves, suffering a decrease in froglet fitness.

Diet, *Discoglossus galganoi*, heat wave, temperature.

(P13) **Avaliação dos efeitos dos parâmetros ambientais sobre o crescimento, a actividade antioxidante e peroxidação lipídica em *Pelophylax perezi***

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Os anfíbios no seu habitat natural estão frequentemente expostos a uma ampla variedade de xenobióticos que podem afetar a sua homeostase e, em última análise, a sua sobrevivência. Muitas vezes, de modo a poder avaliar os efeitos dos xenobióticos nos anfíbios são utilizados biomarcadores envolvidos na atividade antioxidante. Considerando que as populações naturais, para além dos xenobióticos, já se encontram a lidar com alterações ambientais, tais como a acidificação dos cursos de água ou aumento de temperatura, é essencial compreender como é que estas variações irão afetar biomarcadores utilizados comumente. Com este objetivo avaliámos separadamente, em *Pelophylax perezi*, o efeito de três parâmetros diferentes, pH (5,5; 6,5; 7,5 e 8,5), temperatura (16, 20, 24 and 28°C) e densidade animal (10, 20, 40 e 80 girinos/L) no crescimento, atividade enzimática antioxidante (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependente (GPx-Se), total-Glutathione Peroxidase (GPx-total) e Glutathione Redutase (GRed)) e peroxidação lipídica. Para ambos os ensaios de pH e temperatura, expuseram-se ovos no oitavo estágio de desenvolvimento, segundo Gosner, às várias condições. Para cada condição diferente, em cada um dos ensaios (pH e temperatura), a exposição terminou quando todos os girinos atingiram o estágio 25 de Gosner. Quanto à densidade de animais, a exposição começou com girinos no estágio 20 de Gosner e durou até todos os girinos na mesma condição terem alcançado o estágio 25. Apesar da maioria dos parâmetros avaliados não terem mostrado diferenças estatisticamente significativas, a maioria apresentava tendências bastante claras. O comprimento corporal revelou ser afetado pelo pH, tendo os valores mais altos deste originado girinos mais pequenos. De igual forma, a nível antioxidante, para a maioria das enzimas (GPx-total, GRed and GSTs) os girinos apresentam aparentemente uma atividade mais elevada para valores de pH mais baixos, concomitantemente com a peroxidação lipídica mais baixa. Relativamente à temperatura, existe um aumento óbvio do tamanho corporal, relacionado com temperaturas mais elevadas. Em acréscimo, a atividade das GSTs revelou a tendência de valores mais elevados de acordo com valores mais elevados de temperatura, e simultaneamente menos peroxidação lipídica. Relativamente ao ensaio das densidades animais, foi possível ver uma relação entre densidades mais altas e uma menor atividade antioxidante para a maioria das enzimas (GPx-Se, GPx-total, GRed), o que está de acordo com o aumento da peroxidação lipídica em conjunto com densidades maiores. No geral, este trabalho revela que as futuras alterações ambientais irão, certamente, afetar o sistema de defesa antioxidante dos anfíbios.

Evaluation of the effects of environmental parameters on growth, antioxidant activity and lipid peroxidation in *Pelophylax perezi*

Amphibians in their natural habitat are often exposed to a wide variety of xenobiotics that can affect their homeostasis and ultimately their survival. Many times, in order to assess the effects of xenobiotics on amphibians, biomarkers involved in the antioxidant activity are used. Taking into consideration that, besides xenobiotics, natural populations are already dealing with environmental alterations such as acidification of water courses or increased temperatures, it is essential to understand how these parameters by themselves will affect commonly determined biomarkers. With this purpose we assessed separately, in *Pelophylax perezi*, the effect of three different parameters, pH (5.5, 6.5, 7.5 and 8.5), temperature (16, 20, 24 and 28°C) and animal density (10, 20, 40, 80 tadpoles/L) on growth, antioxidant enzymes' activity (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependent (GPx-Se), total-Glutathione Peroxidase (GPx-total) and Glutathione Reductase (GRed)) and lipid peroxidation. For both pH and temperature assays, eggs in the 8th Gosner stage were exposed to the various conditions. For each different condition within each of the assays (pH and temperature) the exposure ended once all tadpoles reached the 25th stage. As for animal densities, the exposure began with tadpoles in the 20th stage and lasted until every tadpole in the same condition reached the 25th stage. Even though many of the assessed parameters did not result in statistically significant differences, the majority of them presented clear trends. Body length revealed to be affected by pH with higher values of pH originating smaller tadpoles. Also, at an antioxidant level for the majority of the enzymes (GPx-total, GRed and GSTs) the tadpoles apparently showed a higher activity at lower pHs, which was concomitant with the lower lipid peroxidation. As for temperature, there was an obvious increase in body size, related to higher temperatures. Furthermore, GSTs activity presented higher levels for higher temperatures, and simultaneously low levels of lipid peroxidation were recorded. When concerning the animal density assay, we observed a negative correlation between density and antioxidant activity for the majority of the enzymes (GPx-Se, GPx-total, GRed), which

resulted in an increase in lipid peroxidation for higher densities. Overall, this work reveals that future environmental alterations will most certainly affect the antioxidant defense system of amphibians.

Amphibians, *Pelophylax perezii*, pH, population density, temperature, antioxidant enzymes.

(P14) Ecophysiology tracks phylogeny and meets ecological models in an Iberian geckoCATARINA RATO, MIGUEL A. CARRETERO

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Because the fitness of ectotherms, including reptiles, is highly dependent on temperature and water availability the study of ecophysiological traits, such as preferred temperature and water loss, may allow developing mechanistic models of potential distribution and provide evidence about possible factors restricting species ranges. The Moorish gecko, *Tarentola mauritanica* has a circum-Mediterranean distribution, and is a species-complex, comprising at least six distinct evolutionary lineages. In the Iberian Peninsula, two forms of the complex are found; an endemic clade distributed in central and south/western Iberia (Iberian Clade) and an European Clade, widely introduced throughout the Mediterranean Basin, which occurs along the Iberian coastline. In the present study, we measured the preferred temperatures (T_p) and water loss rates (WLR) for a total of 84 males from seven distinct populations belonging to the two lineages present in Iberia, in order to evaluate the existence of trait conservatism or, if on the contrary, there is an ecophysiological shift between populations, lineages or both. Additionally we tested for a trade-off between water and thermal traits and assessed if climate regime had any influence on the ecophysiological patterns. Our results demonstrate that T_p is quite conserved at both the population and lineage levels but also corroborate the general idea that nocturnal/crepuscular geckos select a wider and lower range of body temperatures when compared to diurnal lizards. No correspondence between mean T_p and body size was found. In contrast, water-loss experiments reveal clear differences not only between populations but also between the two Iberian lineages of *T. mauritanica*; with the Doñana population (European lineage) presenting the highest water loss ratio while Murcia (Iberian lineage) having the lowest. T_p and WLR were uncorrelated at all levels. Overall, the European lineage displayed a trend for higher water loss with more differences between populations when compared to the Iberian lineage. The identification of a phylogenetic signal regarding WLR suggests distinct evolutionary physiological responses to the environment in both lineages, which may account for the differences in their patterns of range expansion, as already suggested by correlative distribution models.

Tarentola mauritanica, ecophysiology, evolutionary ecology, Iberian Peninsula, preferred temperature.

(P15) **Ecofisiología de *Iberolacerta monticola* y *Podarcis hispanica* 1A simpátricas en Serra de Estrela, Portugal**

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Comparamos las temperaturas preferidas (Tp) y tasas de pérdida de agua (EWL) en dos lacértidos simpátricos que exhiben una segregación altitudinal parcial si bien solapan en el macizo montañoso de Serra Estrela en Portugal: *Iberolacerta monticola* y *Podarcis hispanica* 1A. Dado que la Tp presenta una considerable inercia filogenética, es posible que se observen patrones ecofisiológicos semejantes a los ya detectados en otros pares de especies similares. En el caso ya estudiado en Eslovenia, se demostró que *Iberolacerta* era capaz de termorregular en un rango estacional de Tp más estrecho, así como de perder menos agua que *Podarcis* en un intervalo experimental de ocho horas. Tanto una menor variación estacional de Tp como una baja EWL pueden relacionarse con una mayor precisión termorreguladora, que representa una ventaja considerable en especies que habitan los ambientes rigurosos de la alta montaña. Capturamos igualmente lagartijas en un área de simpatría de Serra da Estrela en Portugal en primavera y verano. En primavera, los animales fueron primero expuestos a un gradiente térmico para obtener datos de Tp y al día siguiente a un experimento de pérdida de agua. En verano las ejemplares solo pasaron por el experimento de temperatura preferidas. Pese a lo esperado, no detectamos diferencias significativas de Tp entre estaciones. Del mismo modo, tampoco encontramos diferencias de EWL entre especies. Dado que ambas especies difieren mucho en longitud hocico-cloaca y masa corporal, siendo *I. monticola* más larga y pesada que *P. hispanica*, incluimos ambas variables en el análisis de Tp y EWL. El hecho de que los resultados se mantengan aún así implica que la ecofisiología no depende de la talla de las lagartijas. A continuación evaluamos la dependencia de la variación de Tp (error estándar) respecto a la talla. En este caso, observamos que los ejemplares mayores, sea dentro de cada especie sea *I. monticola* respecto a *P. hispanica*, presentaron menor variación de Tp y, por tanto, termorregularon de forma más precisa. Esto añade un elemento más que señala a *Iberolacerta* como termorregulador más preciso que *Podarcis*, tal como ya ha sido evidenciado en Eslovenia.

Eco-physiology of *Iberolacerta monticola* and *Podarcis hispanica* 1A sympatric in Serra de Estrela, Portugal

We compared preferred temperatures (Tp) and water loss rates (EWL) of two sympatric lacertids that exhibit partial altitudinal segregation but co-habit on the mountainous area of Serra da Estrela in Portugal; *Iberolacerta monticola* and *Podarcis hispanica* 1A. Because Tp carries substantial phylogenetic inertia, we expected to observe similar eco-physiological patterns to those detected in other similar species tandems. In a co-habiting pair from Slovenia, *Iberolacerta* was found to be able to thermoregulate to a seasonally narrower range of Tp and lost less water after eight hours of experiment than *Podarcis*. Narrower seasonal range in Tp and lower EWL were related to a higher precision in thermoregulation which is an advantage in species that live in harsher environments like mountain tops. In this study, we similarly collected lizards in a sympatric area of Serra da Estrela in Portugal in spring and in summer. In spring, lizards were first placed in a thermal gradient to obtain data on Tp and the next day we tested them for water loss. Lizards collected in summer were used only in experiments of preferred body temperatures. Despite our expectations, we failed to record significant differences between species in seasonal variation in Tp. Likewise we did not find interspecific differences in water loss. Since both species greatly differed in snout to vent length and body mass, *I. monticola* is heavier and longer than *P. hispanica*, we included these two variables in analyses of Tp and EWL. The fact that results remain unchanged implies that eco-physiology is not depended on size of the lizards. Secondly we tested for dependence of Tp variation (standard error) and body size. In this case, we observed that bigger lizards, both within each species or *I. monticola* regarding *P. hispanica*, exhibited less variation in Tp and, hence, thermoregulated more accurately. This adds another evidence suggesting that *Iberolacerta* is a more accurate thermoregulator than *Podarcis*, as already found in Slovenia.

Ecophysiology, preferred temperature, water loss, *Iberolacerta*, *Podarcis*.

(P16) **O Custo da nidificação: Condição corporal e parâmetros sanguíneos em fêmeas nidificantes de jacaré-tinga (*Caiman crocodilus*) na Amazônia Central**

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As quatro espécies de crocodilianos que ocorrem na Amazônia Central constroem os seus ninhos durante a época de seca anual. Alguns autores sugerem que vários aspectos associados à nidificação (e.g. produção e postura de ovos, construção e manutenção do ninho) podem influenciar a condição corporal das fêmeas em crocodilianos. No entanto, para crocodilianos amazônicos não existe qualquer informação sobre este assunto. Por esse motivo, elaboramos um estudo com o objectivo de determinar a condição corporal, segundo o factor de Fulton "K", e os níveis de glicose e triglicérides plasmáticos de fêmeas nidificantes de uma população de jacaré-tinga (*Caiman crocodilus*) da Reserva de Desenvolvimento Sustentável Piagaçu-Purus, Amazônia Central, Brasil. Durante a época de nidificação de 2012, capturamos 43 fêmeas nidificantes junto ao respectivo ninho (F_{ninho}). Durante o mesmo período, capturamos também 15 não nidificantes durante a noite no canal de água principal ($F_{\text{água}}$). Logo após a imobilização do animal e, antes do manuseio para a morfometria, foi colectada uma amostra de sangue da região do seio occipital, centrifugada a 6400 r.p.m. durante cinco minutos para a separação e congelamento imediato do plasma em nitrogénio líquido. Os níveis plasmáticos de glicose e triglicérides foram determinados em sistema automatizado (Cobas Mira Plus®), utilizando kits de reacção específicos (LabTest™). Os valores obtidos evidenciam diferenças significativas na condição corporal (ANOVA, média \pm DP; $F_{\text{ninho}} = 2,16 \pm 0,16$; $F_{\text{água}} = 2,39 \pm 0,17$; $p < 0,001$), e nos níveis de glicose ($F_{\text{ninho}} = 49,5 \text{ mg/dL} \pm 13,4$; $F_{\text{água}} = 87,7 \pm 33,7$; $p < 0,001$) e triglicérides plasmáticos ($F_{\text{ninho}} = 34,2 \text{ mg/dL} \pm 11,4$; $F_{\text{água}} = 141,0 \pm 81,4$; $p < 0,001$) entre fêmeas nidificantes e não nidificantes. Estes resultados indicam que o cuidado do ninho tem influência na condição corporal das fêmeas nidificantes, associando ao sucesso de eclosão dos ovos um custo metabólico dependente das reservas energéticas disponíveis. Por manterem-se próximas ao ninho durante o período de incubação, estas fêmeas tem menos acesso e oportunidade de se alimentar. Essas informações poderão permitir a avaliação do estado fisiológico das fêmeas enquanto guardam o ninho e a percepção da influência deste na resposta à captura. Além disso, são subsídios também importantes para programas de monitoramento que se baseiam na procura de ninhos e/ou captura das fêmeas.

The metabolic cost of nesting: Body condition and blood parameters of nesting females of spectacled caiman (*Caiman crocodilus*) in Central Amazonia

All four crocodilian species that occur in Central Amazonia build their nests during the annual dry season. Some authors have already suggested that several aspects associated with nesting (e.g. egg production and laying, construction and maintenance of the nest, among others) may influence body condition of nesting females. However, for amazonian crocodilians, there is still no available information on this subject. For that reason, we set up an study to evaluate body condition (according to Fulton's "K" body condition factor) and plasmatic concentrations of glucose and triglycerides of nesting females in a population of spectacled caiman (*Caiman crocodilus*) that occur in Piagaçu-Purus Sustainable Development Reserve (PP-SDR), Central Amazonia, Brazil. During the nesting season of 2012, we captured 43 nesting females near their respective nest (F_{nest}). During the same period, we also captured 15 non-nesting females during the night in the main water channel (F_{water}). Immediately after the immobilization of the animal and, before taking morphometric measures, a blood sample was collected from the occipital sinus. All blood samples were centrifuged at 6400 r.p.m and the resulting plasma was frozen in liquid nitrogen. Plasmatic concentrations of glucose and triglycerides were determined with Cobas Mira Plus® automated system, using specific colorimetric reaction kits (LabTest™). All values obtained demonstrated significant differences in body condition (ANOVA, mean \pm SD; $F_{\text{nest}} = 2,16 \pm 0,16$; $F_{\text{water}} = 2,39 \pm 0,17$; $p < 0,001$) and plasmatic concentrations of glucose ($F_{\text{nest}} = 49,5 \text{ mg/dL} \pm 13,4$; $F_{\text{water}} = 87,7 \text{ mg/dL} \pm 33,7$; $p < 0,001$) and triglycerides (F_{nest}

= 34,2 mg/dL \pm 11,4; F_{water} = 141,0 mg/dL \pm 81,4; $p < 0,001$) between nesting and non-nesting females. Our results point out that nest attendance has influence on female body condition and exhibits a metabolic cost associated to hatching success, which may be dependent of female energetic reserves. As nesting females of *C. crocodilus* remain near the nest during all incubation period, they have less access and opportunity to feed. This information may enable the evaluation of the physiological state of females while attending their nest, and may even allow us to better understand its influence on individual response to capture. Also, this information are very important to monitoring and conservation programs, especially those which rely on nest surveys and/or capture of attending females.

Amazon, blood, body condition, caiman, nest, Varzea.

(P17) **La función de múltiples señales visuales en la lagartija *Psammodromus algirus*: indicando diferentes cualidades a diferentes receptores**

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De todas las señales y recursos que emplean los organismos para comunicarse, la coloración es una de las que más interés ha suscitado en los últimos años. Las señales basadas en coloración pueden comunicar diferentes cualidades de los individuos, y estar dirigidas a diferentes receptores. La lagartija colilarga (*Psammodromus algirus*) del sureste de la Península Ibérica posee varias manchas de pigmentación en la zona ventro-lateral del cuerpo: naranja en las comisuras, amarilla en la zona gular, y ocelos azules en los laterales. En este trabajo estudiamos en conjunto y de manera cuantitativa la coloración ventral y las manchas de pigmentación, y su relación con diferentes aspectos biológicos de esta lagartija, para tratar de discernir la función de cada rasgo. Todo bajo el marco de un gradiente altitudinal de 2200 m s.n.m.

Nuestros resultados apuntan que la coloración ventral y las señales cromáticas en *P. algirus* son señales honestas de la calidad de los portadores. Los ornamentos cromáticos aparecen sobre todo durante la estación reproductiva. Además, mientras que el color de las comisuras y el número de ocelos actúan como señal del sexo del portador (comisuras amarillas en las hembras y naranjas y mayor número de ocelos en los machos), la presencia de la mancha amarilla en la zona gular es principalmente indicativa de la edad, la madurez reproductiva y probablemente del estatus social. El tamaño de las manchas indican la condición física, y probablemente el estatus reproductivo (fertilidad y receptibilidad en hembras) y la fuerza de mordisco (en machos). Pero existe un coste asociado a las señales vistosas, ya que las lagartijas con señales más visibles sufren más depredación. Finalmente, los individuos procedentes de altitudes mayores mostraron señales cromáticas más llamativas respecto a los de zonas más bajas, así como un menor dicromatismo sexual entre machos y hembras. Nuestros resultados sugieren que algunas señales visuales están bajo selección sexual (color de la comisura, número de ocelos) mientras que otras intervienen en comunicación agonística o intrasexual (tamaño de las manchas de pigmentación). En conclusión, las diferentes señales visuales presentes en esta lagartija indican diferentes cualidades, y se combinan para aumentar la cantidad y la calidad de la información.

The function of multiple visual cues at the lizard *Psammodromus algirus*: different cues indicates different qualities to different receptors

Among all the signals and resources that animals can use in communication, coloration is one of the things that generate more interest. Signals based on coloration can inform about different qualities of the bearer, and to different receptors. Large *Psammodromus* (*Psammodromus algirus*) from southeastern Spain shows different chromatic patches in ventral and lateral sides: orange in the commissures, yellow in the gular region, and blue blotches in the flanks. In this work we studied as a whole and in a quantitative way the ventral coloration and pigmentary patches. Moreover, we related these colour characteristics with information about quality and status of the individuals. All the study was done in the framework of an elevational gradient of 2200m a.s.l.

The results pointed to ventral coloration and colour badges of *P. algirus* as social and/or honest signals of the individual's quality. Colourful ornaments appeared especially during breeding season. Moreover, whereas colour of the commissures and number of ocelli were distinguishing of the sex, the presence of yellow pigmentary patches was likely indicative of age, reproductive maturity, and probably social status. In addition, the pigmentary patches are larger in better condition individuals, and probably signalling fertility (in females), and bit strength (in males). However, being more attractive implies a higher risk of predation. Finally, pigmentary patches were more frequent and bright at highlands. Our results suggest that while some visual signals are under sexual selection (commissures colour, or number of ocelli), others works during agonistic or intrasexual encounters (pigmentary patches area). In conclusion, different badges inform about different individual characteristics, and they are combined to increase the intensity and quality of the information.

Elevational gradient, honest signals, lacertids, lizard coloration, visual signals.

(P18) **Subtle spectral differences between polychromatic Iberian and Italian populations of *Podarcis muralis***

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Population polychromatism is particularly common in lizards, and much effort has been devoted to study the evolution of this phenomenon. Recently, some lacertid species, which often show population polymorphic ventral colorations, have attracted much attention. *Podarcis muralis* show a long-wavelength based ventral coloration, resulting in several alternative colour morphs: white, yellow and orange, as well as the white-orange and yellow-orange rare intermediate morphs. Here we performed an objective spectrophotometrical analysis with the aim to compare an Iberian (i.e. Eastern Pyrenees) and an Italian (i.e. Po Plain) polychromatic populations. Although the appearance of *P. muralis* coloration is similar in both populations, surprisingly we found a wide level of chromatic differences in the ventral colour morphs, as well as in the ultraviolet (UV) spots located on the outer ventral scales and the brown dorsal coloration. Assuming that different colour surfaces evolve under different selective regimes, we discuss some hypotheses which may account for these spectral differences. Thus, as the variation in the dorsal coloration is mainly explained by crypsis (i.e. background colour matching), the observed variation may be caused by chromatic differences in the natural backgrounds. Differences in chromatic properties of UV spots may be caused by different intensities of intrasexual selection. Finally, population chromatic differences within ventral morphs may be caused by ecological differences between populations, different intersexual selective intensities, different evolutionary histories, or more probably a combination of some of these factors.

Coloration, evolution, *Podarcis muralis*, population polychromatism.

(P19) Problems with spectral measurements: when colour patches are too smallGUILLEM PÉREZ I DE LANUZA¹, MARÍA DEL CARMEN GARCÍA CUSTODIO², ENRIQUE FONT²¹CIBIO Research Centre in Biodiversity and Genetic Resources, InBIO, Universidade do Porto.²Institut Cavanilles de Biodiversitat i Biologia Evolutiva, Universitat de València. Carrer Catedrático José Beltrán Martínez 2, 46980 Paterna, València, Spain.

During the last decades, reflectance spectrophotometry has become a powerful tool in the evolutionary study of animal coloration. Portable spectrophotometers make it possible to obtain huge amounts of quantitative data, and subsequently calculate many valuable variables. However, spectrophotometers can provide misleading data. If a measurement is well done, the resulting spectrum represents the light reflected from a natural colour patch. In contrast, a wrong measurement gives an artifactual spectrum, which is contaminated by light from more than one colour patch (i.e. a chimeric spectrum). Therefore, these spectra do not represent natural colours and cannot be used in analyses. The problem of artifactual spectra is especially serious when the colour patches of interest are relatively small (i.e. smaller than the light beam of the spectrophotometer) and/or they are shown in a complex pattern. In lacertids, some of the most interesting colour patches are small spots, such as the conspicuous ultraviolet (UV) spots shown by many species in their lateral and ventrolateral surfaces. In many cases, the small size of these patches prevents measurement and causes a loss in sample size. Here, we evaluate the actual impact of chimeric spectra on spectral analyses, testing whether useful information can be obtained from artifactual spectra. We captured 60 adult males *Podarcis muralis* and measured the reflectance of their UV spots ensuring that the light beam only illuminates the spot. In addition, we measured artifactual spectra which included, in addition to the UV spot, a portion of the surrounding black spots, or a portion of the white, yellow or orange ventral background coloration. We extracted hue (i.e. peak location), chroma (UV chroma) and brightness values from the spectra and compared those variables extracted from the correctly measured spectra with those of the artifactual spectra. Correlative analyses and paired-t tests show that chroma and brightness are extremely affected by artifactual measurements. However, hue is relatively unaltered, especially when the contamination comes from a black spot. We suggest that, with due caution, the peak location of artifactual spectra may be used for analyses. However, chroma and brightness measures calculated from artifactual spectra are not valid measures and should be discarded.

Coloration, lizards, methodology, sample size, spectrophotometry.

(P20) Utilização do padrão da íris para a foto-identificação de *Thecadactylus rapicauda*CATARINA SERRA GONÇALVES¹, ADRIÀ LÓPEZ-BAUCELLS^{1,2,3}, RICARDO ROCHA^{1,2,4}¹Centro de Biologia Ambiental, Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa. Bloco C2, Campo Grande, 1749-016 Lisboa, Portugal.²Biological Dynamics of Forest Fragments Project, National Institute for Amazonian Research (INPA) and Smithsonian Tropical Research Institute. C.P. 478, Manaus, AM 69011-970, Brazil.³Museu de Ciències Naturals de Granollers, Àrea Investigació en Quiròpters. Av. Francesc Macià 51, 08402 Granollers, Catalonia, Spain.⁴Metapopulation Research Group, Faculty of Biosciences, University of Helsinki. PO Box 65 (Viikinkaari 1), FI-00014 Helsinki, Finland.

Na presença de marcas naturais que permitam a identificação ao nível do indivíduo a foto-identificação pode ser usada como uma metodologia não-invasiva e de baixo custo para estudos de captura-recaptura. A utilização de foto-identificação em osgas tem sido limitada pela ausência de marcas naturais que permitissem uma fácil e rápida identificação individual. Contudo, um estudo recente demonstrou que o padrão individual da íris pode ser usado na foto-identificação de osgas do género *Tarentola* e que o processo poderia ser facilitado através do uso do programa de identificação de acesso gratuito Interactive Individual Identification System (I3S). O presente estudo pretende avaliar a viabilidade da utilização do programa I3S na foto-identificação de espécies de osgas com padrões de íris mais complexo do que o género *Tarentola*, e comparar a eficiência do programa I3S Classic com o mais recente I3S Contour. A nossa espécie focal é *Thecadactylus rapicauda*, o maior phyllodactílideo neotropical e o trabalho de campo decorreu no Projecto de Dinâmica Biológica de Fragmentos Florestais, na Amazônia Central, Brasil. Foram analisadas 59 fotos correspondentes a 29 indivíduos de *Thecadactylus rapicauda*, e embora tenham sido aplicados pequenos ajustamentos relativamente à metodologia descrita anteriormente, concluiu-se que tanto o I3S Classic como o I3S Contour podem ser usados na foto-identificação da íris de esta espécie. A eficácia dos programas na foto-identificação foi avaliada pela comparação da posição da foto correspondente de indivíduos previamente foto-identificados, e revelou-se significativamente diferente entre os diferentes softwares (Wilcoxon signed-rank test $W = 1278.5$; $P < 0.01$). Verificou-se também que o programa I3S Classic avaliou correctamente 92% das fotos na 1ª posição, enquanto o I3S Contour apenas identificou 68% das fotos referentes ao mesmo indivíduo na 1ª posição. Constatou-se que usando o I3S Classic, a pior posição de uma foto do mesmo indivíduo foi a 5ª, contrastando com a 21ª posição com recurso ao uso do I3S Contour. A foto-identificação pode deste modo, ser utilizada em espécies com padrão de íris relativamente complexo, evitando vários constrangimentos éticos e logísticos associadas a técnicas de marcação invasivos disponíveis para o estudo de osgas.

Iris photo-identification of the Amazonian Turnip-tailed gecko *Thecadactylus rapicauda*

In the presence of naturally occurring variegated markings allowing individual identification, photo-identification (photo-ID) can be used as a non-intrusive and relatively inexpensive methodological alternative to artificial marking techniques commonly applied in mark-recapture-studies. Yet, for a large number of species of the most diverse group of lizards, the geckos, individual identification based on photographs was not possible because no such naturally occurring markings had yet been identified. Recently however, a study demonstrated the possibility of using the individual iris pattern to photo-identify nocturnal geckos of the genus *Tarentola*, using the freely available pattern matching software Interactive Individual Identification System (I3S). Our study was aimed at assessing the feasibility of using I3S for the iris photo-ID of gecko species with a more complex iris patterns than the genus *Tarentola* and, to compare the efficiency of the I3S Classic and the recently released I3S Contour in matching photos of known specimens. Our study species was the Amazonian Turnip-tailed gecko *Thecadactylus rapicauda*, the largest Neotropical phyllodactylid, and fieldwork was conducted at the Biological Dynamics of Forest Fragments Project, Central Amazon, Brazil. Using a library of 59 photos from 29 individuals we have found that, contingent to minor adjustments to the previously described methodology, both I3S Classic and I3S Contour can be used for the iris photo-ID in this species. The matching efficiency, assessed by comparing the matching rank of photos of known specimens, differed between I3S Classic and I3S Contour softwares (Wilcoxon signed-rank test $W = 1278.5$; $P < 0.01$) and whereas I3S Classic correctly matched 92% of the photos in the 1st rank, for I3S Contour this figure was only 68%. Also, when using I3S Classic the worse ranked photo of a known specimen occupied the 5th position, contrasting to the 21st occupied by the worse ranked photo of a known specimen using I3S Contour. Iris photo-ID can therefore be used in gecko species with relatively complex iris patterns and we argue that this technique is more ethically acceptable and circumvents several ecological and logistical constraints associated with invasive marking techniques available for the study of geckos.

Photo-identification, mark-recapture-studies, I3S, Neotropical gecko.

(P21) **Comparative topographic anatomy between three genera of snakes from the Crotalinae subfamily: *Lachesis muta*, *Bothrops jararaca* e *Crotalus durissus***

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The Crotalinae subfamily is composed of venomous vipers found in Asia and the Americas. The snakes of this subfamily are distinguished by the presence of a heat-sensing pit organ located on either side of the head. In this study we compared the topographic anatomy of species from three genera of Crotalinae found in Brazil: *Lachesis muta*, *Bothrops jararaca* and *Crotalus durissus*. *Lachesis muta* has crepuscular habit and is distributed in tropical forests with low human population density and is the only Crotalinae oviparous snake in the Americas. In Brazil the gender *Lachesis* is distributed mainly in North and Northeast states and due to deforestation is in rapid decline, having been included in the Red List of Threatened Species. *Bothrops jararaca* is widespread throughout Brazil, occupying a wide range of habitats, from rainforests to open fields. *Crotalus durissus* is found from the state of Rio Grande do Sul to Mato Grosso do Sul and Minas Gerais, Rondônia, Amazônia, and Pará, always in dry-open areas. Until recently, the genus *Crotalus* was considered absent from areas occupied by the Atlantic rain forests of Brazil; however, specimens of *C. d. terrificus* have been reported in Rio de Janeiro. For this study, ten adult males and ten adult females of *B. jararaca* and *C. durissus* and one female and one male of *L. muta* kept in captivity at the Laboratory of Herpetology were used after their natural death. The animals were dissected and 25 internal structures examined for their position, in percentage, in relation to their snout vent length (SVL). As expected for all snakes, paired organs located on the right side are more cranial than the ones located on the left side, with the exception of the thymus. Although *B. jararaca* and *C. durissus* show differences in their external morphology and size of the scales, there is almost no differences in the position of their internal organs in relation to their SVL. Both species have tracheal lung (33% to 40% of their SVL) and the thyroid is located in the apical region of the heart at a distance equal to 29% to 35% of SVL. *Lachesis muta* is the only Brazilian Crotalinae that has no tracheal lung, but trachea appears separated from the lung. The knowledge of this particularity is crucial to the maintainers, as a simple deworming can lead the animal to death by suffocation, if it is parasitized by pentastomids. The trachea of *L. muta* is 20% smaller than that of *B. jararaca* and *C. durissus*. All organs of *L. muta* are positioned quite cranially compared to the positioning of the same organs in *B. jararaca* and *C. durissus*. The knowledge of the topographic anatomy of a specific species, besides being a key to understand the functioning of the organism and its pathologies, is essential in the clinical examination and interpretation of diagnostic images. Supported by FAPESP: 12/18362-3.

Topographic anatomy, *Crotalus*, *Bothrops*, *Lachesis*.

(P22) Hematology and serum biochemistry of *Lachesis muta* (Serpentes: Viperidae)

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Lachesis muta (bushmaster), the largest venomous snake of Brazil, is distributed in the North of Brazil and also in the Atlantic Forest that ranges from Rio de Janeiro to Ceará. Due to deforestation, the number of bushmasters is in rapid decline and the species has been included in the red list of threatened species of the “International Union for Conservation of Nature and Natural Resources”. Instituto Butantan receives bushmasters only sporadically (in 17 years, only 10 specimens were received), and nowadays the number of *Lachesis* maintained at the Laboratório de Herpetologia is sufficient for the production of antivenom but not for immunobiological researches. Establishing hematological and blood biochemical standards for this species is of great importance, as these data do not exist in the literature and are essential to monitor the healthiness of captive specimens. Hematological data are used to detect conditions affecting the body as a whole, which qualitatively or quantitatively alter blood cells, and through biochemical tests metabolic disorders can be diagnosed. The blood of snakes comprises an extracellular medium, plasma, and nucleated cells represented by erythrocytes, thrombocytes, lymphocytes, azurophils, heterophils and basophils. Biochemical analysis is a diagnostic tool which can be used to obtain important information regarding snakes. In this study we performed the following biochemical tests: uric acid, total protein, phosphorus, calcium, creatinine, creatine, cholesterol, albumin and alkaline phosphatase. Two healthy adults kept in captivity at the Laboratory of Herpetology since 2009 and adults recently acquired (SISBIO No. 34324-1) were used in this study. The blood count among captive and recently acquired *L. muta* showed no significant differences (red cell count/mm³: 594.000±166.973, white cell count/mm³: 6.600±3.228 and trombocyte cell count/mm³: 7.000±1.695) but in the specific leucocyte count we found that basophil and lymphocyte counts were statistically higher in the snakes from the wild. The biochemical tests showed statistical differences in most cases, with higher values in animals kept in captivity (e.g. total protein, calcium, albumin and phosphorus) except the test of cholesterol. These results can be due to the more regular feeding in captivity than in the wild and to the stress that the newly acquired animals were still suffering.

Lachesis, hematology, serum biochemistry.

(P23) Blood transfusion in snakesDIEGO R. ARRUDA, DANIEL R. STUGINSKI, KATHLEEN F. GREGO

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In recent years transfusion medicine, as well as immunohematology studies, have become essential for the treatment of various diseases, which encouraged the development of new technologies and better understanding of the use of blood and its components. Currently, blood transfusions are increasingly common in veterinary medicine and are often used in emergency and surgical procedures. The main indications are life-threatening severe anemia, immune-mediated diseases and severe non-regenerative neonatal isoerythrolysis. In snakes, the main indication for blood transfusion is severe anemia due to internal bleeding caused by traumas and caquexia. The determination of hematological and biochemical parameters is essential to estimate the health status of the snakes recently-caught in nature and those maintained in captivity. Blood typing and compatibility tests are used to prevent incompatible blood transfusion that can cause severe immune-mediated transfusion reactions that endanger the patient's life. Five healthy boas (males and females) from the Laboratório de Herpetologia, with an average weight of 3577.5 g were used as donors. Blood was collected slowly by the boa's vertebral venous plexus with the aid of a 22G scalp and put into suitable pediatric blood bags. The receptors were 10 healthy recently acquired rattlesnakes (five males and five females) with an average weight of 707 g. The rattlesnakes were divided into two groups: experimental group (three males and three females) that received blood transfusion, and control group (two males and two females) that did not receive blood transfusion. The day before the catheter implantation surgery, blood tests were performed to determine the hematocrit value and health state of all the animals. After the catheterization, the volume of blood necessary to lower the hematocrit level to 10% was withdrawn and the same quantity of blood from the donor was transfused to the experimental group only. The vertebral venous plexus was successfully used for the collection of large blood volumes, both in boas and rattlesnakes. The blood transfusion showed considerably significant results in the increase of hematocrit values in the experimental group after the first 24 to 48 hours, when compared to the control group that did not receive blood. Although the donors and receptors belonged to different families, no transfusion reactions were observed.

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Snakes, blood transfusion.

(P24) **Effective plastic reactions of the invasive Mediterranean Painted Frog (*Discoglossus pictus*) in front of two usual native predators**

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In order to assess the real influence of inducible defenses on the population dynamics during biological invasions, it is necessary to explore the impact of this plasticity on the survival of prey. Once tested in previous works that tadpoles of the invasive Mediterranean Painted Frog (*Discoglossus pictus*) react in front of their usual native predators, we conducted a series of predation tests for each predator species to explore the effectiveness of their behavioral and morphological defenses. Tests consisted in leaving three tadpoles with inducible defenses plus three "control tadpoles" and a free ranging predator during 24h in a 30L tank with some refugia. As expected, "induced tadpoles" displayed as inducible defenses in front of both predators a shy behavior and higher tails compared to "control tadpoles". Behavioral changes were very effective confronting *Sympetrum striolatum* (Odonata, Libellulidae), decreasing the number of registered attacks by 54.55%. Even though the predator had a tendency to predate upon tadpoles resembling the control morphology, the proportion of failed attacks did not increase in the induced tadpoles. Concerning *Notonecta glauca* (Heteroptera, Notonectidae), both behavioral and morphological defenses were effective diminishing the number of attacks (84.2% decrease) and the number of effective attacks (15.56% decrease), respectively. Overall, our results highlight the importance of the role of inducible defenses in this system, diminishing tadpole deaths of the invasive frog up to 40% in front of the native predator *Sympetrum striolatum* and up to 86,67% in front of the native *Notonecta glauca*.

Phenotypic plasticity, inducible defenses, *Notonecta glauca*, *Sympetrum striolatum*, predation.

(P25) Diferentes reacciones a sustancias de alarma y kairomonas en renacuajos de sapo partero común

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El riesgo de depredación es una de las presiones evolutivas más importantes a las que se enfrentan las especies, ya que la falta de respuesta a un posible depredador puede ser fatal. Sin embargo, la respuesta antidepredadora debe ser adecuada según el nivel de amenaza, ya que una reacción exagerada puede incurrir en costes excesivos, como la pérdida de oportunidades de alimentación. Dado que diferentes especies (e individuos dentro de las especies) sufren diferentes costes y riesgo de depredación, cada uno hará una asignación de riesgo diferente, lo cual puede implicar tanto una mayor respuesta antidepredadora como una mayor sensibilidad a los indicios de depredación. En los ambientes acuáticos, especialmente en hábitats complejos o aguas turbias, la percepción química (olfato) es una modalidad sensorial crítica, ya que quien sea capaz de reaccionar a los depredadores antes de verlos es más probable que sobreviva. Así, los renacuajos de algunas especies de anfibios no sólo reconocen y reaccionan ante el olor del depredador en sí (kairomonas), sino que también reconocen como un indicio de riesgo de depredación las sustancias de alarma que desprenden sus congéneres heridos. Éstas, al ser detectadas suelen provocar una respuesta antidepredadora similar a la provocada por las kairomonas. El sapo partero común (*Alytes obstetricans*) utiliza para la reproducción una amplia variedad de hábitats acuáticos y algunos renacuajos pasan el invierno en el agua, lo que aumenta sus posibilidades de morir depredados. Por lo tanto, podemos esperar que para los renacuajos de esta especie una mayor capacidad de respuesta a las señales de depredación resulte más ventajosa que costosa. Así, planteamos la hipótesis de que los renacuajos de *A. obstetricans* deberían ser capaces de reconocer y reaccionar a las señales de alarma de la misma especie. Para comprobarlo y examinar si la reacción de los renacuajos está relacionada con el nivel de amenaza, les expusimos a cinco estímulos: agua, sustancias de alarma de un conoespecífico, kairomonas (olor de larva de libélula), el olor de una especie no depredadora (renacuajo de *Rana dalmatina*) y kairomonas y alarma juntas. Como respuesta registramos la actividad de los individuos antes y después de la exposición al estímulo. Predijimos que los renacuajos deberían reaccionar con una respuesta similar a la alarma y a las kairomonas, y con una mayor reacción a los dos estímulos juntos. Nuestros resultados confirman que son capaces de reconocer las sustancias de alarma. Sin embargo, mientras que frente a las kairomonas disminuyen el tiempo moviéndose, se observa la reacción opuesta con las sustancias de alarma. Además, ambos estímulos juntos provocan una reacción intermedia que puede ser debida a la existencia de diferencias inter-individuales (es decir, personalidades).

Alarm cues and kairomones elicit different reactions in midwife toad tadpoles

Predation risk is one of the most important evolutionary pressures faced by species because failure to respond to a potential predator may be fatal. However, antipredatory response has to be appropriate according to the threat level, since an overreaction may incur undue costs, such as loss of feeding opportunities. Since different species (and individuals within species) suffer different costs and risk of predation, each one will make a different risk allocation, which may suppose either an enhanced antipredatory response or an increased responsiveness to the predation cues. In aquatic environments, especially in complex habitats or turbid water, chemical perception (olfaction) is a critical sensory mode since that prey able to react to predators before seeing them is more likely to survive. Thus tadpoles of some amphibian species not only recognise and react in the face of the odour of the predator itself (kairomones), but also recognise the alarm cues released by injured conspecifics as a cue of predation risk. When these alarm cues are detected usually elicit an antipredatory response similar to those elicited by kairomones. The common midwife toad (*Alytes obstetricans*) uses for reproduction a wide variety of aquatic habitats and some tadpoles overwinter in the water, increasing their chances of dying predated. Therefore, we can expect that for tadpoles of this species a higher responsiveness to predation cues must be more advantageous than costly. Thus we hypothesized that *A. obstetricans* tadpoles should be able to recognize and react to conspecific alarm cues. To test this and to examine whether *A. obstetricans* tadpoles reaction is related to threat level, we exposed them to five stimuli: water (control), conspecific alarm cues, kairomones (odour of dragonfly larvae), odour of a non-predator (*Rana dalmatina* tadpole; control) and kairomones + alarm cues. As behavioural response we recorded the activity of individuals before and after exposure to the stimulus. We predicted that tadpoles should react with a similar response to alarm cues and to kairomones, and with a greater intensity to both stimuli together. Our results confirm that *A. obstetricans* tadpoles are able to recognize alarm cues. However, while in face of kairomones tadpoles decrease time spent moving, the opposite reaction is observed with alarm cues. Moreover, both stimuli together elicit an intermediate reaction which may be due to the existence of differences in the response among individuals (i.e. personalities).

Alarm cues, tadpoles, predation, semiochemicals, *Alytes obstetricans*.

(P26) Coloración de anfibios: ¿una respuesta plástica al sustrato?JOSÉ MIGUEL OLIVEIRA¹, NURIA POLO-CAVIA², ALBERTO JOSE REDONDO VILLA³, RAFAEL MÁRQUEZ²¹Av. Dr. Abel das Neves, Ed. Cerca dos Anjos, Bl. 1, 1º B, 3130-204 Soure, Portugal.²Department of Biodiversity and Evolutionary Biology, Spanish National Museum of Natural Sciences, CSIC. José Gutiérrez Abascal 2, 28006 Madrid, Spain.³Department of Zoology, Universidad de Córdoba. 14071 Córdoba, Spain.

El camuflaje es un comportamiento defensivo muy generalizado empleado contra los depredadores y otras amenazas del medio. Por ejemplo, ajustando sus patrones de coloración a las características del hábitat los animales pueden reducir el riesgo de ser detectados y capturados, y de este modo incrementar su eficacia biológica. Si bien la mayor parte de las adaptaciones en la coloración críptica son constitutivas, algunos organismos pueden expresar plasticidad fenotípica en la coloración de la piel. Mostramos una respuesta potencialmente plástica en la coloración del sapo partero común, *Alytes obstetricans*, cuya pigmentación mostró una variación acorde con el color del sustrato. Fotografiamos sapos adultos de una población del Parque de Santa Cruz en Coimbra (centro de Portugal). Mediante análisis cuantitativo de imágenes, comparamos los valores cromáticos medios (luminancia y reflectancia de las componentes RGB) de los sapos y del área de sustrato inmediatamente alrededor. Encontramos una correlación positiva y significativa entre la coloración dorsal de los sapos y el color del sustrato. Estos resultados sugieren que algunas especies de anfibios podrían alterar fenotípicamente la coloración de la piel para ajustarse al entorno que les rodea, beneficiándose así de estrategias de cripsis reversible. Una hipótesis menos probable sería que los sapos son capaces de seleccionar de forma precisa sustratos que se ajustan a su coloración como estrategia antidepredatoria que favorece la ocultación.

Amphibian coloration: a plastic response to backgrounds?

Camouflage is a widespread defensive behavior employed against predators and other environmental threats. For instance, by adjusting their color patterns to habitat features many animals reduce the risk of being detected and captured, thereby improving their fitness. While most adaptations in cryptic coloration are constitutive, some organisms can express phenotypic plasticity in skin coloration. Here we show a potentially plastic response in coloration of common midwife toads, *Alytes obstetricans*, whose pigmentation varied in parallel according to backgrounds. We photographed adult toads from a population occurring in the Santa Cruz Park of Coimbra (central Portugal). Using quantitative image analysis, we compared the average chromatic values (luminosity and reflectance of RGB components) of toads and the area of ground immediately around them. We found a positive and significant correlation between dorsal coloration of toads and ground color. These results suggest that some amphibian species might phenotypically adjust skin coloration to match the surrounding environment, thus benefitting from reversible crypsis strategies. A less supported hypothesis would be that toads accurately select matching backgrounds to improve concealment as an antipredatory strategy.

Alytes obstetricans, amphibians, background matching, coloration, crypsis, phenotypic plasticity.

(P27) **Polychromatic *Podarcis muralis* do not behaviourally hide conspicuousness differences among alternative chromatic morphs**

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The conspicuousness of animal coloration is a relative parameter, which depends on the visual system of the receiver and the environmental context in where they are viewed. The general prediction is that colour signals must evolve to show those chromatic properties that maximize their conspicuousness to primary receivers in their own visual ecosystem. In the ventrally polychromatic (i.e. orange, yellow and white) lacertid *Podarcis muralis*, the use of visual modelling techniques has demonstrated that the different morphs combined with the ultraviolet (UV) ventrolateral spots differ in their conspicuousness; being males sorted in order of conspicuousness as orange, yellow and white. Although these dissimilarities suggest different signal efficacy for each morph (i.e. detectability, discriminability and/or memorability of the ventral coloration and the UV spots), they may be compensated or altered behaviourally.

We quantified the degree of exposure of the lizard ventrolateral and ventral coloration in the field with the aim to confirm that the differences in chromatic contrast may actually result in differences in conspicuousness. We used an exposure classification based on four lizard postures, from a posture in which the ventral surface is completely hidden (posture 1; e.g. when lizards thermoregulate) to a posture in which the exhibition of the conspicuous colour patches is maximized (posture 4; when lizards extend their forelegs and elevate the head, showing the ventrolateral and ventral patches). Controlling for sex, age (categorically assigned by size, body proportions and colour pattern development), colour morph and substrate (because conspicuousness also depend on the chromatic properties of the background in which a colour pattern is displayed), results did not reveal differences in the exhibition of ventral coloration among morphs. Therefore, the different degrees of conspicuousness caused by chromatic contrasts are not attenuated (nor increased) behaviourally in any morph, validating our hypothesis and confirming that the alternative colour morphs incur in different costs and benefits due their detectability by primary receivers of colour signals and other unintended observers (i.e. competitors, predators, prey).

Behaviour, conspicuousness, population polychromatism, signal efficacy, signalling.

(P28) **Morph-specific correlations between immunity, sexual characteristics and personality in male *Podarcis muralis***

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In some lizard species, individuals occur in discrete, stable, genetically based colour morphs (CMs) which coexist at different equilibrium levels within the same population. Theory predicts that colour polymorphism is evolutionarily stable because morphs have exactly equal fitness or a fitness advantage when rare. Different CMs reflect alternative behavioural strategies related with reproduction (e.g. territoriality), which are modulated by complex interactions among environmental pressures in constant change (e.g. social interactions and relative morph density). Thus, the differences in fitness among morphs vary over time. We hypothesize that behavioural strategies reflected by CMs may affect to individual investment in other morphological, behavioural or physiological characteristics (related or not with reproduction) and they may imply different trade-offs or correlations between them. To explore this hypothesis, we measured the following characteristics in pure CMs of male *Podarcis muralis* (orange, yellow and white morphs): sexual signalling investment (number of blue ocelli and phemoral pores), head size (related to male dominance), health status (immune response and parasite load), body condition, and personality (antipredatory and exploratory behaviour). We predict that different evolutionary pressures which promote the persistence of each CM also promote morph-specific correlations between measured male characteristics. We found that CMs differed in head width and immune response (yellow and orange CMs over white in both cases) and number of phemoral pores and blue ocelli (orange and white CMs more than yellow in both cases). Immune response was positively related with body condition in white and yellow morphs, but not in orange ones. In the latter, body condition was positively related to the number of phemoral pores. Boldness to predation risk was positively related to head width in orange males, but negatively related in yellow ones. Finally, yellow males with more ocelli were bolder. Differences detected among CMs and morph-specific correlations support our hypothesis. Results were discussed in the context of different behavioural strategies of each CM.

Colour polymorphism, PHA, personalities, behavioural syndromes, sexual signals, sexual selection.

(P29) Red tails are effective decoys for avian predatorsJOSABEL BELLIURE¹, BELÉN FRESNILLO¹, JOSÉ JAVIER CUERVO²¹Department of Life Sciences, Ecology Section, University of Alcalá. 28271 Alcalá de Henares, Madrid, Spain.²Department of Evolutionary Ecology, Museo Nacional de Ciencias Naturales (CSIC). C/ José Gutiérrez Abascal 2, 28006 Madrid, Spain.

A possible explanation for the presence of conspicuous colouration in juveniles is the decoy hypothesis, which states that such colouration is present in a non-vital part of the body to divert attacks from head and trunk, thus increasing survival probability. To test this hypothesis we made two different plasticine and plaster lizard models: one with red tails and another with dark-and-light striped tails. We based our models on the colour design of *Acanthodactylus erythrurus* hatchlings, which naturally show a dark-and-light striped dorsal pattern and red tail. Lizard models were placed in the field and also presented to captive *Falco tinnunculus*, a common avian lizard predator. The number of attacks and the body part attacked (tail or rest-of-body) were recorded, as well as the number of days until a model was attacked. Our results suggest that models with both colour designs were recognized as prey and attacked at a similar rate, but in the field, red-tailed models were detected, and thus attacked, sooner than striped-tailed. Despite this increase in conspicuousness, red-tailed models effectively diverted attacks to the tail from the more vulnerable body parts, thus supporting the decoy hypothesis. Greater fitness benefits of attack diversion to the tail compared to the costs of increased conspicuousness would explain the evolution and maintenance of red tail colouration in lizards.

Acanthodactylus erythrurus, attack diversion, predator-prey interaction, evolution of signals.

(P30) **¿Son las plantaciones forestales alóctonas trampas ecológicas para los anfibios? Un estudio de campo y experimental con *Lissotriton helveticus***

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Los ambientes antropizados son hábitats simplificados y con presiones selectivas diferentes a las de los naturales que son explotados por especies que aparentemente pueden tolerar o adaptarse a cambios tan dramáticos (diferencias en la presión depredadora, exposición a contaminantes, disponibilidad de alimento, etc.). El proceso de adaptación (evolutivo o plástico) a estos ambientes dependerá de los diversos aspectos fisiológicos y comportamentales que directa o indirectamente puedan verse afectados. El tritón palmeado (*Lissotriton helveticus*) es una especie que a menudo aparece en monocultivos de eucaliptos y pinos. No obstante, se desconoce si estos hábitats y las alteraciones que producen en el medio (liberación de sustancias alelopáticas, acidificación, reducción del número de microhumedales y de la biodiversidad de invertebrados) podrían estar afectando a ésta u otras especies de anfibios. Para evaluar su efecto se examinaron la respuesta inmune y el desarrollo de caracteres sexuales de machos. Resultados preliminares sugerían que pinares y sobre todo eucaliptales afectaban de forma negativa a ambos aspectos. En 2013 se examinaron esos mismos aspectos en alrededor de 20 individuos capturados en cada uno de los seis pinares, seis eucaliptales y seis bosques nativos seleccionados (un total de ~350 individuos), obteniéndose resultados idénticos y muy significativos: un 75% peor en los procedentes de eucaliptales e intermedia en los de pinares. Sin embargo, el efecto de los monocultivos puede ser debido a efectos indirectos (p. ej. escasez de alimento) o directos, consecuencia del estrés provocado por la alteración química del medio acuático. Para examinar esta última hipótesis se mantuvo a 180 machos durante 21 días en nueve mesocosmos: tres de ellos con hojarasca de roble, tres con hojarasca de eucalipto y tres de pino (20 machos por mesocosmos). De nuevo, los resultados fueron similares en el caso de la respuesta inmune, confirmando un efecto directo a través de la alteración de aspectos abióticos del medio acuático. Los efectos detectados, tanto en respuesta inmune como en los mecanismos que sustentan la selección sexual, podrían estar perjudicando a los individuos que habitan las plantaciones forestales, que funcionarían como trampas ecológicas si los animales no son capaces de adaptarse.

Are alien tree plantations ecological traps for amphibians? A field and experimental study with *Lissotriton helveticus*

Anthropized environments are simplified habitats that present different selective pressures from native ones. These environments are exploited by species that can apparently tolerate or adapt to such dramatic changes (i.e., differences on predation pressure, exposure to contaminants, food availability, etc.). The process of adaptation (evolutionary or plastic) to these environments depends on the various physiological and behavioural aspects that may directly or indirectly be affected. The palmate newt (*Lissotriton helveticus*) is a species that often appears in eucalyptus and pine forest plantations. However, it is unknown whether these habitats and associated disturbance (release of allelopathic substances, acidification, and reduction of pond availability and invertebrate biodiversity) could be affecting this or other amphibian species. To evaluate its effect we examined the immune response and development of sexual male characteristics. Preliminary results suggested that pine and especially eucalyptus affect negatively both features. In 2013 these same aspects were examined in 20 individuals captured in each of six native forests, six pine and six eucalyptus plantations (a total of ~ 350 individuals), yielding identical and significant results: individuals performed 75% worse in eucalyptus plantations and intermediately in pine plantations with respect to native forests. However, the effect of exotic plantations may be due to indirect effects (f.e. shortage of food) or direct ones, as a consequence of the stress caused by chemical alteration of the aquatic environment. To examine this hypothesis, 180 male newts were kept for 21 days in nine mesocosms: three of them with oak leaves, three with pine leaves, and three with eucalyptus leaves (20 males per mesocosm tanks). Again, the results were similar in the case of the immune response, confirming a direct effect caused by alteration of aquatic abiotic aspects. The effects detected in both immune response and the mechanisms that underlie sexual selection could be damaging to amphibians that inhabit the forest plantations, which would work as ecological traps if the animals are not able to adapt.

Exotic plantations, palmate newt, ecological traps, immune response, sexual selection, mesocosms.

(P31) Habitat use of multiple populations of *Salamandra algira* along an altitudinal gradientDANIEL ESCORIZA¹, JIHÈNE BEN HASSINE²¹Institute of Aquatic Ecology, University of Girona. Campus Montilivi, 17071 Girona, Spain.²Faculty of Sciences of Tunis, Department of Biology, University Tunis-El Manar. 2092 Tunis, Tunisia.

Salamandra algira is one of the southernmost species in the genus, and most of its ecology remains poorly known. We studied the microhabitat conditions of the sites occupied by several populations of *S. algira* along an altitudinal gradient, and the use of water bodies for reproduction. The microclimate conditions were analyzed at six sites in northern Morocco: one site in Beni Snassen massif (*S. algira spelaea*), two in the Middle Atlas and central Rif mountains (*S. algira splendens*), and three in the western Rif mountains and Peninsula Tingitana (*S. algira tingitana*), where a viviparous population also occurs. The microclimate was characterized using temperature and relative humidity data loggers for a period of two years. We also measured the surface area and depth of the water bodies where we found *S. algira* larvae. Our results showed an autumn-winter reproductive period for all ovoviviparous populations studied. In most of the aquatic habitats examined, larvae were found between November and March, although this period could extend to May at higher altitudes. Larval abundance and size variation did not correlate with water body size or microclimatic conditions. The decrease in the number of larvae per water body coincided with the existence of suitable conditions for post-metamorphic dispersal. *Salamandra algira* occurred in regions with moist conditions (annual average relative humidity greater than 64%) and with mean annual temperatures of 13.6-18.6°C, but populations were largely segregated along a gradient of humidity, with some showing higher and more constant values than others. The viviparous population occurs in a region with maritime influence and greater microclimate stability than the other sites studied.

Viviparity, fire salamander, microhabitat, North Africa.

(P32) **Variación estacional de la distribución a escala local de la lagartija de Valverde, *Algyroides marchi*, en función de sus requerimientos de humedad**

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Algyroides marchi se restringe a las sierras surorientales de la Península ibérica, donde se encuentra típicamente confinada en localidades umbrías y húmedas. Rubio y Carrascal (Biological Conservation 70, 245-250, 1994) mostraron la importancia de una humedad elevada y una baja temperatura en la selección del hábitat de la especie. García-Muñoz y Carretero (Acta Herpetologica 8, 123-128, 2013) mostraron una alta pérdida de agua por evaporación en comparación con *Podarcis hispanica* simpátrica. En esta comunicación se muestra la variación estacional de la abundancia de individuos de *A. marchi* en relación con la variación estacional de humedad en las laderas de un cuerpo de agua. El histograma en tres dimensiones de la distribución de individuos muestra como la población, dentro de una parcela de muestreo dividida en cuadrículas situada en un tramo de arroyo y sus laderas, se concentra en el periodo estival en el lecho del arroyo, con rocas y agua (típico microhábitat seleccionado por *A. marchi*). Los resultados ponen de manifiesto la sensibilidad de la especie a la humedad.

Seasonal variation of local distribution of the Spanish *Algyroides*, *Algyroides marchi*, according to its humidity requirements

Algyroides marchi is confined to the southern Iberian mountains, where it is typically limited to shady and humid localities. Rubio and Carrascal (Biological Conservation 70, 245-250, 1994) stressed the importance of high humidity and low temperatures in the habitat selection of the species. García-Muñoz and Carretero (Acta Herpetologica 8, 123-128, 2013) showed high evaporative water loss compared to sympatric *Podarcis hispanica*. In this communication I show seasonal variation of abundance of individuals of *A. marchi* with distance to water, as a function of the humidity variation of the slopes around a water body. A three dimensional histogram shows how the population within a squared sample plot established on a stream and its slopes, concentrates in summer in the stream bed, with blocks, rocks and water (typical preferred microhabitats of *A. marchi*). The results highlight the sensibility of the species to humidity.

Algyroides marchi, lacertids, hydric requirements.

(P33) How sex modifies spatial patterns? Home ranges of a lizard community of *Darevskia* sp. in ArmeniaNEFTALÍ SILLERO¹, CLAUDIA CORTI², MIGUEL A. CARRETERO³

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In order to elucidate the function of lizard home ranges (HR) in the context of reproductive interactions, we analysed for the first time a sympatric community of *Darevskia* rock lizards, located at Kuckak (Armenia), composed of a bisexual species (*D. valentini*), two parthenogenetic species (*D. armeniaca* and *D. unisexualis*), and the two hybrid forms between them. We marked the lizards with coloured inks for visual identification and georeferenced each lizard's position with a Trimble GPS receiver with a horizontal error around 10 cm after post-processing. We estimated HR areas using Minimum Complex Polygon and 95% of the locations for those individuals with five or more sightings. We also investigated the eventual influences of lizard species and morphology on home range size, perimeter, and total travelled distance. Finally, we counted the number of individual HR intersecting a given HR and those individuals without any overlap. We captured 32 individuals and recaptured 26 with 149 locations. We could calculate HR for 17 individuals with 123 records. The bisexual *D. valentini* was the species with the largest home ranges and travelled distances, and with more intersections as well. There were no significant differences between both unisexual species and hybrids in any comparison. HR size and perimeter were related to male morphological characteristics. Contrarily to a previous study, we found unisexual species with smaller home ranges and with less overlaps, likely because presence of bisexual species makes sexual reproduction available and thus induces parthenogenetic females to compete among them.

Reptiles, parthenogenesis, geographical information systems, spatial statistics, minimum convex polygon.

(P34) Movimentos e conectividade de um sistema de charcos utilizado por tartarugas de água doceFILIPE SERRANO¹, PEDRO SEGURADO²¹Universidade de Évora. Largo dos Colegiais 2, 7004-516 Évora, Portugal.²Centro de Estudos Florestais, Instituto Superior de Agronomia, Universidade Técnica de Lisboa. Tapada da Ajuda, 1349-017 Lisboa, Portugal.

A conectividade é uma propriedade central na gestão e planeamento da paisagem para a conservação de espécies que ocupam parcelas de habitat dispersas. O cágado-de-carapaça-estriada (*Emys orbicularis*) é uma espécie de tartaruga de água doce considerada em perigo em Portugal, que mantém importantes populações em diversos sistemas de charcos na Costa Sudoeste Alentejana (Portugal). Cerca de 56% dos charcos desta região foram destruídos entre 1991 e 2009 em consequência da intensificação agrícola no perímetro de rega do Rio Mira. Esta destruição teve certamente importantes consequências para o cágado-de-carapaça-estriada, nomeadamente ao nível da perda de conectividade entre charcos de todo o sistema da região. Uma população de *E. orbicularis* que ocorre num sistema de charcos nas imediações de uma pequena localidade (Longueira-Almogrove) tem sido regularmente amostrada desde 2003. Um total de 595 capturas, totalizando 205 indivíduos, foram realizadas durante este período. Os dados de movimentos entre lagoas foram usados para estimar a probabilidade de movimentos entre lagoas, separadamente para fêmeas, machos e juvenis. A relação entre essas probabilidades e três medidas de distância entre lagoas – distância linear, de menor custo e de custo acumulado – foi testada através de testes de Mantel. As probabilidades de movimento tenderam a apresentar uma maior correlação com a distância de custo acumulado, apesar de se ter encontrado uma relação significativa também com a distância linear. Uma proporção de movimentos sucessivos entre lagoas diferentes foi significativamente superior nas fêmeas. A variação da probabilidade de movimentos com a distância foi modelada através do ajustamento de uma função exponencial negativa. Esta função foi utilizada para prever a probabilidade de movimentos entre todos os charcos do sistema, incluindo charcos não amostrados ou sub-amostrados. Diversos atributos baseados em grafos espaciais foram estimados, definindo diferentes probabilidades críticas para estabelecer diferentes graus de conectividade entre charcos. Esta metodologia permitiu ordenar a importância dos charcos em termos da sua contribuição para a conectividade funcional global do sistema para *E. orbicularis*.

Interpond movements and connectivity of a pond system used by freshwater turtles

Connectivity is currently a central issue in landscape management and planning for the conservation of wildlife species occupying scarce habitat patches. The European pond turtle (*Emys orbicularis*) is an endangered species in Portugal that has important populations in several pond systems in the SW Alentejo region of Portugal. About 56% of the ponds of this region have been destroyed from 1991 to 2009, primarily as a consequence of the agricultural intensification. This destruction possibly have had severe consequences for the European pond turtle, namely in the interpond connectivity of the overall pond system. A population of *E. orbicularis* living in a pond system in the surroundings of a small locality (Longueira-Almogrove) has been regularly surveyed since 2003. A total of 595 captures, totaling 205 individuals, were performed during this period. The data on interpond movements were used to estimate movement probabilities among pairs of ponds separately for females, males and juveniles. The relationship between these movement probabilities and three measures of interpond distances – linear, least cost and accumulated cost distance – were tested through Mantel tests. Movement probabilities tended to be more correlated with accumulated cost distance estimates, although a significant relationship with linear distances was also found. Females had a higher proportion of successive movements between different ponds than males. The variation of the movement between pairs of ponds with distance was modelled by fitting a negative exponential function. This function was used to predict the interpond movement probabilities among all the ponds of the system, including non-sampled or undersampled ponds. Based on these probabilities, defining several alternative threshold probabilities to settle different degrees of connection among ponds, several graph-based attributes were then estimated in order to rank the ponds in terms of their importance to the overall functional connectivity for the European pond turtle.

Movement, connectivity, *Emys orbicularis*, graph analysis, least cost distance.

(P35) **Movimentos e actividades de postura do cágado-de-carapaça-estriada num sistema de charcos no Sudoeste Alentejano, Portugal**

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Desde 2009, a Associação Nacional de Conservação da Natureza, Quercus, tem vindo a promover uma acção de gestão de um sistema de charcos de carácter temporário no Sudoeste Alentejano para a conservação do cágado-de-carapaça-estriada (*Emys orbicularis*). Este sistema de charcos encontra-se em grande parte incluído numa propriedade privada dedicada predominantemente à criação de gado. Esta acção envolve um acordo com o proprietário que garante boas práticas agrícolas e de criação de gado, nomeadamente sob o ponto de vista da conservação dos cágados. A acção mais relevante foi a instalação de uma vedação em torno de dois importantes charcos, por forma a impedir o acesso do gado. Um aspecto importante que deveria ser considerado neste esforço de conservação refere-se à protecção dos locais de postura dos cágados. Para este propósito, em 2003, foi realizado um programa de radio telemetria para estudar os movimentos e actividades de postura de tartarugas fêmeas. O principal objectivo deste estudo foi o de identificar e caracterizar os principais locais de postura. Foram colocados transmissores de rádio em 10 fêmeas adultas e todas as frequências individuais foram controladas duas vezes por dia de Junho a Agosto. Um total de nove eventos de comportamento de postura foram detectados, 6 dos quais foram confirmados como locais de postura. A distância dos locais de postura ao corpo de água mais próximo variou entre os 20 e os 1110 metros. Os movimentos totais de fêmeas variaram entre poucas centenas de metros (no mesmo charco) até cerca de 3000 metros (4 charcos utilizados e diversas valas). O movimento mais longo detectado exclusivamente em terra foi de cerca de 400 metros. Não foi observado nenhum comportamento de agregação de fêmeas na área de estudo. Todos os locais de postura foram encontrados em zonas abertas, predominantemente de pastagem usadas por gado. No final da época de reprodução, à medida que os charcos secavam, as fêmeas tenderam a mover-se para uma pastagem de regadio, com um sistema de valas associado, onde os cágados encontraram refúgio. Este estudo forneceu dados preliminares sobre os movimentos individuais de fêmeas ao longo do período reprodutor. De futuro, é necessário recolher mais dados sobre o movimento das tartarugas, nomeadamente ao longo de todo o ciclo de actividade e estendido aos machos adultos, para apoiar medidas de gestão efectivas para a conservação desta importante população.

Movements and nesting activity of the European pond turtle in a pond system in SW Alentejo, Portugal

Since 2009, the National Association for Nature Conservation, Quercus, has been promoting a management action for the conservation of the European pond turtle, *Emys orbicularis*, in a system of ponds located in SW Alentejo, Portugal. This system is located within a private property mainly dedicated to livestock farming. This action involves an agreement with the land owner to ensure good agriculture and livestock practices, namely from the point of view of turtle conservation. A main action involved the installation of fences surrounding two important ponds, in order to prevent access from the livestock. An important aspect that should be taken into account in this conservation effort is the protection of the turtle main nesting sites. For this purpose, in 2003, a radio telemetry program was conducted to study the movements and nesting activity of female turtles. The main objective of this study was to identify and characterize the main nesting sites. Ten females were fitted with transmitters and all individual's frequencies were searched twice a day from early June to August. A total of nine nesting behaviour events were identified, but only six were confirmed as effective nesting sites. The distance of nesting sites to the nearest pond varied between 20 to 110 m. Total movements of tagged females ranged from a few hundreds of meters (same pond) to over 3000 meters (4 ponds used and several ditches). The maximum movement exclusively through land was approximately 400 m. No aggregating behaviour of females seems to occur in the study area. All nesting sites were found in open areas dominated by pastures used by the cattle. At the end of the reproductive season, as the ponds dried up, females tended to move to an irrigated pasture, with an associated ditch system. This study provided preliminary data on individual female turtle's movements along the reproductive season. Further data on turtle movement activity along the whole activity cycle and extended to adult males are needed to support effective management actions for the conservation of this important population.

Conservation, *Emys orbicularis*, movements, nesting sites, telemetry.

(P36) **De que maneira a localização do ninho influencia o sex-ratio das posturas de *Chelonia mydas* em Poilão, Guiné-Bissau?**

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O sexo das tartarugas marinhas é determinado pela temperatura de incubação. Estes animais possuem um padrão “macho-fêmea”, onde temperaturas de incubação mais baixas produzem machos e temperaturas mais altas produzem fêmeas. A temperatura pivotal produz uma proporção equilibrada entre os sexos e varia entre espécies e entre populações da mesma espécie. As temperaturas pivotais estimadas em tartarugas marinhas estão descritas entre 29-30°C. As praias de nidificação podem apresentar várias zonas térmicas que podem influenciar o sex-ratio. Este trabalho teve como objectivo determinar a influência da localização do ninho (exposto, na floresta e na transição entre os dois habitats) no sex-ratio dos ninhos de Tartaruga-verde (*Chelonia mydas*). O estudo decorreu na ilha de Poilão (10°52'N, 15°43'W), Guiné-Bissau, entre 18 de Agosto e 22 de Novembro de 2013. Foram monitorizadas as temperaturas de incubação, de 45 ninhos, através de “loggers” e determinados os sexos das crias ($5 \pm 0,84$ por ninho) através da análise histológica das gónadas. O período de incubação variou consoante o local do ninho, sendo mais longo em zonas de floresta. Nas zonas expostas, 92% dos ninhos produziram um sex-ratio 100% de fêmeas e todos os ninhos em zonas de floresta produziram um sex-ratio 100% de machos. Em Poilão o local do ninho tem influência no sex-ratio da população de *C. mydas*, uma vez que a produção de machos depende do ensombramento proporcionado pelas zonas de floresta. Com o aquecimento global e sendo as tartarugas marinhas espécies ameaçadas mundialmente, a conservação da floresta litoral terá uma elevada importância para o equilíbrio dos sex-ratios destas populações de tartarugas.

How does the location of the nest influences the sex-ratio of *Chelonia mydas* in Poilão, Guinea-Bissau?

The sex of turtles is determined by the incubation temperature. These animals have a “male-female” pattern in such a way that lower incubation temperatures produce males and warmer temperatures produce females. The pivotal temperature produces a balanced sex-ratio and varies among species and among populations of the same species. Pivotal temperatures for marine turtles are described between 29-30°C. The nesting beaches may have several zones with different temperatures each, which may influence the sex-ratio. This study aimed to determine the influence of the location of the nest (an area exposed to the sun, a sheltered area in the forest, or a transition between the two habitats) in the sex-ratio of the nests of Green Turtle (*Chelonia mydas*). The study took place on the island of Poilão (10°52'N, 15°43'W), Guinea-Bissau, between 18th August and 22nd November 2013. Incubation temperatures of 45 nests were monitored by loggers, and the sex of the offspring of these nests was determined by histological analysis of the gonads (5 ± 0.84 hatchlings per nest). The incubation period varied depending on the location of the nest, being longer in forest areas. In exposed areas, 92% of the nests produced a sex-ratio of 100% females and nests on forest produced a sex-ratio 100% males. In Poilão the nest site has an influence on the sex-ratio of the population of *C. mydas*, since the production of males depends on the shadowing provided by the forest areas. Considering global warming and being a globally endangered sea turtle species, conservation of coastal forest has a high importance for the balance of the sex-ratios of this turtle populations.

Incubation temperature, sex-ratio, habitat, *Chelonia mydas*, Guinea-Bissau.

(P37) **¿Competencia o segregación? Caracterización del nicho de *Calotriton asper*, *Salamandra salamandra* y *Salmo trutta* en la Serra del Cadí**

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Se prospectaron un total de 102 torrentes en la vertiente norte de la Serra del Cadí. De éstos, 60 presentan un periodo de estiaje que impide la presencia de la trucha común y tritón pirenaico. De los 42 cursos de agua que presentan agua todo el año en 41 se detectó la presencia de *Salmo trutta* o *Calotriton asper*. Únicamente en el 17,1% de los torrentes cohabitan las dos especies. *Salamandra salamandra* ocupa la mayoría de los torrentes al tener un ciclo larvario corto. Sin embargo, es mucho menos abundante en los torrentes en los que se encuentra la trucha común. Para observar la segregación o la posible competencia entre las tres especies se eligieron dos torrentes de vertiente norte de la Sierra del Cadí (Vall de Pi y Vall d'Inglà). En el primer torrente coexisten el tritón pirenaico y la Salamandra común en su fase larvaria. En el segundo, *Calotriton asper* coexiste con la trucha común (*Salmo trutta*). Se ha analizado el nicho espacial, temporal y trófico de tres especies que cohabitan en los dos torrentes. *Salmo trutta* desplaza espacial y tróficamente a los adultos de *Calotriton asper* y depreda sobre las larvas de *S. salamandra* y *C. asper*. Los resultados indican que las larvas de *S. salamandra* se segregan espacialmente con *C. asper* tanto en fase adulta como larvaria. Las mayores densidades de la salamandra se observan en las zonas donde *C. asper* es más escaso. Se ha observado que las larvas de *S. salamandra* compiten tróficamente por los mismos recursos que *C. asper* tanto a nivel taxonómico como de tallas de presa.

Competition or segregation? Niche characterization of *Calotriton asper*, *Salamandra salamandra* and *Salmo trutta* in Serra del Cadi

102 streams were surveyed in the north face of Serra del Cadi. Of these, 60 have a drought period which prevents the presence of brown trout and Pyrenean newt. In 41 of the 42 other streams the presence of *Salmo trutta* or *Calotriton asper* was detected. Only in 17.1% of the brooks the two species coexist. *Salamandra salamandra* occupies most of the streams due to its short larval cycle, but presents low densities in the streams where the trout lives. To observe segregation or potential competition between the three species, two streams of the north face of the Serra del Cadi (Vall de Pi and Vall d'Inglà) were chosen. In the first stream Pyrenean newt and Fire Salamander coexist in their larval stage. In the second one, *C. asper* coexists with brown trout (*Salmo trutta fario*). We analyzed the spatial, temporal and trophic niche of the three species cohabiting two streams. *Salmo trutta* displaces *C. asper* adults and predaes on larvae of *S. salamandra* and *C. asper*. The results indicate that the larvae of *S. salamandra* are spatially displaced by *C. asper* in both its larval and adult stages. The highest densities of salamanders were observed in areas where *C. asper* is scarce. Larvae of *S. salamandra* compete for the same trophic resources than *C. asper* both in size and taxonomic groups.

Calotriton, niche, competence, segregation, *Salamandra*, trout.

(P38) **Potential competition influence on the spatial structure of three lizard communities in Slovenia: a case of *Podarcis muralis* and *Iberolacerta horvathi***

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Life is directly influenced by space. However, how the animal communities use the space has been rarely studied, mainly due to the absence of high resolution spatial information. Nowadays, professional GPS and remote sensing images facilitate to perform these studies at local scale. The analysis of the spatial structure of a species community allows us to understand how individuals share the space and modify their home ranges when together with other species (competitors, predators, prey) or under the influence of different environmental components (light, shelters). The spatial structure of the individuals inside the community can be random, regular, or in clusters. The main aim of this work was to analyse if the presence of a species can modify the space use of another species, and consequently the community' spatial structure. When two species are living together, their distribution structure should be more clustered compared to a single species community, as a consequence of spatial segregation due to competition. If competition is asymmetrical (one species is outcompeting another), only one species should have a more clustered distribution. If the community is composed by only one species, we expect that the spatial distribution will be less clustered, i.e. more regular or random. For this, we studied three lizard communities in Northern Dinaric Mountains, southern Slovenia: two were composed by only one of the species (*Iberolacerta horvathi* or *Podarcis muralis*), and the third community was composed of both species (*I. horvathi* and *P. muralis*). We performed numerous surveys during 28 days (from May to September of 2012). Each survey was a route that we walked from an initial point of the study area and finished in the other side; after a pause of 15 minutes we began the next survey starting from the final point to the initial one. The position of each lizard was recorded with a professional GPS (Trimble GeoExplorer 2008 HX), with an accuracy around 10 cm after post-processing. We applied spatial statistical test to determine the spatial structure of species locations, namely global and local Moran's I, as well as joint pair distances. We recorded 415 *P. muralis* at Mala gora; 11 *P. muralis* and 50 *I. horvathi* at Kuzeljska stena; and 245 *I. horvathi* at Velike Bele stene. The species' records were globally and locally clustered. We present preliminary results focusing in the differences among the three lizard communities.

Lacertids, spatial statistics, GIS, GPS, spatial ecology, interactions.

(P39) Dieta y selección de presa de *Psammodromus algirus* en su límite altitudinal

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Las especies consiguen su máximo nivel de fitness en condiciones ambientales óptimas. En hábitats marginales, como son los ambientes alpinos, la diversidad de presas generalmente es menor, y muchas especies se vuelven peores competidoras, por lo que pueden llegar a afrontar una dieta subóptima con el consecuente impacto negativo en su eficacia biológica. En el presente estudio, analizamos la dieta y la selección de presa de la lagartija *Psammodromus algirus* en su límite altitudinal superior (2200-2500 msnm) para comprobar si su dieta es subóptima en ambientes alpinos, comparada con otros lugares de menor altitud. El análisis de la dieta se basó en muestras de heces recogidas individualmente. Para conocer la disponibilidad de presas, usamos trampas de caída. La selección de presa se estimó comparando la frecuencia consumida de cada tipo de presa -para cada sexo- con la frecuencia de dicha presa en las trampas de caída. La composición de la dieta fue muy similar entre machos y hembras respecto a la abundancia relativa de cada tipo de presa, y el número de presas por muestra fecal tampoco difirió entre sexos. Sin embargo, la amplitud de nicho trófico y la presencia relativa de cada presa fueron mayores en hembras que en machos, consumiendo éstas un mayor porcentaje de cada categoría de presa. Orthoptera, Coleoptera, y Hemiptera fueron las categorías más abundantes en la dieta, seguidas por Larvae y Araneae. Orthoptera y Larvae fueron seleccionadas positivamente (consumo vs. disponibilidad) por la lagartija, mientras que Acarina y Coleoptera fueron activamente rechazadas. Nuestros resultados muestran que *P. algirus* se comporta como una especie generalista en su límite altitudinal superior, y su dieta es similar a las descritas para otras poblaciones mediterráneas, rechazando la hipótesis de una dieta subóptima en un ambiente alpino para esta especie.

Diet and prey selection of *Psammodromus algirus* in its upper elevational limit

Species obtain their maximal fitness in optimal environmental conditions. In marginal habitats, like alpine environments, prey diversity is usually low and some species may be worse competitors, so, they might face a suboptimal diet with negative impact on fitness. In the present study, we analysed the diet and prey selection of the lizard *Psammodromus algirus* in its maximum elevation limit (2200-2500 m asl) to test whether its diet is suboptimal in alpine environments, in comparison to other lowland places. Diet analysis was based on individually collected faecal samples. To assess prey availability, we used pitfall traps. Prey selection was estimated by comparing the frequency of each prey consumed -by sex- with the frequency of that prey in pitfalls. Diet composition was very similar between males and females with respect to the relative abundance of each prey type, and number of prey per faecal pellet did not differ between sexes, either. Nevertheless, trophic niche breadth and relative occurrence of prey were higher in females than in males, with females consuming a higher percentage of each prey category. Orthoptera, Coleoptera, and Hemiptera were the most abundant prey types in diet, followed by Larvae and Araneae. Orthoptera and Larvae were positively selected (consumption vs. availability) by the lizard, while Acarina and Coleoptera were actively rejected. Our findings show that *P. algirus* behaves as a generalist species in its upper elevational limit, and its diet is similar to that reported in other Mediterranean populations, rejecting the hypothesis of suboptimal diet in alpine environments for this species.

Elevational gradient, lacertid diet, maximum elevation limit, *Psammodromus algirus*.

(P40) La dieta de los cocodrilos del oeste de África (*Crocodylus suchus*) analizada con isótopos establesSANDRA NAVARRO¹, JOÃO CAMPOS², XAVIER SANTOS², JOSÉ C. BRITO², CAROLA SANPERA¹¹Departament de Biologia Animal, and Institut de Recerca de la Biodiversitat (IRBio), Universitat de Barcelona. Barcelona, Spain.²CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório Associado, Universidade do Porto. Campus Agrário de Vairão, Rua Padre Armando Quintas 7, 4485-661 Vairão, Vila do Conde, Portugal.

Los análisis con isótopos estables son una herramienta muy útil para reconstruir la dieta de los animales ya que las tasas de los depredadores reflejan las tasas de sus presas. Para las especies de interés para la conservación y con un limitado conocimiento de sus rasgos ecológicos básicos, este método puede ser de gran ayuda para la preparación de los programas dirigidos a proteger sus poblaciones. Hemos analizado los isótopos estables de carbono ($\delta^{13}\text{C}$) y de nitrógeno ($\delta^{15}\text{N}$) de las muestras de *Crocodylus suchus* recogidas en pequeños puntos aislados de agua en el sur de Mauritania. Se recogieron muestras, tanto en gueltas como en tâmoûrts, de escamas de cocodrilos vivos y muertos así como muestras de sus potenciales presas (peces, anfibios, pájaros y mamíferos). Se compararon los $\delta^{13}\text{C}$ y $\delta^{15}\text{N}$ entre los cocodrilos con un Modelo General Linear usando como factores el tamaño de los cocodrilos (no adultos y adultos), la subregión (Tagant y Affolé) y los puntos de agua (guelta y tâmoûrt). El resultado más claro fue un incremento de los valores de $\delta^{15}\text{N}$ con la talla de los cocodrilos en todas las subregiones y los puntos de agua. Este hecho sugiere un cambio ontogénico en la dieta de esta especie, con los cocodrilos adultos alimentándose de presas de un mayor nivel trófico. Las comparaciones de $\delta^{13}\text{C}$ fueron menos claras: los adultos de los tâmoûrts en Affolé presentaron valores de $\delta^{13}\text{C}$ más bajos, sugiriendo un mayor consumo de presas terrestres en esos hábitats. La dieta de los cocodrilos podría estar influenciada por la disponibilidad de los diferentes tipos de presas que a su vez probablemente están relacionadas con el tipo de punto de agua. Son necesarios nuevos análisis para reconstruir la dieta de los cocodrilos de Mauritania según los valores isotópicos de referencia locales e incluyendo el efecto del tamaño del cocodrilo y el tipo de punto de agua. Estas conclusiones serán útiles para proteger los hábitats donde viven los cocodrilos incluyendo la calidad del agua y la presencia de sus presas.

Diet of West African crocodiles (*Crocodylus suchus*) examined with stable isotopes

The analysis of stable isotope ratios is a powerful tool to reconstruct animals' diets since rates of consumers/predators reflect those of their prey. For species of conservation concern and with a limited knowledge of basic ecological traits, this method may be of great help for preparing guideline programs in order to protect their populations. We have examined the stable carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) isotopes for the West African crocodile *Crocodylus suchus* collected in small and isolated water points in Southern Mauritania. Scale samples from living and death crocodiles as well as from their potential prey (birds, mammals, fish and amphibians) were collected in gueltas and tâmoûrts. Isotopes $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were compared among crocodiles by General Linear Models using crocodile size (non-adult and adult), sub-region (Tagant and Affolé) and water body (gueltas and tâmoûrt) as factors. The clearest result was an increase of $\delta^{15}\text{N}$ values from non-adult to adult individuals in all sub-regions and water bodies, suggesting thus an ontogenetic shift on the diet of this species, adult crocodiles foraging on prey species from higher trophic levels. $\delta^{13}\text{C}$ comparisons were less unequivocal: adult crocodiles collected in tâmoûrts at Affolé showed the lowest $\delta^{13}\text{C}$ values, suggesting the use of more terrestrial prey by the biggest individuals in these temporary habitats. Crocodile diet may be influenced by the available prey types that on turn probably vary according to the water-body type. New lab analyses are needed to reconstruct Mauritanian crocodile's diet according to local baseline isotopic values and including the effect of crocodile size and the type of water body (i.e. gueltas and tâmoûrts). These conclusions will be useful to protect the habitats where crocodiles live including the water quality and the presence of their prey.

Stable isotopes, crocodiles, Sahara, desert, trophic ecology.

(P41) **A comparison of the diet of the Smoky Jungle Frog, *Leptodactylus pentadactylus* (Anura, Leptodactylidae), in an urban forest fragment and continuous forest, in Central Amazonia**

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Habitat fragmentation is a major threat for many amphibian populations worldwide, and a growing concern for conservationists in tropical regions. In this study we compared the diet of populations of *Leptodactylus pentadactylus*, a large neotropical anuran, from an urban forest fragment and a continuous forest site, in Central Amazonia. The diet samples were obtained by stomach flushing the individuals on location. Each prey item was measured, identified and categorized by taxa. For each category we determined the number of items, the frequency of occurrence; the relative volume; and the index of relative importance (IRI). The Shannon-Wiener index was used to estimate the trophic niche breadth of *L. pentadactylus* on both areas and the values obtained were compared using a t-test. We used Pearson's correlation to test if the volume of the largest prey consumed related to the body size of the individual. We identified a total of 127 prey items belonging to 18 different categories, being arthropods the main source of food on both areas. The IRI values obtained show that Aranea, Scorpiones and Diplopoda were the most important prey items on the forest fragment, where a small Squamata was also part of the diet. On the continuous forest site Diplopoda, Aranea, and Diptera were the most important prey items. There was no significant difference between the trophic niche breadth values obtained for the two areas, and no relation between the largest prey item consumed and body size. Our results suggest that *L. pentadactylus* is a generalist predator that is able to maintain a diverse diet, even on fragmented habitats.

Amphibia, Arthropoda, Neotropical, habitat fragmentation, generalist, stomach flushing.

(P42) **La reflectancia del mentón en el espectro UV está positivamente relacionado con la infección por parásitos sanguíneos en *Gallotia galloti* (Lacertidae: Gallotiinae)**

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Se sabe que los machos de muchas especies animales tienen señales sexuales secundarias que reflejan en el espectro ultravioleta. Recientemente se ha descubierto que la coloración estructural, tal y como lo es aquella basada en la reflectancia de UV, es tan costosa de producir y mantener como la basada en pigmentos. Concretamente, se sabe que los lacértidos utilizan este tipo de señales para comunicarse con sus conespecíficos. Estas señales son interpretadas por ambos sexos de manera que por ejemplo pueden identificar machos más dominantes o agresivos. Estas señales podrían evitar que machos subordinados entren en conflicto con machos de mejor condición física o podrían ayudar a las hembras a identificar machos de mejor calidad genética. En este contexto, la hipótesis de Hamilton y Zuk predice que los individuos del sexo elegible más brillantes serán sexualmente seleccionados por el sexo opuesto en base a las señales sexuales honestas de los primeros que señalarían sobre su capacidad de resistencia a infecciones parasitarias. Sin embargo, hemos encontrado que los machos más brillantes de *Gallotia galloti palmae* presentan mayor carga de parásitos sanguíneos. Este resultado está en consonancia con la hipótesis del hándicap de la inmunocompetencia que predice un aumento de los niveles de testosterona durante la época de celo, este incremento en los niveles de testosterona tendrá dos efectos antagónicos: por un lado el incremento de los caracteres sexuales secundarios, pero al mismo tiempo un aumento de la susceptibilidad a parásitos. Por tanto, nuestros resultados sugieren que en ciertos sistemas con elevada presión por parasitación, pero alta disponibilidad de alimento y sin cuidados parentales el color de los machos podría estar indicando su capacidad de soportar infecciones elevadas de parasitemia sin sufrir efectos negativos. Sin embargo, falta por entender el papel de la testosterona en relación a la expresión de señales sexualmente dimórficas basadas en UV.

UV-chin reflection is positively related to blood parasite infection in male *Gallotia galloti* (Lacertidae: Gallotiinae) from La Palma, Canary Islands

Males of several species are known to show UV-based secondary sexual signals. In recent years structural signals, such as UV-based signals, are known to be so costly to produce and maintain as pigment-based signals. Specifically, lacertid lizards are known to use these signals on conspecifics which are used by both sexes as messages to detect more dominant or aggressive males. These signals may prevent other males against entering in direct male-male competition or help females to identify a male with good quality genes. In this context, the Hamilton and Zuk's hypothesis predicts that the brighter individuals within the eligible sex would be more often selected by the choosing sex as mates since they are honestly signaling their ability to resist parasite infections. However we found that brighter males of *Gallotia galloti palmae* have higher blood parasite loads. This result is in agreement with the immunocompetence handicap hypothesis which predicts that a high elevation of testosterone levels during the mating season, will lead at the same time to two antagonistic effects; higher expression of secondary sexual traits and an immunosuppression increasing the susceptibility of the individuals to diseases. Therefore, our results suggest that in specific systems with high parasitemia pressure, high food income rate and no parental care male secondary sexual signal expression may indicate the capacity of males to support relatively high parasitemia without suffering from detrimental effects. However, the effect of testosterone on male UV signals remains to be demonstrated.

Gallotia, ultraviolet, sexual selection, host-parasite interaction, Hamilton and Zuk hypothesis.

(P43) Filogenia molecular de los coccidios de reptiles: Algunas respuestas a preguntas evolutivasRODRIGO MEGÍA-PALMA¹, JAVIER MARTÍNEZ², SANTIAGO MERINO¹¹Departamento de Ecología Evolutiva, Museo Nacional de Ciencias Naturales-CSIC. C/ José Gutiérrez Abascal 2, 28006 Madrid, Spain.²Departamento de Microbiología y Parasitología, Facultad de Farmacia, Universidad de Alcalá de Henares. 28871 Alcalá de Henares, Madrid, Spain.

La evolución de los parásitos de reptiles de los géneros *Isospora* y *Eimeria* (s.l.) es prácticamente desconocida. Históricamente, las relaciones evolutivas y la taxonomía de estos organismos se habían basado en su morfología y en el tipo de desarrollo endógeno en el interior de sus hospedadores. Sin embargo, el uso de técnicas moleculares nos ha permitido comparar secuencias de ADN para comprender las relaciones evolutivas entre estos organismos. Nuestra hipótesis evolutiva basada en filogenias moleculares de secuencias del gen 18s de ARNr de parásitos del género *Isospora* infectando reptiles mostró 1) la polifilia de este género, y 2) el origen evolutivo del género *Lankesterella*, parásito sanguíneos de aves, anfibios y reptiles. Por otra parte, los parásitos de reptiles del género *Eimeria* (s.l.) evolucionaron como el grupo hermano de los eimeriorinos que infectan actualmente a mamíferos y aves. Este clado de eiméridos de reptiles es parafilético puesto que engloba tres géneros que difieren en la morfología del ooquiste y en el lugar en el que se desarrollan en el interior de su hospedador específico. En este sentido, aportamos información filogenética que apoya la validez del género *Acroeimeria*.

Molecular phylogeny of coccidia (Apicomplexa: Eimeriorina) in lizards: some answers to evolutionary questions

The evolution of intestinal parasites of reptiles under the genera *Isospora* and *Eimeria* (s.l.) remains largely unknown. Historically, the relationships and taxonomy of these organisms was based on the morphology and the endogenous development of these parasites in the internal organs of their hosts. However, the use of molecular tools allows us to compare DNA sequences to understand the evolutionary relationships among these organisms. Molecular phylogeny of the 18s rRNA gene sequence of parasites of the genus *Isospora* in lizards revealed 1) the polyphyly of the genus and 2) the evolutionary origin of avian, frog and reptile blood parasites of the genus *Lankesterella*. On the other hand, the parasites of the genus *Eimeria* (s.l.) of reptiles evolved as the sister group to the eimeriorina that evolved parasitizing mammals and birds. This clade of reptile eimeriids is considered paraphyletic based on the occurrence of three different genera within it differing in the endogenous development of these reptile-specific taxa. In this respect we here add phylogenetic data to support the validity of the genera *Acroeimeria*.

Parasite, Protozoa, Apicomplexa, *Eimeria* (s.l.), *Isospora*, phylogeny.

(P44) Incendios y parásitos de reptiles: Metodología de estudio de los parásitos como bioindicadoresVICENTE ROCA¹, JOSABEL BELLIURE², XAVIER SANTOS³, JULI PAUSAS⁴¹Departament de Zoologia, Universitat de València. Dr. Moliner 50, 46100 Burjassot, València, Spain.²Departamento de Ciencias de la Vida, Facultad de Biología, Ciencias Ambientales y Química, Campus Científico-Tecnológico de la Universidad de Alcalá de Henares. Carretera Madrid-Barcelona km. 33,600,28271 Alcalá de Henares, Madrid, Spain.³CIBIO, Univ do Porto. Campus Agrário de Vairão, Rua Padre Armando Quintas 7, 4485-661 Vairão, Vila do Conde, Portugal.⁴Departament d'Ecologia Vegetal, Centre d'Investigació sobre la Desertificació (CIDE). Carretera Moncada-Náquera km. 4,5, 43113 Mocado, València, Spain.

Los incendios forestales provocan fuertes cambios en la estructura del hábitat y ello puede influir sobre diversas características del ciclo vital de los reptiles, entre ellas las relaciones parásito-hospedador. Aunque los parásitos representan un amplio componente de la biodiversidad animal y pueden afectar a la condición de los hospedadores y a su dinámica poblacional, raramente se han incluido en estudios acerca de la respuesta de hospedadores vertebrados a perturbaciones causadas por el fuego.

Este estudio se enmarca en el trabajo que se está desarrollando en la provincia de Valencia tras dos incendios de grandes dimensiones que ocurrieron en verano de 2012. En la primavera de 2013 y 2014 se llevaron a cabo muestreos visuales en parcelas no ardidas situadas en la periferia de ambos incendios y en zonas incendiadas situadas a diferentes distancias de la zona no ardida más próxima. El objetivo de este estudio es examinar y tratar de dilucidar si los parásitos gastrointestinales de lacértidos en el ámbito mediterráneo pueden ser bioindicadores de este tipo de perturbaciones. La investigación que se presenta se constituye como pionera en España y en Europa.

Durante los muestreos se capturaron individuos de las dos especies de lacértidos más representativas del sotobosque mediterráneo de la Comunitat Valenciana, la lagartija colilarga, *Psammotromus algirus* y la lagartija cenicienta, *Psammotromus hispanicus*. Tras su captura se procedió a la obtención de heces frescas que se conservaron en alcohol 70% para su análisis parasitológico. Este consistió en la filtración de las muestras, su procesamiento mediante técnicas de sedimentación y flotación y su análisis mediante microscopía óptica.

La hipótesis a testar es la posible diferencia de infección parasitaria de estas dos lagartijas entre tres áreas bien delimitadas: una zona no quemada afín al área original afectada por el fuego; una zona limítrofe con la afectada por el incendio; y una zona en el interior del epicentro del incendio. Los estudios previos relativos a la helmintofauna que albergan estos dos hospedadores sugieren que se podrían encontrar alguna especie de cestodo y algunas especies de nematodos oxiúridos. Además, los primeros análisis llevados a cabo muestran la presencia de al menos una especie de protista. Las fases de vida libre, que pasan por el sustrato antes de llegar al hospedador definitivo o intermediario, pueden servir como bioindicadores de la recuperación del suelo y de la vegetación de la zona afectada por el fuego.

Fires and parasites of reptiles: Methodology study of parasites as bioindicators

Forest fires produce strong shifts in habitat structure, and this fact may influence several life-history traits of reptiles including host-parasite relationships. Although parasites represent a large component of biodiversity and can affect host fitness and population dynamics, they are rarely included in studies regarding vertebrate host responses to disturbances by fires.

This study is part of the research conducted in the Valencia province (Spain) after two big fires occurred in summer 2012. In spring 2013 and 2014, we surveyed reptiles in several sampling points located in unburned and burned areas located at different distances from the fire edge. The main objective of this study is to examine whether gastrointestinal parasites of lizards may be adequate bioindicators of this disturbance in the Mediterranean basin. This is a novel research in Spain and Europe.

Lizards of the two more representative species in Mediterranean woodlands, *Psammotromus algirus* and *Psammotromus hispanicus*, were captured and fresh faeces stored in 70% alcohol, processed using flotation and sedimentation techniques and observed under the microscope.

The hypothesis to be tested is the variation in parasite infection of these two lizards among three well-defined areas: an unburned area similar to the original area affected by the fire; a burned area located near the edge of the fire; and a burned area located in the epicenter of the fire. Previous studies regarding the helminthfauna of both hosts suggest that at least a species of cestode and some species of oxyurid nematodes may be found. Moreover, preliminary analyses of the faecal samples show some species of protists. Parasite free-life stages passing through the substrate before reaching the final or intermediate host can serve as bioindicators of the soil and vegetation recovery in the areas affected by fires.

Forest fires, parasites, lacertid lizards.

(P45) **Diversity of *Hepatozoon* parasites in *Tarentola* Wall geckos of the Mediterranean Basin and Canary Islands**

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Parasites are an important element of ecosystems, influencing the fitness of hosts and thus the dynamics of their interactions. Yet many parasites have received little attention and much of their biodiversity remains to be described or even detected. Such is the case of many members of the phylum Apicomplexa. It is estimated that only 0.1% of the total species of this phylum has been described, despite including some of the most notorious parasites of medical and veterinary importance. Reptiles are hosts to a wide variety of organisms, but their most common hemoparasites are of the apicomplexan genus *Hepatozoon*. Species of *Hepatozoon* have been found around the world infecting all groups of terrestrial vertebrates and a range of hematophagous invertebrates, requiring both these types of hosts to complete their life cycle. The pathogenicity of *Hepatozoon* parasites in the wild is still to be fully assessed, along with their biodiversity and distribution, so that their level of threat and conservation significance can be correctly evaluated. Here we analysed tissue samples of 499 wild individuals of seven species of the Wall gecko genus *Tarentola* across the Mediterranean region and Canary Islands. Using primers specific for the 18S rRNA gene of hemogregarines, we found 22 infected geckos with *Hepatozoon* spp. The results of the phylogenetic analysis showed that most individuals were infected with *Hepatozoon* spp. from two lineages closely related to others previously identified from *Tarentola* and other gecko genera. On the other hand, another of the retrieved sequences clustered within a clade of *Hepatozoon* spp. found in lacertids and skinks, as well as snakes. This finding could constitute a new lineage for gecko hosts, or a dead-end infection (i.e. the parasite infects a host that is not part of its normal life cycle). Finally, a completely new *Hepatozoon* lineage was detected in two *Tarentola* specimens from Morocco. This study highlights that, although a large number of hemogregarines from reptile hosts have already been detected, there is still new diversity to be found. Only after understanding their life cycles, their diversity and their effects on the hosts, will we finally be able to truly assess the role of parasites in ecosystems and take them into account in conservation actions.

Biodiversity, parasite, hemogregarine, 18S rRNA, reptile, Gekkota.

(P46) **Prevalencia y diversidad genética de las infecciones mixtas de parásitos sanguíneos en el galápago leproso (*Mauremys leprosa*)**

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Los parásitos apicomplexos son un grupo de parásitos sanguíneos que incluyen, entre otros, los géneros *Hemoproteus*, *Plasmodium* y *Hepatozoon*. A pesar de que han sido registrados en más de 300 especies de reptiles, los estudios de la prevalencia e intensidad de estos hemoparásitos en reptiles son aún escasos. Los hospedadores vertebrados a menudo se encuentran parasitados por dos o más especies de parásitos sanguíneos, pero pese a esta abundancia, los estudios de los efectos de estas infecciones mixtas son escasos y arrojan resultados no concluyentes. En este trabajo hemos analizado la prevalencia y diversidad genética de la infección por tres géneros de hemoparásitos (*Plasmodium*, *Haemoproteus* y *Hepatozoon*), así como su relación con la condición corporal, valores de hematocrito e inmunidad en individuos de dos poblaciones cercanas de galápago leproso (*Mauremys leprosa*), una especie emblemática de la Península Ibérica sobre la que no existen estudios de parasitismo sanguíneo. Utilizando métodos moleculares, encontramos siete especies diferentes de parásitos infectando a los galápagos, siendo cuatro de ellas de nuevo descubrimiento. Además, se hallaron diferencias significativas en la prevalencia de infección entre las dos localidades de estudio, debidas principalmente a diferencias inter-poblaciones en la infección por *Hepatozoon*. Por último, observamos que la condición corporal de los galápagos no se vio afectada por el tipo de infección hemoparásita; pero sí comprobamos que los individuos con infección mixta fueron los que tuvieron menores valores de hematocrito y mayor respuesta inmune celular.

Prevalence and genetic diversity of mixed infections of blood parasites in the Mediterranean Turtle (*Mauremys leprosa*)

Apicomplexan parasites are a group of blood parasites including, among others, the genera *Hemoproteus*, *Plasmodium* and *Hepatozoon*. Although they have been found in more than 300 species of reptiles, the studies on the prevalence and intensity of these haemoparasites are still scarce. Vertebrate hosts are often parasitized by two or more species of blood parasites but, in spite of this abundance, the studies on the effects of these mixed infections are scarce and do not provide concluding results. In this work we have analysed the prevalence and genetic diversity of the infection by three haemoparasite genera (*Plasmodium*, *Haemoproteus* and *Hepatozoon*), as well as their relationship with body condition, haematocrit and immunity in individuals from two nearby populations of Mediterranean Turtle (*Mauremys leprosa*), an emblematic species from the Iberian Peninsula for which no studies on blood parasitism are known. Using molecular techniques, we found seven different species of parasites infecting the terrapins, four of which were newly discovered. Furthermore, we found significant differences in infection prevalence between the two studied sites, mainly because of inter-population differences in the infection by *Hepatozoon*. Finally, we observed that body condition was not affected by the type of haemoparasite infection, although we confirmed that the individuals showing a mixed infection had the lowest haematocrit values and the highest cellular immune responsiveness.

Haemoproteus, haematocrit, *Hepatozoon*, *Plasmodium*, immune response.

(P47) **Fauna helmíntica de *Trachemys scripta elegans* y *Mauremys leprosa* en el este de España: intercambio potencial de parásitos autóctonos y exóticos**

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El galápagos invasor *Trachemys scripta elegans*, ejerce un impacto negativo directo sobre dos especies de galápagos autóctonos, *Mauremys leprosa* y *Emys orbicularis*, que se encuentran catalogadas como amenazadas. Un efecto indirecto poco estudiado trata sobre la existencia de intercambio de parásitos entre la especie invasora y las autóctonas, y si dicho intercambio podría afectar a la conservación de las especies autóctonas. En un estudio parasitológico previo de 47 individuos de *T. scripta elegans* capturadas en cinco localidades del este de España, donde esta especie coexiste con los galápagos autóctonos, se encontraron tres taxones de helmintos: el digeneo *Telochis solivagus* y el nematodo *Serpinema microcephalus*, aparentemente procedentes de las tortugas nativas, y un monogeneo típico encontrado en las tortugas invasoras, *Neopolystoma orbiculare*, en una localidad. Estos datos sugieren que (1) *T. scripta elegans* podría funcionar como hospedador competente para parásitos autóctonos (efecto “spill-back”), incrementando potencialmente las infecciones de dichos parásitos en las especies autóctonas y (2) *N. orbiculare*, un parásito potencialmente patogénico, podría infectar a las especies autóctonas (efecto “spill-over”), como ya se ha demostrado en cautividad. En el presente estudio se analizaron 10 individuos de *T. scripta elegans* procedentes de dos nuevas localidades (Almenara y Río Vaca) y 10 individuos de *M. leprosa* de una tercera localidad nueva, Moixent. En los galápagos de Almenara se detectó *N. orbiculare* (Prevalencia (P): 10.6%) lo que sugiere que este parásito exótico ha colonizado más de una localidad en la región; además, se halló el digeneo *Telorchis singularis* (P: 35.1%) que, al igual que *N. orbiculare*, es una especie exótica propia de *T. scripta elegans*. En *M. leprosa* solo se encontraron ejemplares de *S. microcephalus* (P: 100%). Estos resultados indican que existe un riesgo potencial de efectos “spill-over” en el área, pero todavía no hay datos que los confirmen.

Las tortugas detectadas en este estudio han sido proporcionadas por la Conselleria d'infraestructures, territori i medi ambient dentro del proyecto LIFE-*Trachemys*.

Helminth fauna of *Trachemys scripta elegans* and *Mauremys leprosa* in eastern Spain: potential exchange of indigenous and exotic parasites

The invasive terrapin *Trachemys scripta elegans* has a direct negative impact on two native terrapins, *Mauremys leprosa* and *Emys orbicularis*, both catalogued as endangered. A poorly studied indirect effect is whether there is parasite exchange between invasive and native turtles, and whether this could have an impact on native terrapin conservation. In a previous parasitological study of 47 individuals of *T. scripta elegans* collected in five localities from eastern Spain, where this species coexists with the native terrapins, three helminth taxa were found: the digenean *Telochis solivagus* and the nematode *Serpinema microcephalus*, likely acquired from native terrapins, and a monogenean typically found in the invasive terrapin, *Neopolystoma orbiculare*, detected in one locality. These data suggest that (1) *T. scripta elegans* could be a competent host for native parasites (“spill-back” effect), potentially increasing the infections of these parasites in native turtles, and (2) *N. orbiculare*, a potentially pathogenic parasite, could infect the native terrapin species (“spill-over” effect), as it has been demonstrated in captivity. In the present study, we analyzed 10 individuals of *T. scripta elegans* from two new localities (Almenara and Río Vaca) and 10 individuals of *M. leprosa* from a third new locality, Moixent. In Almenara we detected *N. orbiculare* (Prevalence (P): 10.6%), thus suggesting that this exotic parasite has colonized more than one locality in the region, and the digenean *Telorchis singularis* (P: 35.1%) which, as *N. orbiculare*, is an exotic parasite species typically found in *T. scripta elegans*. In *M. leprosa* we only detected *S. microcephalus* (P: 100%). Our results indicate that there is a potential risk of “spill-over” in the region, although no data have confirmed it yet.

Turtles used in the present study were provided by the Conselleria d'infraestructures, territori i medi ambient within the LIFE-*Trachemys* project.

Invasive terrapins, Valencia, helminth, parasite exchange.

(P48) *Protopolystoma xenopodis*, um parasita de anfíbios invasores a residir nas ribeiras de OeirasRICARDO RODRIGUES¹, VANESSA PINTO¹, NUNO GONÇALVES¹, RUI REBELO¹, RICHARD TINSLEY²¹Centro de Biologia Ambiental / Departamento de Biologia Animal, Faculdade de Ciências da Universidade de Lisboa. 1749-016 Lisboa, Portugal.²School of Biological Sciences, University of Bristol. BS8 1UG Bristol, United Kingdom.

As invasões biológicas por espécies não-nativas constituem uma das principais ameaças aos ecossistemas naturais e à biodiversidade. Milhares de espécies foram extintas ou estão em risco devido a espécies introduzidas, em resultado de interações directas, competição ou transmissão de parasitas e agentes patogénicos. A maior parte dos animais consegue escapar dos seus parasitas quando são introduzidos em novos habitats, contudo alguns persistem nos seus hospedeiros e podem afectar seriamente as comunidades nativas. Neste trabalho caracterizou-se a parasitofauna da população invasora de *Xenopus laevis* (Daudin, 1802). Oitenta *X. laevis* de duas ribeiras do Concelho de Oeiras foram capturados e examinados para a presença de parasitas. Uma das espécies encontrada foi *Protopolystoma xenopodis*, num total de 114 exemplares. A taxa de infecção foi semelhante em ambos os sexos de *X. laevis*, com 53% dos machos e 60% das fêmeas infectados. Este parasita ocorreu com uma prevalência de 68,75%, uma abundância de 1,49 indivíduos na população e uma intensidade média de 2,78 por hospedeiro infectado nos *X. laevis* amostrados. A diferença entre o número de *P. xenopodis* em machos e fêmeas de *X. laevis* não foi significativa, no entanto as suas dimensões foram superiores nos machos. Existiu também uma correlação positiva entre o comprimento de *P. xenopodis* e o comprimento focinho-urostilo (SUL) dos *X. laevis* fêmeas. As dimensões de *P. xenopodis* estiveram ainda negativamente correlacionada com a época do ano. A presença desta espécie exótica de parasita nas ribeiras de Oeiras faz com que a infecção de anfíbios nativos, como *Pelophylax perezi*, seja uma possibilidade.

***Protopolystoma xenopodis*, a parasite of invasive amphibians residing in Oeiras' streams**

Biological invasions by non-native species constitute one of the leading threats to natural ecosystems and biodiversity. Thousands of species have been extinguished or are at risk due to invasive aliens, as a result of direct interactions, competition or parasite and pathogens transmission. Most of the animals can escape from its parasites when they are introduced into new habitats, however some persist in their hosts and may seriously affect the native communities. This project characterizes the parasite fauna of the invasive population of *Xenopus laevis* (Daudin, 1802). Eighty *X. laevis* from two streams of Oeiras were captured and examined for the presence of parasites. One of the species found was *Protopolystoma xenopodis*, in a total of 114 individuals. The infection rate was similar in both genders of *X. laevis*, with 53% of males and 60% of females infected. This parasite occurred with a prevalence of 68,75%, an abundance of 1,49 individuals in the total population and a mean intensity of 2,78 per infected host in the sampled *X. laevis*. The difference between the number of *P. xenopodis* in males and females of *X. laevis* was not significant. However their sizes were superior in males. There was also a positive correlation between the length of *P. xenopodis* and female *X. laevis*' snout-urostile length (SUL). The dimensions of *P. xenopodis* were negatively correlated with the term of the year. The presence of this exotic species of parasite in Oeiras streams may cause the infection of native anuran, *Pelophylax perezi*.

Protopolystoma xenopodis, parasite, invasive species, *Xenopus laevis*, infection rate, Oeiras.

(P49) Análisis helmintofaunístico de *Salamandra salamandra* (Linnaeus, 1758) en la Sierra de Gredos

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En el curso de investigaciones herpetológicas llevadas a cabo conjuntamente por equipos de las universidades de Salamanca y Valencia, en los meses de julio, agosto y septiembre de los años 1987, 88 y 89, se colectaron 106 ejemplares de *Salamandra salamandra* en la Sierra de Gredos. El examen parasitológico dio como resultado una prevalencia de parasitación helmíntica superior al 90%. No obstante, a pesar del elevado número de hospedadores afectados, la parasitofauna de este urodelo se ha revelado como muy pobre. Sólo se aislaron tres especies vermicianas, dos trematodos con ciclos vitales heteroxenos, *Haematoloechus carbonelli* y *Gorgoderina vitelliloba* y un nematodo monoxeno, *Oxysomatium brevicaudatum*. Los hospedadores proceden de dos enclaves diferentes de la Sierra de Gredos, Laguna Grande y Cinco Lagunas, mostrando una fauna parasitaria significativamente diferente. Sin embargo, no se han encontrado diferencias significativas en el tamaño corporal o el sexo de los hospedadores analizados en ambas localidades. Tampoco se han observado interacciones de ningún tipo entre los tres helmintos aislados. *Oxysomatium brevicaudatum*, que accede a su hospedador directamente por medio de la ingesta de huevos embrionados o de larvas infectivas del suelo, es muy abundante en ambos enclaves. El ciclo vital de *G. vitelliloba* transcurre por medio de un molusco primer hospedador intermediario, habitualmente del género *Pisidium* y, aunque en la Laguna Grande de Gredos ha sido citado *P. casertanum*, nunca se ha registrado Gorgoderidae alguno infectando anfibios en este enclave. Aunque el ciclo biológico de *H. carbonelli* es desconocido, el de una especie congénérica *H. pyrenaicus*, propia de zonas montañosas, transcurre por medio del molusco *Ancylus fluviatilis* como primer hospedador intermediario. Este molusco, relativamente abundante en nuestras prospecciones en Cinco Lagunas, pero no en Laguna Grande, es probablemente, dada la escasez de moluscos dulceacuícolas en la zona, el primer hospedador intermediario de *H. carbonelli*.

Helminthofaunistic analysis of *Salamandra salamandra* (Linnaeus, 1758) from the Sierra de Gredos

During the herpetological research done jointly by teams from the universities of Salamanca and Valencia, in the months of July, August and September in 1987, 88 and 89, 106 specimens of *Salamandra salamandra* from Sierra de Gredos were collected. The parasitological survey showed a helminthic prevalence over 90%. However, in spite of the large number of hosts affected, the parasitofauna of this Urodela has been revealed as extremely poor. Only three parasite worm species were isolated, two trematodes with heteroxenous life cycles, *Haematoloechus carbonelli* and *Gorgoderina vitelliloba* and a monoxenous nematode, *Oxysomatium brevicaudatum*. Hosts analysed came from two different sites in Sierra de Gredos, Laguna Grande and Cinco Lagunas, and had a different parasitic fauna. However, no significant differences were found among localities in the body size or sex of the hosts. We did not detect any type of interaction among the three isolated helminthes either. *Oxysomatium brevicaudatum*, who accesses its host directly through ingestion of infective larvae or embryonated eggs from the soil, is very abundant in both sites. *Gorgoderina vitelliloba* life cycle includes a freshwater snail as first intermediate host, usually of the genus *Pisidium* and, although in the Laguna Grande of Gredos was cited *P. casertanum*, never a Gorgoderidae has been detected parasitizing amphibians in this site. Although the life cycle of *H. carbonelli* is unknown, the cycle of the congeneric mountain species *H. pyrenaicus* has the mollusk *Ancylus fluviatilis* as the first intermediate host. This mollusk, quite abundant in Cinco Lagunas, but not in Laguna Grande, is most probably the first intermediate host of *H. carbonelli*.

Salamandra salamandra, Gredos, Laguna Grande, Cinco Lagunas, helminth parasites.

(P50) Datos comparativos de la helmintofauna de *Bufo spinosus* en varias áreas de la Península Ibérica

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Se han estudiado, desde el punto de vista parasitológico, 98 ejemplares completos de *Bufo spinosus* de distintas localidades de la Península Ibérica, 66 de las Sierras de Gredos y de Francia, 15 del Pirineo Central, 11 de la Sierra de Grazalema y seis de varios enclaves de la Comunidad Valenciana. De las 16 especies parásitas detectadas (seis trematodos, un cestodo, ocho nematodos y un acantocéfalo), los nematodos monoxenos, que ingresan en el hospedador al ser ingeridos del suelo en fase de huevo o de larva infectiva, son los más abundantes, llegando a alcanzar, en el caso de *Oxysomatium caucasicum*, una intensidad máxima de 309 en el hospedador más afectado. En el otro extremo, los trematodos *Haplometra cylindracea*, *Sonsinotrema tacapense* y *Pleurogenoides stromi* y el acantocéfalo *Echinorhynchoides* sp. pueden considerarse parásitos accidentales en este anuro, al haber sido encontrados en un único ejemplar. Sólo el cestodo *Nematotaenia dispar* fue observado en las cuatro regiones prospectadas. Las restantes especies se encontraron en una sola localidad, con la excepción de *O. caucasicum*, que se detectó en la Comunidad Valenciana y en la Sierra de Grazalema. Se ha constatado la existencia de diferencias significativas en el tamaño de los hospedadores y también en la riqueza y la abundancia de los parásitos entre áreas estudiadas. Así, mientras que la mayor riqueza media de helmintos se da en el Pirineo Central, la mayor abundancia media aparece en la Comunidad Valenciana. Respecto a los hospedadores, los mayores ejemplares proceden de la Sierra de Grazalema, seguidos de los de la Comunidad Valenciana, Sierras de Gredos y Francia y, por último, los del Pirineo Central. Sin embargo, no se ha observado correlación alguna entre la talla de estos hospedadores y su abundancia o riqueza helmíntica.

Comparative data of the helminth fauna of *Bufo spinosus* in several areas of the Iberian Peninsula

Parasites of 98 specimens of *Bufo spinosus* from different localities of the Iberian Peninsula were analyzed, 66 from the Sierra de Gredos and Sierra de Francia, 15 from the Central Pyrenees, 11 from Sierra de Grazalema and six from several locations in the Valencian Community. Out of the 16 detected parasite species (six trematodes, one cestode, eight nematodes and one acanthocephalan), the monoxenous nematodes, which enter into the host by direct ingestion of eggs or infective larvae, are the most abundant, with *Oxysomatium caucasicum* reaching a maximum intensity of 309 in the affected host. On the other side, the trematodes *Haplometra cylindracea*, *Pleurogenoides stromi* and *Sonsinotrema tacapense* and the acanthocephalan *Echinorhynchoides* sp. can be considered as accidental parasites in this amphibian, since they were found in a single host specimen. *Nematotaenia dispar* was the only cestode observed in the four areas of origin of the hosts. The remaining species were found in a single location, with the exception of *O. caucasicum*, detected in the Valencian Community and in the Sierra de Grazalema. We found significant differences among studied areas in the size of the hosts and in the richness and abundance of parasites. So, while the highest mean helminth richness occurs in Central Pyrenees, the highest average abundance is found in the Valencian Community. Regarding the hosts, the largest specimens were found in Sierra de Grazalema, followed by those of the Valencian Community, Sierra de Gredos, Sierra de Francia and finally that of Central Pyrenees. However, we did not find correlation between the size of these hosts and the abundance or richness of helminthes.

Bufo spinosus, helminth parasites, Gredos, Grazalema, Valencian Community, Central Pyrenees.

(P51) **Diversidade diferencial entre o microbiota da pele rã-verde (*Pelophylax perezi*) amostradas num local contaminado e num local de referência**

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A microbiota da pele dos anfíbios é considerada parte do seu sistema imunitário já com provas dadas de inclusivamente poder inibir o fungo patogénico *Batrachochytrium dendrobatidis*, que é reconhecido como estando associado a mortalidade em massa de anfíbios. Contudo, a composição do microbiota pode variar entre locais dependendo de vários parâmetros ambientais. Considerando que alguns contaminantes como os metais, são persistentes, podendo afetar as comunidades bacterianas, e que inclusivamente algumas espécies anfíbias como a rã verde (*Pelophylax perezi*) podem ser encontradas em locais contaminados por metais, é essencial estudar a composição do seu microbiota. Tendo isto em mente, assim como o facto de que as espécies cultiváveis do microbiota da pele podem revelar-se no futuro de extrema importância como profiláticas contra agentes patogénicos, o nosso trabalho teve como objetivo comparar as espécies cultiváveis do microbiota da espécie *P. perezi* proveniente de um local de referência e de um local contaminado. Para tal, sete adultos de *P. perezi*, quatro provenientes do rio Vouga (local não contaminado) e três da ribeira da Pantanha (local contaminado por metais) foram amostradas através de esfregaços da pele dorsal, lateral e ventral, para obter bactérias cultiváveis sendo as culturas inoculadas no local em meio PCA. Os isolados bacterianos de cada local foram obtidos posteriormente e identificados através de métodos moleculares. A presença ou ausência de espécies permitiu agrupar claramente o microbiota da pele entre as rãs do local de referência contra o local contaminado, sugerindo agrupamentos bacterianos características. Do total de 73 bactérias isoladas, 16 espécies foram obtidas a partir do local de referência e 27 espécies do local contaminado. A diversidade mais baixa de bactérias cultiváveis foi observada no local de referência, sendo *Pseudomonas* o género predominante com sete espécies, enquanto no local de referência *Chryseobacterium* foi o género predominante com cinco espécies. Dentro de algumas estirpes das espécies bacterianas isoladas já existem algumas características descritas na bibliografia, tal como a capacidade de inibir o crescimento de *B. dendrobatidis*. As características gerais das espécies identificadas serão posteriormente discutidas, bem como a sua distribuição. No entanto, um resultado imediato a partir deste estudo é que o perfil microbiota cultivável dos locais estudados é bastante diferente.

Differential diversity between skin microbiota of green frogs (*Pelophylax perezi*) from a polluted and a reference site

The amphibians' skin microbiota is considered part of their innate immune system with given evidences that it can even inhibit the pathogenic fungus *Batrachochytrium dendrobatidis*, which is known for being responsible for amphibian massive die offs. Nonetheless, the composition of the microbiota may vary among sites depending on various environmental parameters. Considering that some contaminants such as metals are persistent and that can affect microbial communities, and also that some amphibian species such as the Iberian green frog (*Pelophylax perezi*) can be found in metal contaminated sites, it is essential to study how their microbiota is composed. Bearing this in mind, as well as the fact that cultivable skin microbiota species can prove in the future to be of the utmost importance as prophylactic against amphibian pathogens, our work aimed to compare the cultivable microbiota species of *P. perezi*'s skin from a reference site against a metal contaminated site. For that purpose, seven *P. perezi* adults, four from Vouga river (uncontaminated site) and three from Pantanhas' stream (metal contaminated stream) were sampled for cultivable bacteria through swabbing of their dorsal, side and ventral skin following inoculation onsite in PCA culture media. Bacterial isolates from each site were then obtained and identified by molecular methods. The presence or absence of assigned species allowed to clearly cluster the skin microbiota between the frogs from reference site against the contaminated site, suggesting characteristic bacterial assemblages. From the total of 73 isolated bacteria, 16 species recovered from the reference site and 27 species from the contaminated site. The lower diversity of cultivable bacteria was observed in the reference site with the predominant genus being *Pseudomonas* with seven species while in the contaminated site, *Chryseobacterium* was the dominant genus, with five species. Within some strains of the isolated bacterial species various features have already been described in literature, such as the ability of inhibiting the *B. dendrobatidis* growth. The overall traits of the identified species are further discussed as well as their distribution. Nonetheless, an immediate result from this study is that the cultivable microbiota profile from the studied sites is quite different.

Microbiota diversity, metal contamination, *Pelophylax perezi*, amphibians.

(P52) **Modificações histológicas no epitélio de *Pelophylax perezi* (Seoane, 1885) de locais contaminados com agropesticidas**

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Os agropesticidas são um dos principais contaminantes dos ecossistemas aquáticos. Escoamentos hídricos superficiais contaminam lagoas e charcos temporários, afectando os organismos aquáticos, particularmente anfíbios em estágios de desenvolvimento iniciais.

A pele dos anfíbios contém dois tipos de glândulas exócrinas: glândulas granulares e glândulas mucosas, ambas amplamente distribuídas pelo epitélio. Esta última responsável pela produção de um muco polissacarídeo. Dados científicos confirmam que as glândulas mucosas desempenham um papel importante no transporte transepitelial e na osmoregulação, com o muco libertado a actuar como um mecanismo alternativo contra a perda de água por evaporação. Alterações na morfologia da pele, tais como a produção de melanina excessiva ou redução da camada queratinizada foram já observadas para outras espécies de anfíbios provenientes de áreas contaminadas com pesticidas.

O objetivo deste estudo passou por avaliar diferenças na relação de área / volume de glândulas mucosas de populações naturais de *Pelophylax perezi* expostas a agropesticidas em locais contaminados.

Sete fêmeas adultas de quatro áreas de amostragem foram coletadas. Os adultos foram submetidos a eutanásia com MS222, de acordo com as permissões da DGV (Direcção Geral de Veterinária). Parâmetros morfométricos foram determinados e a pele foi retirada para posterior análise histológica. Pedacos de pele foram fixados em solução de Bouin e processados para inclusão em parafina, utilizando protocolos de rotina. Posteriormente, seções de 6 µm foram cortadas e coradas com várias colorações (p.e. H&E, Azul Alciano e PAS (Ácido Schiff)). Os cortes foram analisados ao microscópio de fluorescência e a medição de cada glândula foi realizada para observar diferenças entre locais.

A análise preliminar dos dados mostra que existe um aumento significativo ($p < 0,05$) na área das glândulas em organismos de áreas contaminadas, quando comparados com o local não contaminado (controlo).

Histological changes on the epithelium of *Pelophylax perezi* (Seoane, 1885) from agropesticide contaminated areas

Agropesticides are one of the major contaminants of aquatic ecosystems. Surface water runoffs contaminate lagoons and temporary ponds, affecting aquatic organisms, particularly amphibians in early development life stages.

Amphibian skin contains two types of exocrine glands: a granular gland and a mucous gland, widely distributed and responsible for the production of a polysaccharide mucous. Scientific data confirm that mucous glands play an important role in transepithelial transport and osmoregulation with the mucous functioning as a mechanism for evaporative water loss. Alterations in skin morphology such as excessive melanin production or reduction of the keratinized layer have already been reported for other amphibian species exposed to pesticide-contaminated sites.

The purpose of this study was to evaluate differences in the area/volume ratio of mucous glands of natural populations of *Pelophylax perezi* exposed to agropesticides within contaminated sites.

Seven adult females from four sampling areas were collected. Adults were euthanized with MS222. Morphometric parameters were determined and skin was removed for posterior histological analysis. Pieces of skin were fixed in Bouin solution and then processed for paraffin-wax embedding using routine protocols. Subsequently, 6 µm sections were cut and stained with several colorations (e.g. H&E, Alcian Blue and PAS). Sections were analyzed at the fluorescence microscope and the measure of each gland was carried out to observe differences between sites.

Preliminary data analysis shows that there is a significant increase ($p < 0.05$) in gland area ratio in organisms from a contaminated area when compared with the control site.

Agropesticides, histology analysis, mucous glands, osmoregulation, *P. perezi*.

(P53) **Variabilidade intrapopulacional na sensibilidade de dois estádios iniciais de *Pelophylax perezi* a três químicos: influência da água do charco**

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Os ecossistemas dulçaquícolas estão sob considerável pressão ambiental, estando os anfíbios entre os grupos grandemente afetados por este distúrbio. A resposta destes organismos a situações de stress é complexa, sendo modulada por vários componentes: as características inerentes à espécie, exigências do ciclo de vida, características individuais, condições ambientais ou composição do ecossistema. Assim, este grupo pode ser muito heterogéneo relativamente à sensibilidade a perturbações ambientais. Deste modo, o propósito deste trabalho foi avaliar a variabilidade intra-populacional na sensibilidade de uma população natural de rã-verde *Pelophylax perezi* a diferentes compostos: NaCl, cobre e o pesticida carbendazim. Três objetivos específicos foram delineados: (i) estimar a variabilidade entre a sensibilidade de massas de ovos a três substâncias; (ii) avaliar a influência do meio ambiente na sensibilidade dos girinos às três substâncias; e (iii) avaliar se existe uma associação entre a sensibilidade aos três compostos. Dezassete massas de ovos foram individualmente expostas a duas concentrações de cada composto. Girinos das mesmas massas iniciais, mantidos em solução de FETAX (controlo) e em água do charco de origem, foram também expostos aos três stressores. No fim dos ensaios, avaliou-se a taxa de mortalidade dos organismos e os efeitos sub-letais: malformações, edemas, comprimento e biomarcadores enzimáticos. Os dados de mortalidade evidenciaram uma sensibilidade diferente entre massas e entre químicos, nas concentrações mais baixas ($p \leq 0,01$). Em relação à influência do meio na sensibilidade, a água do charco e o FETAX também induziram diferenças significativas na mortalidade entre massas ($p < 0,01$), pois os girinos mantidos em água do charco exibiram mortalidade mais elevada ao cobre e mais baixa ao sal. Relativamente ao objetivo (iii), os resultados preliminares sugerem que algumas massas têm tendência para serem mais sensíveis aos três compostos, sugerindo a ocorrência de co-sensibilidade a diferentes químicos. Concluindo, a sensibilidade de uma população pode ser grandemente determinada pela ação combinada entre indivíduo-ambiente. Neste caso, massas diferentes do mesmo sítio mostraram diferenças na sensibilidade aos stressores ambientais. Igualmente, a influência do meio onde os organismos se desenvolveram pode alterar a sua sensibilidade ao longo do ciclo de vida.

Intra-population variability in the sensitivity of two early life stages of *Pelophylax perezi* to three chemicals: influence of the pond water

Freshwater ecosystems are under considerable environmental pressure, and amphibians are among one of the groups greatly affected by such disturbance. The response of these organisms to stress conditions is complex, being modulated by many components: the species inherent traits, life-cycle demands, individual features, environmental conditions or ecosystem composition. Therefore, it can be a very heterogeneous group regarding sensitivity to environmental perturbations. Thus, the main goal of this work was to evaluate the intra-population variability in the sensitivity of a natural population of the green frog *Pelophylax perezi* to different compounds: NaCl, copper and the pesticide carbendazim. Three specific objectives were delineated: (i) to assess the among egg mass variability in sensitivity to the three chemicals; (ii) to evaluate the influence of the environment in the sensitivity of tadpoles to the three chemicals; and (iii) to evaluate if there is an association in sensitivity to the three chemicals. Seventeen egg masses were individually exposed to two concentrations of each compound. Tadpoles from the same initial masses, maintained in FETAX solution (control) and water from the original pond, were also exposed to the three stressors. At the end of the assays, the organisms were checked for mortality rate and sub-lethal effects: malformations, edemas, length and enzymatic biomarkers. Mortality data showed a differential sensitivity among egg masses and within chemicals at the lower concentrations ($p \leq 0.01$). Concerning the influence of media in the sensitivity, pond water and FETAX also induced significant differences in mortality rates among masses ($p < 0.01$) since tadpoles from pond water exhibited higher mortality when exposed to copper, and lower mortality when exposed to salt. Finally, for the objective (iii), preliminary results suggest that some masses tend to be more sensitive to the three compounds, suggesting the occurrence of co-sensitivity to different chemicals. In conclusion, the sensitivity of a population can be greatly determined by the individual-environment interplay. In this case, different masses from the same site showed differences in sensitivity to the environmental stressors. Also, the influence of the medium where the organisms developed could alter their sensitivity along the life cycle.

Amphibia, egg masses, natural pond water, NaCl, copper, carbendazim.

(P54) **Os efeitos combinados de cloreto de sódio (NaCl), temperatura e densidade no desenvolvimento de embriões e larvas da rã verde (*Pelophylax perezi*)**

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A salinização dos sistemas aquáticos afeta muitas espécies de anfíbios em todo o mundo. As fases mais novas do ciclo de vida dos anuros são muito sensíveis à salinização. Tendo esta informação em consideração, este trabalho teve como objectivo a determinação dos efeitos da salinização em embriões e girinos de *Pelophylax perezi*, assim como, a influência da temperatura e da densidade nesses mesmos efeitos. Assim, foram expostos embriões (estádio de Gosner 10-11) e girinos (estádio de Gosner 25) a uma gama de concentrações de cloreto de sódio (0-7,32 g/l para embriões e 0-10,4 g/l para girinos) durante 96 h e com a avaliação de três densidades (1, 3 e 6 organismos por unidade de volume, para os dois estádios de vida). Os dois ensaios foram ainda realizados a duas temperaturas diferentes (23°C e 26°C). Os seguintes parâmetros foram avaliados diariamente: mortalidade, malformações, edemas e comprimento total do corpo. No final dos ensaios, os organismos sobreviventes foram congelados em azoto líquido para posterior análise da actividade de três marcadores bioquímicos: colinesterase, lactato-desidrogenase e glutathione-S-transferase. O valor de LC50 (e limites de confiança a 95%) para as 96 h a 23°C e a 26°C foi calculado: (i) para embriões – 6,62 (6,44-6,80) g/l e 6,75 (6,61-6,89) g/l, respectivamente, e (ii) para girinos – 7,92 g/l e 7,80 (7,51-8,08) g/l respectivamente. Não foram observadas diferenças significativas na mortalidade, comprimento final ou na taxa de malformações, entre densidades, nos dois ensaios. Para o ensaio dos embriões, nas duas concentrações mais altas de NaCl (6,66 e 7,32 g/l), foram registados 53,3% e 93,3% de mortalidade a 23°C e 43,3% e 100% a 26°C. Todos os organismos sobreviventes nas duas concentrações mais altas de NaCl apresentaram malformações severas e na concentração de 6,66 g/l, houve uma taxa de ocorrência de edemas de 85,7% a 23°C e 41,2% a 26°C. Para os ensaios com girinos, nas duas concentrações mais altas de NaCl (8 e 10,4 g/l) registou-se uma taxa de mortalidade de 33,3% and 100% a 23°C e 56,6% e 100% a 26°C. No desenvolvimento geral para os embriões expostos a 6,66 g/l houve uma redução no comprimento total final de 20% a 23°C e de 28,3% a 26°C. Quando expostos a 8 g/l, os girinos exibiram uma redução de 16% no comprimento a 23°C e 22,46% a 26°C. As concentrações (e limites de confiança a 95%) que causaram diminuição do crescimento foram calculadas, apenas para 10 e 20%: EC20: 5,91 (2,69-6,51) g/l, EC10: 4,35 (0,13-5,29) para 23°C e EC20: 9,08 (7,80-13,10) g/l e EC10: 5,12 (4,14-5,50) g/l para 26°C em embriões. Os valores para girinos a 23°C foram: EC20: 9,08 (7,80-13,10) g/l e EC10: 7,05 (6,24-8,41) g/l, e a 26°C EC20: 8,00 (6,03-24,60) g/l e EC10: 3,59 (1,49-4,66) g/l. A análise para os marcadores bioquímicos ainda está a decorrer e será apresentada no poster.

The combined effects of sodium chloride (NaCl), temperature and density on embryonic and larval development of the Iberian green frog (*Pelophylax perezi*)

Salinization of the freshwater bodies affects many amphibian species around the globe. The early stages of the life cycle of anurans are very sensitive to salinization. Accordingly, this work aimed at assessing the effects of salinization in embryos and tadpoles of *Pelophylax perezi*, as well as the influence of temperature and density on such effects. We exposed embryos (Gosner stage 10-11) and tadpoles (Gosner stage 25) to several concentrations of sodium chloride (0-7.32 g/L for embryos and 0-10.4 g/L for tadpoles) during 96 h and under three densities (1, 3 and 6 individuals per volume unit for the two evaluated life stages). Moreover, both assays were performed under two different temperatures (23°C and 26°C). The following endpoints were monitored each day: mortality, malformations, edemas and total body length. At the end of the assays, surviving organisms were frozen in liquid nitrogen for further quantification of the activity of three biochemical markers: cholinesterase, lactate dehydrogenase, and glutathione S-transferase. We derived LC50 (and 95% confidence limits) for 96 h at 23°C and 26°C: (i) for embryos - 6.62 (6.44-6.80) g/L and 6.75 (6.61-6.89) g/L, respectively, and (ii) for tadpoles - 7.92 g/L and 7.80 (7.51-8.08) g/L, respectively. No significant differences were observed in mortality, final length or malformation rate between densities in both assays. In the two highest NaCl concentrations of the embryos' assay (6.66 and 7.32 g/L), we recorded 53.3% and 93.3% mortality at 23°C and 43.3% and 100% at 26°C. All the surviving embryos in the two highest NaCl concentrations showed severe malformations, and at 6.66 g/L edemas occurred in 85.7% at 23°C and 41.2% at 26°C. In the two highest NaCl concentrations of the tadpole assay (8 and 10.4 g/L), we recorded 33.3% and 100% mortality at 23°C, and 56.6% and 100% at 26°C. In embryos exposed to 6.66 g/L there was a reduction in final total length of 20% at 23°C and 28.3% at 26°C. When exposed to 8 g/L, tadpoles exhibited 16% reduction in length at 23°C and 22.46% at 26°C. The concentrations (and 95% confidence limits) that cause 10% and 20% of growth reduction were also calculated as follows: (i) for embryos- EC20: 5.91 (2.69-6.51) g/L EC10: 4.35 (0.13-5.29) at 23°C, and EC20: 9.08 (7.80-13.10) g/L and EC10: 5.12 (4.14-5.50) g/L at 26°C; and (ii) for tadpoles- EC20: 9.08 (7.80-13.10) g/L and EC10: 7.05 (6.24-8.41) g/L for 23° and

EC20: 8.00 (6.03-24.60) g/L and EC10: 3.59 (1.49-4.66) g/L for 26°C. The analysis for the biochemical markers activity is still in progress, and will be presented in the poster.

Salinity, *Pelophylax perezii*, larval development, acute effects.

(P55) Effects of parabens in Perez's frog embryos: apical and biochemical endpointsCARLA QUINTANEIRO, RUI SANTOS, BRUNA TEIXEIRA, MARTA MONTEIRO, AMADEU SOARES

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Parabens are commonly found in surface water due to their large use as preservatives in personal care products. Methylparaben (MP) and propylparaben (PP) are amongst the most used parabens and their toxicity to aquatic organisms is not fully understood. The main objective of this study was to evaluate the effects of MP and PP on embryos of the Perez's frog *Pelophylax perezi*, using apical and biochemical endpoints. Perez's frog embryos were exposed to increasing concentrations of parabens (0.1 – 10000 µg/l) during 168h, following the Frog Embryo Teratogenesis Assay guidelines (FETAX). Hatching rate, malformations and mortality were the apical endpoints assessed and the activity of the enzymes cholinesterase (ChE), catalase and glutathione S-transferase (GST) were the biochemical endpoints measured. MP did not induced malformations and alterations in the hatching of embryos, while PP induced up to 60% malformations and a decrease of 30% in the hatching rate. Concerning biomarkers, MP increased GST activity and PP inhibited ChE, both in a dose-response manner. The present results suggest that MP is less toxic to Perez's frog embryos than PP, since it only affects the activity of GST that might be answering to the stress as a detoxifying enzyme. PP affected both embryos apical and biochemical endpoints, namely at the neurotransmission level that might compromise equilibrium and frog larvae behavior.

Amphibians, biomarkers, embryo malformations, *Pelophylax perezi*, personal care products.

(P56) Efeitos enzimáticos e de crescimento em girinos de *Pelophylax perezi* expostos a indometacinaMATTHEW O. MOREIRA^{1,2}, RUTH PEREIRA³, FERNANDO GONÇALVES^{1,2}, SÉRGIO M. MARQUES^{1,2}¹Departamento de Biologia da Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.²CESAM (Centro de Estudos do Ambiente e do Mar), Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.³Departamento de Biologia, Faculdade de Ciências & CIIMAR - Interdisciplinary Centre of Marine and Environmental Research. Universidade do Porto, Porto, Portugal.

Nos anos mais recentes tem havido um aumento da consciência do risco ambiental que os compostos emergentes representam. Muitos destes compostos permanecem não regulamentados e podem ser encontrados frequentemente em cursos de água, uma vez que o tratamento convencional das estações de tratamento de águas residuais não é capaz de os eliminar eficazmente. Um destes químicos é a indometacina, que é um composto emergente, pertencente à classe farmacêutica e amplamente utilizado em hospitais como princípio ativo de medicamentos anti-inflamatórios. A presença deste composto farmacêutico nos influentes e efluentes já foi registado, revelando que a sua taxa de remoção é insignificante. Assim os organismos presentes nos cursos de água recetores estarão sujeitos a uma exposição contínua e cumulativa a este composto. Considerando que muitas espécies anfíbias enfrentam uma redução do número dos seus efetivos, e que a maioria das massas de água geralmente abrigam espécies anfíbias, este estudo teve como objetivo avaliar os efeitos da exposição de girinos de *Pelophylax perezi* à indometacina. Para atingir este objetivo expuseram-se girinos na fase 20 de Gosner de desenvolvimento a várias concentrações de indometacina (0,136; 0,272; 0,544; 1,088 e 2,176 µg/l) mais dois controlos (FETAX e FETAX+etanol). A exposição foi realizada com quatro réplicas por tratamento, com 10 girinos cada, e acabou assim que todos os girinos no controlo FETAX atingiram a fase de desenvolvimento 25 de Gosner. Os parâmetros avaliados foram o crescimento, a atividade de enzimas antioxidantes (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependente (GPx-Se), total-Glutathione Peroxidase (GPx-total) e Glutathione Redutase (GRed)) e peroxidação lipídica. O crescimento não foi afetado em nenhum tratamento, incluindo o controlo FETAX+etanol. Relativamente às enzimas antioxidantes avaliadas, apesar de não terem sido detetadas diferenças estatisticamente significativas, foram observadas algumas tendências claras para GSTs, com uma diminuição da atividade nas concentrações mais elevadas de indometacina, e GPx-Se, com um aumento da atividade nas concentrações mais elevadas. Estes resultados indicam que a indometacina pode ter potencial para desequilibrar o sistema antioxidante, o qual é essencial para manter a homeostase do organismo.

Growth and enzymatic effects in *Pelophylax perezi* tadpoles exposed to indomethacin

In the most recent years there has been an increase awareness of the environmental risk that emerging compounds represent. Many of these compounds remain unregulated and can be frequently found in water courses since conventional treatments of wastewater treatment plants are not able to efficiently remove them from wastewaters. One of such chemicals is indomethacin, which is an emerging compound belonging to the pharmaceutical class and widely used in hospitals as the active ingredient of anti-inflammatory drugs. The presence of this pharmaceutical compound in urban influents and effluents has already been recorded revealing that the removal rate is insignificant. Thus the organisms present in the receiving water bodies will undergo a continuous and cumulative exposure to this compound. Considering that many amphibian species are facing a decrease in the number of their effectives and that the majority of the water bodies usually harbor amphibian species, our study aimed to assess the effects of the exposure of *Pelophylax perezi* tadpoles to indomethacin. To achieve this purpose, tadpoles in the 20th Gosner stage were exposed to several concentrations of indomethacin (0.136, 0.272, 0.544, 1.088 and 2.176 µg/l) plus two controls (FETAX medium and FETAX+ethanol). The exposure was carried out with four replicas per treatment, each with 10 tadpoles, and ended once the 25th Gosner stage was reached by all tadpoles in the FETAX control. The parameters evaluated were growth, antioxidant enzyme activity (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependent (GPx-Se), total-Glutathione Peroxidase (GPx-total) and Glutathione Reductase (GRed)) and lipid peroxidation. Growth was not affected in any of the treatments including the control with ethanol. As for the antioxidant enzymes assessed, even though no statistical significant differences were detected, some clear trends were observed for GSTs, with a decrease of activity in the higher concentrations of indomethacin, and GPx-Se, with an increase of activity in the higher concentrations. These results indicate that indomethacin may have the potential to unbalance the antioxidant system, which is essential for maintaining the homeostasis of the organism.

Emergent compounds, antioxidant enzymes, *Pelophylax perezi*, amphibians, indomethacin.

(P57) Afecta o diatrizoato de sódio o crescimento e enzimas antioxidantes em girinos de *Pelophylax perezi*?LIANE CARVALHO^{1,2}, RUTH PEREIRA³, FERNANDO GONÇALVES^{1,2}, SÉRGIO M. MARQUES^{1,2}¹Departamento de Biologia da Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.²CESAM (Centro de Estudos do Ambiente e do Mar), Universidade de Aveiro. Campus de Santiago, 3810-193 Aveiro, Portugal.³Departamento de Biologia, Faculdade de Ciências & CIIMAR - Interdisciplinary Centre of Marine and Environmental Research. Universidade do Porto, Porto, Portugal.

Nas décadas mais recentes os anfíbios têm enfrentado um grande declínio nas suas populações. Várias razões podem ser apontadas como responsáveis por este declínio, tais como doenças ou perda de habitat. Uma das causas que não pode ser ignorada é o aumento de xenobióticos no ambiente, que é piorado no caso de xenobióticos sem regulação adequada. Os compostos emergentes tais como o diatrizoato de sódio, um agente contrastante para raio-x, pertencente à classe de compostos farmacêuticos emergentes, são incluídos nesta descrição e os seus efeitos na fauna local são normalmente desconhecidos. Em acréscimo, as estações de tratamento de águas residuais nem sempre conseguem remover este tipo de compostos químicos das águas residuais, contribuindo assim para a sua persistência no ambiente. Desta forma, é importante juntar informação sobre como os organismos serão afetados, nomeadamente os anfíbios. Tendo isto em mente, o nosso estudo teve como objetivo tentar perceber de que forma o diatrizoato de sódio afeta os girinos de *Pelophylax perezi*. Com esse fim, girinos no estágio de desenvolvimento 20 de Gosner, foram expostos a várias concentrações de diatrizoato de sódio (0,003; 0,006; 0,012; 0,024; 0,048 e 0,096 mg/l) mais um controlo FETAX. A exposição foi levada a cabo com quatro réplicas por tratamento, cada uma com 10 girinos no estágio de desenvolvimento 20 de Gosner e terminou assim que todos os girinos do controlo atingiram o estágio de desenvolvimento 25 de Gosner. Os parâmetros avaliados foram o crescimento, a actividade de enzimas antioxidantes (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependente (GPx-Se), total-Glutathione Peroxidase (GPx-total) e Glutathione Redutase (GRed)) e peroxidação lipídica. O crescimento não foi afetado em nenhum dos tratamentos. Relativamente à atividade enzimática, existiram dois diferentes tipos de resposta, com a GRed e a GPx-total a apresentar uma tendência de aumento de atividade com o aumento da concentração do diatrizoato de sódio, enquanto a GSTs e a GPx-Se revelaram uma tendência oposta. Contudo, não foi observada peroxidação lipídica.

Does sodium diatrizoate affect growth and antioxidant enzymes in *Pelophylax perezi* tadpoles?

Over the most recent decades, amphibians have been facing a great decline in their populations. Several reasons can be pointed as responsible for this decline, such as diseases or loss of habitat. One of the reasons that cannot be overlooked is the increase of xenobiotics in the environment which is worst in the case of xenobiotics without proper regulation. Emerging chemicals, such as sodium diatrizoate, an x-ray contrast agent, is included in this description and its effects on freshwater fauna are unknown. Furthermore, the conventional treatments of urban wastewater treatment plants do not remove this kind of chemicals from the wastewaters, contributing for their release into the environment. Thus, it is important to perceive how these compounds will affect aquatic organisms, namely amphibians. Bearing this in mind our study aimed at perceiving the effects of sodium diatrizoate in *Pelophylax perezi* tadpoles. To meet this objective, tadpoles in the 20th Gosner stage were exposed to several concentrations of sodium diatrizoate (0.003, 0.006, 0.012, 0.024, 0.048 and 0.096 mg/l) plus a FETAX control. The exposure was carried out with four replicas per treatment, each with 10 tadpoles, and ended once the 25th Gosner stage was reached by all the tadpoles in the FETAX control. The parameters evaluated were growth, antioxidant enzyme activity (Glutathione S-Transferases (GSTs), Glutathione Peroxidase Se-dependent (GPx-Se), total-Glutathione Peroxidase (GPx-total) and Glutathione Reductase (GRed)) and lipid peroxidation. Growth was not affected in any treatment. As for enzymatic activity two different kinds of response were observed, with GRed and GPx-total showing a tendency to increase their activity with the increase of sodium diatrizoate concentration, while GSTs and GPx-Se revealed the opposite tendency. Nonetheless, no lipid peroxidation was observed.

Emergent compounds, antioxidant enzymes, *Pelophylax perezi*, amphibians, sodium diatrizoate.

(P58) Efeito do benzo[α]pireno nos melanomacrófagos hepáticos de *Physalaemus cuvieri* (Anura: Leptodactylidae)

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No Brasil ocorre a maior diversidade de anfíbios, mas estes animais estão expostos a contaminantes e sujeitos à redução no número de espécies. Dentre os poluentes, os hidrocarbonetos policíclicos aromáticos são gerados da combustão incompleta de matéria orgânica e estão presentes no ambiente destes organismos. Em particular, o benzo[α]pireno (BAP) é um composto mutagênico e carcinogênico metabolizado no fígado e transportado até ele por meio da corrente sanguínea. Neste órgão há células especiais, os melanomacrófagos (MMCs), envolvidas nos mecanismos inespecíficos de defesa e relacionadas com a melanina em seu citoplasma. Nosso objetivo foi avaliar o efeito do BAP nos MMCs do fígado de *Physalaemus cuvieri*. O experimento foi realizado em 36 animais (aprovado pela Comissão de Ética no Uso de Animais, protocolo 094 e RAN/IBAMA/MMA 18573- 1), nos tempos experimentais de 1, 7 e 13 dias, depois os animais receberam a concentração de 2 mg/kg de BAP. Após a eutanásia, fragmentos do fígado foram retirados e incluídos em historesina para análise em microscopia de luz. Os cortes foram corados com Hematoxilina-Eosina e as contagens da pigmentação foram realizadas por meio do programa Image Pro-Plus (versão 6.0). Os resultados mostraram que houve variação da quantidade de pigmento melânico presente nos MMCs apenas em 7 dias, onde houve redução da quantidade da melanina hepática. Na breve exposição de 1 dia e na mais duradoura de 13 dias não houve alteração quando estes grupos são comparados aos seus respectivos controles. Entretanto, quando comparamos todos os tempos experimentais observamos um maior aumento da pigmentação com 13 dias se comparado com 1 e 7 dias, respectivamente. Alguns autores explicam essa redução afirmando que em níveis elevados de poluição, a fagocitose realizada pelos MMCs pode ser prejudicada, dessa forma é mais susceptível ocorrer alterações metabólicas que alteram a função dos MMCs, o que leva a diminuição da melanina presente no MMC quando exposto ao contaminante. Nesse estudo interpretamos que o BAP não altera a área pigmentar ocupada pelos MMCs com a exposição curta de 1 dia, mas efeitos são observados após 7 dias de tratamento, onde essa substância foi reduzida. Por fim, transcorridos 13 dias de experimentos pode ter ocorrido um ajuste fisiológico.

Effect of benzo[α]pyrene in hepatic melanomacrophages of *Physalaemus cuvieri* (Anura: Leptodactylidae)

Brazil presents the highest diversity of amphibians, and these animals are exposed to contaminants and subject to a decrease in the number of species. Among the pollutants, polycyclic aromatic hydrocarbons are generated from the incomplete combustion of organic matter and are present in these organisms' environment. In particular, benzo[α]pyrene (BAP) is a mutagenic and carcinogenic compound metabolized in the liver and transported to it through the bloodstream. In the liver there are special cells, melanomacrophages (MMCs), involved in nonspecific defense mechanisms and associated with cytoplasmic melanin. Our objective was to evaluate the effect of BAP in MMCs of the liver in the species *Physalaemus cuvieri*. The experiment was conducted on 36 animals (approved by the Ethics Committee on Animal Use, protocol 094 and RAN / IBAMA / MMA 18573-1) at experimental days 1, 7 and 13 after the animals received 2 mg/kg of BAP. After euthanasia, fragments of the liver were removed and embedded in historesin for analysis by light microscopy. The sections were stained with hematoxylin-eosin and pigmentation scores were performed using Image-Pro Plus (version 6.0) program. The results presented a variation of the amount of melanin in the MMCs in 7 days, only where there was a reduction in the amount of hepatic melanin. In the brief exposition of 1 day and the longest, lasting 13 days, there were not changes when compared to their respective controls. However, when comparing all experimental times we observed a greater increase in pigmentation in 13 days when compared with 1 and 7 days, respectively. Some authors explain this reduction by stating that with high levels of pollution, phagocytosis by MMCs may be impaired, thus metabolic changes that alter the function of MMCs are more likely to occur, which leads to a decreased quantity of melanin present in MMCs when exposed to the contaminant. In this study we interpreted that BAP does not alter the pigmented area occupied by MMCs by a short exposure of 1 day, but effects are observed after 7 days of treatment, in which the substance has been reduced. Finally, after 13 days of experiment a physiological adjustment that elevated the melanin of the animals exposed to BAP may have occurred.

Melanomacrophages, anuran, benzo[α]pyrene, pigmentation.

(P59) **Estrutura etária e crescimento das populações invasoras da rã *Xenopus laevis* em Portugal: um estudo esqueletocronológico**

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Há duas populações invasoras da rã *Xenopus laevis* em Portugal, nas ribeiras da Laje e de Barcarena, em Oeiras. Aparentemente essas duas populações estão isoladas entre si. Estudou-se a documentada diferença de tamanhos das rãs entre as duas ribeiras, usando a esqueletocronologia para avaliar a estrutura etária, longevidade e crescimento delas. A população da rib^a da Laje está envelhecida, pois as rãs apresentam maiores valores de tamanho corporal, idade e maturação sexual mais tardia nas fêmeas. Na rib^a de Barcarena há uma estrutura etária jovem, com valores menores de tamanho e idade. Isto sugere que Barcarena pode ter boas condições de reprodução, mas piores condições de sobrevivência de adultos, ao contrário da Laje. Aqui a estrutura etária está envelhecida, porventura em resultado da anterior campanha de erradicação de rãs. Porém, tais resultados também poderão ser justificáveis em face das diferenças nas condições ambientais vigentes nas ribeiras.

Age structure and growth of invasive populations of the frog *Xenopus laevis* in Portugal: a skeletochronological approach

There are two populations of *Xenopus laevis* in Portugal, living in two streams, Laje and Barcarena, located in Oeiras. Previous studies have found differences in the size of individuals of both populations. The present work intends to analyze this size difference through the analysis of the age structure, longevity and the growth of these animals, using skeletochronology. Laje's is characterized by an aged population with higher values of individual length, age and longevity. Barcarena's depicted a younger age structure with lower values of individual length, age and longevity. These results suggest that Barcarena may have better conditions for reproduction, but worse conditions for adult survival, while the opposite may happen in Laje. The environmental differences could explain these results, but the aged population of Laje may also be a result of the eradication program. We suggest different approaches for the continuation of the eradication program.

Xenopus laevis, skeletochronology, growth, age structure, longevity.

(P60) Prevención de la entrada accidental de anfibios alóctonos en la importación de planta viva procedente de China

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En septiembre de 2012, el personal de Aduanas del Puerto de Valencia detectó varios ejemplares de anfibios alóctonos (un ejemplar de sapo asiático *Duttaphrynus melanostictus* y tres ejemplares de rana china *Hylarana guentheri*) en el interior de dos contenedores en los que se almacenaban varios miles de bonsáis procedentes de China, y que eran propiedad de una empresa situada en Camarles (Catalunya). El Comité Científico del Ministerio de Agricultura, Alimentación y Medio Ambiente (MAGRAMA) emitió un dictamen de acuerdo con el cual la especie *Duttaphrynus melanostictus* tiene un elevado potencial invasor y recomendó evitar su entrada accidental a través del tráfico de plantas vivas. Por ello, los contenedores fueron retenidos en el Puerto de Valencia. Debido al elevado valor económico de los bonsáis y al grave perjuicio que podía causar a la empresa la pérdida de miles de ejemplares de plantas, el propietario solicitó a la Dirección General del Medio Natural (DGMN) del Departamento de Agricultura, Ganadería, Pesca, Alimentación y Medio Natural (DAAM) de la Generalitat de Catalunya, que intercediera ante el MAGRAMA y la Aduana del Puerto de Valencia para que éstos autorizaran el traslado de los contenedores a las instalaciones de la empresa, lo cual sucedió el 11 de octubre de 2012.

En este trabajo se explica el plan de confinamiento e inspección meticulosa que desarrollaron técnicos de la DGMN, de la empresa pública Forestal Catalana SA, y agentes forestales del Cos d'Agents Rurals durante seis días enteros, el cual resultó en la inspección de unas 20.000 plantas y la localización de cuatro ejemplares de *Hylarana guentheri*, dos vivos y dos muertos. Previamente y con posterioridad a la descarga de los contenedores se efectuaron varias inspecciones, algunas nocturnas, para descartar el escape de cualquier ejemplar. Así mismo, se quiere hacer incidencia en la importancia de la cooperación entre las distintas administraciones para evitar la entrada de especies invasoras de forma accidental, y a la vez evitar un perjuicio colateral a las empresas de importación de plantas vivas, en este caso.

Prevention of accidental entry of alien species of amphibians in live plant imports from China

In September 2012, Port of Valencia's Custom officers found several specimens of alien amphibian species (one Asian toad *Duttaphrynus melanostictus* and three Chinese frogs *Hylarana guentheri*) inside two containers that stored several thousands of bonsais from China, and which were owned by a company located in Camarles (Catalunya). The Scientific Committee of the Ministry of Agriculture, Food and Environment (MAGRAMA) issued an opinion according to which the species *Duttaphrynus melanostictus* has a high invasive potential and recommended to prevent its accidental entry through the traffic of live plants. Therefore, the containers were retained in the Port of Valencia. Due to the high economic value of the bonsai and the serious damage that the loss of thousands of specimens of plants could cause to the company, the owner asked the General Directorate of the Environment (DGMN) of the Department of Agriculture, Livestock, Fisheries, Food and Natural Environment (DAAM) of the Generalitat de Catalunya, to intercede with the MAGRAMA and Customs of the Port of Valencia so that they would authorize the transfer of containers to the premises of the company, which happened on October 11, 2012.

In this poster we explain the confinement plan and meticulous inspection that fauna experts of the DGMN, the public company Forestal Catalana SA, and rangers from Cos d'Agents Rurals developed for six whole days, which resulted in the inspection of about 20 000 plants and the finding of four specimens of *Hylarana guentheri*, two living and two dead. Prior and subsequent to the unloading of the containers several inspections were performed, even at night time, to discard the escape of any specimen. Also, we want to advocate the importance of cooperation between the various authorities to prevent the accidental introduction of invasive species while avoiding collateral damage to the businesses importing live plants, in this case.

Invasive exotic species, customs, import.

(P61) Assessing the vulnerability of species to environmental changes: an approach with amphibians and reptiles

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Climate and land use change is resulting in the geographic shifting of species' suitable environmental conditions. Species might survive this rapid change by shifting their distributions, but in most cases patches of suitable habitats are separated by a matrix of uninhabitable space. Thus, successful shifting will depend on species dispersal abilities, the availability of suitable habitats and the connectivity between them. In this context, amphibians and reptiles are assumed to have low dispersal abilities, and therefore they are among the species that might be more affected by ongoing environmental change. Ecological realized niche models coupled with connectivity analysis provide a valuable approach to assess potential migration corridors accounting for differential landscape permeability and thus allowing evaluate vulnerability to extinction or isolation. Here we describe the use of such a framework to assess the vulnerability of amphibians and reptiles to a rapid change in the distribution and spatial connectivity of suitable areas in Portugal. We build habitat suitability models using seven modelling techniques and with the best five models we produce a consensus model to project the future distribution in two different scenarios of change. We assessed the functional connectivity between the current and future suitable areas. This study is funded by FEDER Funds through Programa Operacional Factores de Competitividade - COMPETE and by National Funds through FCT - Fundação para a Ciência e a Tecnologia, in the context of project FCOMP-01-0124-FEDER-027863 (IND_CHANGE).

Vulnerability, ecological modelling, connectivity, amphibians, reptiles.

(P62) Amphibians in a human-altered wetland landscapes: water matters, even when there is plenty

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Amphibians are one of the most threatened taxa worldwide and it is fundamental to understand the factors influencing their distribution, abundance and diversity in order to protect their populations. Baixo Vouga Lagunar is a very heterogeneous human-shaped wetland in North-western Portugal, encompassing several municipalities, namely Estarreja and Murtosa, where our study was conducted. This region is composed of a mosaic of different habitats, the most representative being: salt marshes, rushes, reed beds, forested areas, Bocage, rice and maize fields. The area comprises several seasonal or permanently flooded habitats, suitable for the establishment of different species of amphibians. The aim of our study was to characterize the distribution, abundance and diversity patterns of amphibians and to identify the factors that influence those patterns in the mosaic of habitats that constitutes the Baixo Vouga Lagunar. Different sampling methods were applied in a standardized way with the aim of detecting amphibians with different ecological features and in different stages of development. We captured 1534 amphibians from 12 different species, which represent about 70% of the species found in Portugal and, therefore, a large proportion of the regional amphibian diversity. Food availability (measured by insect diversity and abundance) revealed to be of particular value to the amphibian community composition. Aquatic habitats are widespread and abundant in the study area, but water-related variables were always included in the models that best explained species richness patterns. Distance to the nearest well was the single variable in the model that best explained amphibian richness pattern. This can suggest that in this region, strongly influenced by human activities, amphibians were able to adapt and benefit from human presence. Amphibian species richness was also positively influenced by presence of temporary water bodies. These results reinforce findings in the Mediterranean region, which reveal the importance of these seasonally available habitats. We conclude by reinforcing the need for conservation of this semi-natural landscape, of natural and human-made water bodies (such as wells), and of the traditional agricultural practices that promote the mosaic of habitats.

Amphibians, anthropogenic habitats, conservation, mediterranean wetlands, natural habitats, temporary.

(P63) Conectividade ecológica de anfíbios sob mudanças climáticas na Mata Atlântica do sul da Bahia, Brasil

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Embora a conectividade de fragmentos florestais ter sido foco de estratégias de conservação ao longo de décadas, análises conjuntas entre as mudanças climáticas e conectividade ecológica ainda exigem uma atenção constante e renovada em relação a manutenção de fragmentos florestais alvos de conservação. No entanto, avaliações realistas dos impactos das mudanças climáticas em animais ectotérmicos tropicais ainda são insuficientes para traçar metas de conservação conclusivas. Nesse sentido, este estudo avaliou os níveis de conectividade de anfíbios anuros entre os fragmentos de Floresta Atlântica no sul do estado da Bahia, Nordeste do Brasil, estimando as condições de deslocamento destas espécies em diferentes cenários de mudanças climáticas previsíveis. O modelo climático de alcance espacial dos fragmentos florestais do sul da Bahia foi construído com base no mapa da distribuição dos atuais remanescentes de Floresta Atlântica do Brasil. O método de modelagem utilizado para gerar os mapas de distribuição previstos para os fragmentos foi o de máxima entropia, o qual foi gerado pelo programa MAXENT, versão 3.3.2. As projeções do modelo foram realizadas com base em sete variáveis climáticas (média anual de temperatura, sazonalidade de temperatura, temperatura média do trimestre mais quente, temperatura média do trimestre mais frio, precipitação anual, precipitação do trimestre mais úmido e precipitação do trimestre mais seco). A conectividade ecológica foi quantificada através do índice de probabilidade de conectividade (dPC), calculado para todos os fragmentos avaliados em relação aos cenários climáticos do presente e do futuro, utilizando o software Conefor 2.6. A distância de dispersão média para anfíbios anuros foi determinada de acordo com a revisão realizada por Smith e Green (2005; *Ecography*, vol. 28, pp. 110-128), onde foi proposta uma distância média estimada de 400 m para as espécies avaliadas. Os resultados obtidos indicaram que as mudanças climáticas previstas poderão gerar uma perda de 53% de área florestada, o que irá corresponder a uma perda de 38% de conectividade ecológica em relação à dispersão da comunidade de anfíbios anuros que habitam a região estudada. Estes dados mostraram que os fragmentos posicionados na parte costeira do sul da Bahia merecem uma atenção especial em futuros planos de conservação, de modo que estes fragmentos possuem a maior proporção de conectividade ecológica entre os demais avaliados. Assim, este estudo contribui para a compreensão de como os padrões espaciais de dispersão de anfíbios podem ser informativos para planejamentos de conservação em escalas regionais.

Ecological connectivity of amphibians under climate changes in the Atlantic Forest of southern Bahia, Brazil

Although the connectivity of forest fragments has received much attention from conservation strategies for decades, joint analyses between climate change and ecological connectivity still require constant attention in order to maintain the forest fragments as conservation targets. However, realistic assessments of the impacts of climate change on tropical ectotherms are still insufficient to draw conclusive conservation goals. In this sense, this study evaluated the levels of connectivity of anuran amphibians among the remaining fragments of Atlantic Forest in the southern state of Bahia, northeastern Brazil, estimating the conditions of displacement of these species in different scenarios predicted by climate changes. The climate model of the spatial ranges of forest fragments in southern Bahia was built based on the distribution map of the current remnants of the Atlantic Forest in Brazil. The modeling method used was the maximum entropy, which was generated by MAXENT software, version 3.3.2. The model projections were based on seven climatic variables (i.e., mean annual temperature, temperature seasonality, mean temperature of the warmest quarter, mean temperature of the coldest quarter, annual precipitation, precipitation of the wettest quarter, and precipitation of the driest quarter). The ecological connectivity was quantified through the probability of connectivity index (dPC), which was calculated for all the fragments evaluated in relation to the current and future climate scenarios, using the Conefor 2.6 software. The average dispersal distance for amphibians was determined according to the review proposed by Smith and Green (2005; *Ecography*, vol. 28, pp. 110-128), where an average distance of 400 m for the assessed species was established. The results indicated that the predicted climate changes may generate a loss of 53% of forested area, which will correspond to a loss of 38% of ecological connectivity in relation to the dispersion of the community of amphibians that inhabits the study area. These data showed that the fragments positioned in the coastal part of southern Bahia deserve special attention in future conservation plans because these fragments have the highest proportion of ecological connectivity among those evaluated. Thus, this study

contributes to the understanding of how the spatial patterns of dispersal of amphibians may be informative for the conservation planning at regional scales.

Amphibians, Atlantic Forest, connectivity, climate changes, conservation planning.

(P64) **Arrojamentos de tartarugas marinhas ao longo da costa continental Portuguesa: padrões de distribuição e indícios de captura acidental**

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A informação sobre as tartarugas marinhas em águas costeiras de Portugal continental tem sido limitada a alguns avistamentos ou a alguns dados não padronizados de arrojamentos, sendo a tartaruga comum (*Caretta caretta*) e a tartaruga de couro (*Dermochelys coriacea*), as espécies mais registadas. Geograficamente, a localização de Portugal continental, na ponta oeste da Península Ibérica, parece ser uma zona importante de passagem tanto para as tartarugas, comuns do Atlântico norte e Mediterrâneo, como para as tartarugas de couro do Atlântico oeste. Este trabalho é a primeira tentativa de realizar uma análise completa dos registos de tartarugas marinhas ao longo da costa continental Portuguesa, utilizando uma base de dados de 36 anos (1978 a 2013) de animais arrojados e capturados acidentalmente/resgatados.

Dos 806 indivíduos registados, foram observadas tartarugas comuns (a mais frequente com 57,1% dos registos), tartarugas de couro (41,8%) e com valores mais residuais tartarugas verdes, *Chelonia mydas* (1,0%) e tartarugas de Kemp, *Lepidochelys kempii* (0,1%). Tendo em conta apenas as duas espécies mais frequentes, a tartaruga comum e a de couro, estas encontram-se presentes durante todo o ano, ao longo de toda a costa continental Portuguesa, no entanto, apresentam padrões de distribuição espacial e sazonal diferentes. O número de registos anuais de tartaruga comum aumentou em anos mais recentes, tendo havido um pico distinto nos anos de 2010 e 2011, sendo que as densidades mais elevadas para esta espécie ocorrem na costa sul durante a Primavera e Verão. No caso da tartaruga de couro, as densidades mais elevadas foram observadas na região Norte/Centro durante o Outono, e na região Sul durante o Verão. Para ambas as espécies, a maioria dos animais registados eram sobretudo juvenis imaturos. A principal causa de morte foi a interação com as artes de pesca, responsável por 42,6% dos registos (44,7% para a tartaruga comum e 47,8% para a tartaruga de couro). Para ambas as espécies, a interação mais frequente, foi com as redes de emalhar/tresmalho (65,6% e 34,7% para a tartaruga comum e de couro, respectivamente). Os resultados demonstram pela primeira vez que as águas costeiras continentais portuguesas são áreas importantes de alimentação para animais juvenis e sub-adultos da tartaruga comum e de couro durante todo o ano. De um modo geral, a abordagem de distribuições geográfica e sazonal das tartarugas marinhas em águas portuguesas, assim como das suas principais ameaças, irá contribuir para que no futuro se estabeleçam prioridades e medidas de conservação para estas espécies em Portugal.

Sea turtle strandings along the Portuguese continental coast: distribution patterns and insights of bycatch

Information on sea turtles in mainland Portugal has been limited to a few sightings or nationwide not standardized information of stranding data, with loggerhead sea turtles (*Caretta caretta*) and leatherback sea turtles (*Dermochelys coriacea*) referred as the most common species. Geographically, the location of mainland Portugal in western Iberia seems to be an important crossing area for many individuals of both loggerheads and leatherbacks moving along Atlantic hemispheres and/or using the Atlantic-Mediterranean passage. This work is the first attempt to perform a comprehensive analysis of sea turtle records along the Portuguese mainland coast, using a 36-year database (1978 to 2013) for stranded and delivered animals.

Out of the 806 individuals recorded, loggerhead turtles comprised the majority of the records 57.1%, followed by leatherback turtles 41.8%, while residual numbers were recorded for the green turtle, *Chelonia mydas* (1.0%) and Kemp's ridleys, *Lepidochelys kempii* (0.1%). Considering both of the most frequent species, loggerhead and leatherback turtles are present off the whole Portuguese mainland coast throughout the year, although clear spatial and seasonal distribution differences were observed. Generally, the annual number of loggerhead records increased, especially in 2010 and 2011, with higher density in the southern coast during the spring and summer months. With respect to leatherbacks, stranding densities were higher in the north-central western coast during the fall and in the southern coast during the summer. For both species, recorded animals were mainly juvenile-immature individuals. Interactions with coastal fisheries were the main cause of death, accounting for 42.6% of the records (44.7% for loggerheads and 47.8% for leatherbacks). The most frequent interactions for both species occurred with set gill/trammel net fisheries (65.6% and 34.7% of the incidental captures records for loggerheads and leatherbacks, respectively). Results show for the first time that waters off the Portuguese mainland coast are important foraging areas for both loggerhead and leatherback turtles in the North Atlantic

region. Overall, by addressing geographical and seasonal sea turtle distributions and their main threats, this study will contribute to establishing future conservation priorities and actions in Portugal.

Caretta caretta, *Dermochelys coriacea*, strandings.

(P65) **Recuperación de los hábitats acuáticos para anfibios en la Red Natura 2000 en la provincia de Cuenca (España)**

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Durante 2012-2013 un proyecto financiado por el Gobierno regional (JCCM) ha sido desarrollado por la Asociación Herpetológica Española (AHE) en las zonas incluidas en Red Natura 2000 de la provincia de Cuenca. Los objetivos del proyecto fueron: 1 - crear un catálogo de los hábitats reproductivos de anfibios; 2 - crear una base de datos con los registros históricos y actuales de las especies que se reproducen en cada hábitat acuático, especialmente para las incluidas en los catálogos regionales y nacionales; y 3 - proponer actividades de rehabilitación y adecuación de puntos de agua, con el fin de recuperar los hábitats existentes y también crear nuevos puntos que permitan la conectividad entre las diferentes poblaciones de anfibios, con énfasis en las especies que se señalan como en peligro o que tienen una distribución limitada en la zona. Para lograr estos objetivos, se procedió a:

Objetivo 1. Visitamos todos los puntos de agua conocidos por los técnicos, guardas, así como el catálogo de fuentes publicado por el Gobierno de la Provincia que incluye el mapeo y el trabajo de inventario en el Atlas de la provincia de Cuenca que se encontraban dentro o en las inmediaciones de la Red Natura 2000. Cada hábitat acuático que se considera adecuado para la supervivencia y reproducción de los anfibios fue caracterizado con el número de especies detectadas y las posibles acciones a tomar. Se recopiló información sobre 344 puntos de agua a través de las zonas incluidas en Red Natura 2000.

Objetivo 2. Hemos recopilado una base de datos con los registros de anfibios en las zonas Natura 2000. Con esta base de datos, se han desarrollado mapas de distribución con el fin de proporcionar toda la información sobre la distribución de los anfibios en el área de estudio. Se detectaron nueve especies de anfibios, 8 anuros y un urodelo.

Objetivo 3. Proponer acciones para evitar o corregir los problemas típicos de estos ambientes acuáticos: 1 - Adecuación de acceso y medidas de evacuación (instalación de rampas) a los hábitats de reproducción artificial como pilones, abrevaderos, balsas de riego. 2 - La rehabilitación de las estructuras tradicionales que han sido dañadas o abandonadas y pueden albergar poblaciones de anfibios. 3 - Adecuación de los cuerpos de agua que sufren eutrofización, la contaminación y / o el pisoteo del ganado, tratando de permitir la supervivencia de los anfibios y también su utilización por el ganado.

Recovery of aquatic habitats for amphibians in Natura 2000 areas of Cuenca (eastern Spain)

During 2012-2013 a project funded by the regional government (JCCM) was developed by the Spanish Herpetological Society (AHE) in Natura 2000 areas of Cuenca province. The aims of the project were: 1 - to create a catalogue of reproductive habitats for amphibians; 2 - to create a database with historical and present records of species breeding in each aquatic habitat, especially for those included in regional and national Red Lists; and 3 - to propose rehabilitation activities focused on water sites, in order to recover existing habitats and also to create new points that allow connectivity between different amphibian populations, with emphasis on those listed as endangered or those with limited distribution in the area. To achieve these objectives we proceeded to:

Objective 1. We visited all water points known by technicians, rangers, and also those listed in the catalogue of fountains released by the Provincial Government including the mapping and inventory work in the Atlas of the province of Cuenca, that were within or in the vicinity of the Natura 2000 network. Each aquatic habitat that was considered suitable for the survival and/or reproduction of amphibians was characterized with the number of species detected and possible actions to be taken were recorded. We collected information about 344 water sites across Natura 2000 areas.

Objective 2. We compiled a database with the amphibian records in the Natura 2000 areas. With this database, we have developed distribution maps in order to provide all information about amphibian distribution in the study area. Nine species of amphibians were detected, 8 anurans and one newt.

Objective 3. Propose actions to avoid or correct the typical problems of these aquatic environments, including: 1 - Adaptation of access and escape measures (installation of ramps) in artificial reproductive habitats as water tanks, watering troughs, or irrigation pools. 2 - Rehabilitation of traditional structures that have been damaged or abandoned and may harbor populations of amphibians. 3 - Adaptation of water bodies that suffer eutrophication, pollution and/or trampling by livestock, trying to allow amphibian survival and also their use by livestock.

Amphibians, recovery, aquatic habitats, Natura 2000 Areas.

(P66) Refugios para anfibios y reptiles en la Alhambra y el Generalife. Resultados preliminares

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El conjunto de la Alhambra y el Generalife alberga una herpetofauna de trece especies (cuatro anfibios y nueve reptiles). En un entorno arquitectónico histórico en el que, dentro de la ordenación a que se ve sometido el monumento y su entorno, se dan actuaciones sistemáticas de retirada de escombros y desbroces de la cubierta de matorral y de herbáceas, la presencia de refugios terrestres en suelo se convierte en un recurso escaso para pequeños vertebrados. Por otra parte, el control de acceso de las visitas y de depredadores como gatos y perros, y la implementación en los últimos años de técnicas de manejo integrado de huertas y jardines, van propiciando la recuperación de las poblaciones perdidas, con extinciones en tiempos recientes de algunas especies, como el lagarto ocelado (*Timon lepidus*). Se han instalado 160 refugios en tríadas de distintos materiales (arcilla, madera y madera con recubrimiento de arpillera) e iguales dimensiones (25 x 12 x 10 cm) con orientaciones distintas (solana vs. umbría) en las inmediaciones de albercas y acequias. Hasta la fecha, se ha detectado ocupación de *Bufo spinosus*, como principal ocupante (99% de los casos a partir de postmetamórficos). Secundariamente, juveniles de *Pelophylax perezi*, *Podarcis hispanica* s.l. y *Natrix maura*, e individuos adultos de *P. hispanica*, *Hemidactylus turcicus*, *Blanus cinereus* y *Tarentola mauritanica*.

Artificial refugia for amphibians and reptiles in the Alhambra and Generalife. Preliminary results

The monument of the Alhambra and Generalife is the home for thirteen herp species (four amphibians and nine reptiles). In this historical architectonic site there are periodical systematic actions to remove rubble and clear scrubs and herbaceous vegetation, and the presence of terrestrial refugia for small vertebrates is a scarce resource. On the other hand, restricted access of visitors and control of predators (cats and dogs), and implementation in recent years of integrated management techniques in orchards and gardens, are encouraging the recovery of lost populations, including recent extinctions of some species, like the ocellated lizard (*Timon lepidus*). We have placed 160 shelters of similar dimensions (25 x 12 x 10 cm) in triads of different materials (clay, wood and wood coated burlap) in different exposures (sunny vs. shaded) in the surroundings of ponds and ditches. So far we have detected *Bufo spinosus*, as the main occupier (99% of cases from postmetamorphic stages), but also juveniles of *Pelophylax perezi*, *Podarcis hispanica* s.l., and *Natrix maura*, and adults of *P. hispanica*, *Hemidactylus turcicus*, *Blanus cinereus* and *Tarentola mauritanica*.

Artificial terrestrial refugia, herps, Alhambra, Generalife, conservation.

(P67) Estado actual de la herpetofauna en la zona española de la Eurociudad Vasca Bayona-San Sebastián

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La Eurociudad Vasca Bayona-San Sebastián se viene conformando como un corredor urbano entre dos estados fronterizos, poblado por unos 600.000 habitantes. La parte española de la Eurociudad, conformada en torno a la conurbación San Sebastián-Irún, compuesta por trece municipios, ronda los 400.000 habitantes, que aglomeran el 55 % de la población de la provincia de Gipuzkoa. Este territorio, situado en la costa cantábrica, está conformado por montañas de baja altitud en la que el uso humano ha modelado un paisaje a base de campiña (prados y cultivos atlánticos) con residuos de los antiguos bosques caducifolios, dominados por el robledal pedunculado, y plantaciones forestales basadas en pinares, desde el último cuarto del siglo XIX. La herpetofauna del territorio está compuesta mayoritariamente por especies europeas de rango más o menos extendido por el continente, algunas de ellas occidentales o presentes sólo en Francia y España. De las 21 especies actualmente constatadas, nueve corresponden a anfibios y doce a reptiles. La mayor parte de ellas son características del medio humanizado de la campiña, y están presentes en el ámbito forestal y en el ecotono de los bosques y los prados, viviendo con frecuencia a expensas de un ambiente rural transformado por la actividad secular humana. Los bordes de prados, los setos, los matorrales y pastizales que han sustituido a los bosques y las construcciones humanas constituyen el hábitat preferente del 80% de las especies, algunas de las cuales se encuentran catalogadas (entre otras, *Triturus marmoratus*, *Bufo calamita*, *Hyla meridionalis* y *Zamenis longissimus*). La actualización de los datos disponibles, considerados a partir de 2005 y basados en diversos estudios realizados en los últimos años, muestra una distribución más menos generalizada por todo el territorio considerado para la mayor parte de las especies. Entre las excepciones, dos taxones amenazados a escala regional (*Bufo calamita* y *Hyla meridionalis*), únicamente presentes en dos poblaciones costeras aisladas. A falta de estudios específicos detallados (censos poblacionales para buena parte de las especies), la composición herpetológica de las áreas periurbanas es semejante a la de las áreas naturales, muchas de ellas englobadas en espacios protegidos (parques naturales) o áreas gestionadas (zonas de especial conservación). La densa trama urbana y la tupida red de viales de comunicación pueden ser una barrera a la comunicación y expansión de las poblaciones, fenómeno todavía pendiente de investigación que deberá ser explorado en próximos estudios. Un buen diseño urbano, acompañado de una buena gestión de los espacios verdes, facilitaría la supervivencia de las poblaciones urbanas y su interconexión, así como la de las poblaciones en ambientes periféricos naturales.

Current status of the herpetofauna in the Spanish zone of Basque Eurocity Bayonne-San Sebastián

The transboundary Basque Eurocity Bayona (Aquitaine, France)-San Sebastián (Basque Country, Spain) has been shaped as an urban corridor between two border states, populated by some 600,000 inhabitants. The Spanish part of the Eurocity, around the conurbation San Sebastián-Irún, is composed of thirteen municipalities, peaking at around 400,000 inhabitants, which gather the 55 % of the population of the province of Gipuzkoa. This territory, situated on the Cantabrian coast, is composed of low-altitude mountains in which human use since the last quarter of the nineteenth century has created a landscape consisting mostly of meadows and fields of Atlantic crops, with traces of the ancient deciduous woodland, dominated by the oak (*Quercus robur*), and plantations based on pine forests. The herpetofauna of the territory is mainly composed of European species of more or less extended range in the continent, some of them present in western or only in France and Spain. Nine of the 21 species currently found correspond to amphibians and twelve to reptiles. Most of them are features of the humanised environment of the countryside, and are present in the forest and in the ecotone between the forests and meadows, often living at the expense of a rural environment transformed by the secular human activity. The edges of meadows, hedges, shrubs and grasslands that have replaced forests and human constructions are the preferred habitat of 80% of the species, some of which are cataloged (among others, *Triturus marmoratus*, *Bufo calamita*, *Hyla meridionalis*, and *Zamenis longissimus*). An update of the available data, considered from 2005 and based on several studies in recent years, shows a more or less widespread range throughout the considered territory for most of the species. The exceptions are two regionally threatened taxa (*Bufo calamita* and *Hyla meridionalis*), present only in two isolated coastal populations. In the absence of specific detailed studies (population censuses for good part of the species), the herpetological composition of peri-urban areas is similar to that of natural areas, many of them included in protected areas (natural parks) or managed areas (special areas of conservation). The dense urban matrix and network of vials of communication can be a barrier to the connection and expansion of populations, a phenomenon still not investigated in this territory that should be explored in future studies. A good urban design, close to a good management of green spaces, will facilitate the survival of urban populations and their interconnection, as well as the peripheral populations in natural environments.

Amphibians, reptiles, distribution, conservation, Basque Country.

(P68) **Distribución y estado de conservación de los anfibios en la Reserva de la Biosfera de las Bardenas Reales de Navarra**

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La Reserva de la Biosfera (y Parque Natural) de las Bardenas Reales, en el sur de Navarra, es una vasta extensión de origen terciario de unas 40.000 ha, caracterizada por su aridez, con suelos de arcillas, yesos y areniscas, cuya vegetación natural se compone fundamentalmente de formaciones de matorrales (algunas de ellas catalogadas en la Directiva de Hábitats), pinares carrascos y saladares, con la mitad de su superficie dedicada a los cultivos cerealistas extensivos. Algunos afloramientos de agua dan lugar a barrancos húmedos permanentes, con frecuencia densamente poblados de carrizo. La precipitación, de régimen torrencial, contribuye fuertemente a la severa erosión que modela el paisaje y abastece los barrancos estacionales. Existe una gran tradición agroganadera, en especial de ganado ovino, que ha generado la construcción de corrales, repartidos por todo el enclave y que frecuentemente llevan asociadas balsas ganaderas. En un estudio realizado en 2012 con el objetivo de actualizar el estatus de la comunidad de anfibios se catalogaron más de 500 puntos de agua, la mayoría de origen antrópico, y algunos de ellos actualmente no funcionales debido a la colmatación, a la apertura de drenajes en las cubetas de las balsas o al abandono como abrevaderos del ganado. Las balsas ganaderas conforman el principal hábitat de reproducción para las cuatro principales especies de anfibios que actualmente habitan la Reserva de la Biosfera: *Pelophylax perezi*, *Bufo calamita*, *Pelodytes punctatus* y *Pelobates cultripes*. La otra especie presente, *Bufo spinosus*, se reproduce en barrancos húmedos de escasa corriente, cubiertos de densos carrizales. Una sexta especie detectada en las inmediaciones de la Reserva, *Triturus marmoratus*, se ha localizado en densidades ínfimas en una balsa situada a menos de un kilómetro del enclave. En balsas infestadas por *Procambarus clarkii* aparece prácticamente como único anfibio *P. perezi*, y en alguna ocasión se han observado larvas de *P. cultripes* visiblemente atacadas por los cangrejos. Parece indudable que las poblaciones de anfibios más comunes en este espacio protegido han resultado beneficiadas por la recreación histórica de enclaves húmedos para el abastecimiento del ganado. Sin embargo, el intenso aprovechamiento agrícola de numerosos sectores puede estar afectando negativamente a estas especies (y otras antiguamente citadas en el mismo) por el uso de fitosanitarios y la pérdida superficial de un hábitat terrestre adecuado para ellas.

Distribution and conservation status of amphibians in Bardenas Reales Biosphere Reserve (Navarre, Spain)

Bardenas Reales Biosphere Reserve (and Natural Park), in Southern Navarre (Spain), is a 40.000 Ha territory originated in the Tertiary and characterized by its aridity, with soils composed by clays, gypsums and sandstones, and natural vegetation including mainly Mediterranean shrubs (some of them included in the Habitats Directive), Aleppo pines and halophile species, with half of its area dedicated to cereal extensive crops. Moreover, there are water springs that result in permanent streams, often densely populated by reed. There are also seasonal streams fed by torrential rainfall regime, which strongly contributes to the severe erosion that shapes the landscape. The area maintains a large agricultural and farming tradition, hosting numerous herds of sheep, which have generated the construction of corrals, spread throughout the territory and often with associated livestock ponds. In a study conducted in 2012 with the aim of updating the status of the amphibian community, more than 500 wetlands were inventoried, the vast majority human-made, and some of them currently not functional due to silting, draining or abandonment for their use for cattle watering. Livestock ponds form the main breeding habitat for the four species of amphibians that currently inhabit the Biosphere Reserve: *Pelophylax perezi*, *Bufo calamita*, *Pelobates cultripes* and *Pelodytes punctatus*. The other species present, *Bufo spinosus*, is distributed in specific and restricted sectors of the Bardenas breeding in low-flow streams, covered with dense reedbeds. A sixth species detected in the vicinity of the reserve, *Triturus marmoratus*, is located at extremely low densities in a single pond located within a kilometre of the Reserve. In ponds infested by *Procambarus clarkii*, *Pelophylax perezi* seems to be the only amphibian species present, but sometimes *Pelobates cultripes* tadpoles have been observed, visibly damaged by crabs. There is little doubt that the most common amphibian populations in this protected area have benefited from historical creation of wetlands to supply water for livestock. However, intense agricultural use in many sectors may be adversely affecting these species (and others that were formerly reported there) by the use of pesticides and the loss of suitable terrestrial habitat.

Amphibians, Bardenas Reales, distribution, Biosphere Reserve.

(P69) **Comparación de los métodos de marcaje-recaptura y las estimas genéticas del tamaño poblacional en una población urbana de salamandra común (*Salamandra salamandra*) de la ciudad de Oviedo**

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La pérdida y fragmentación del hábitat causada por las actividades humanas está considerada como una de las mayores amenazas para la biodiversidad. En la ciudad de Oviedo algunas poblaciones de salamandra común permanecen aisladas desde el S. VIII, después de la construcción de varios edificios y una muralla defensiva que rodeaba la ciudad. Otras poblaciones se fueron aislando paulatinamente a medida que avanzaba el desarrollo urbanístico de la ciudad. Esto es posible gracias a que esta especie evolucionó hacia el viviparismo (nacimiento de larvas terrestres completamente metamorfoseadas) durante el periodo Plioceno-Pleistoceno, lo que les permitió sobrevivir en pequeñas parcelas dentro de la ciudad donde la presencia de masas de agua es muy escasa o totalmente inexistente. Las especiales condiciones de estas poblaciones urbanas hacen de la ciudad de Oviedo un perfecto laboratorio para el estudio de la dinámica de poblaciones y nos dan la oportunidad de comparar diferentes métodos de análisis en poblaciones fragmentadas. Nuestros objetivos son: 1) obtener censos de población (N_c) y compararlos con estimas de población efectiva (N_e); y 2) discutir los resultados de N_e calculados con diferentes métodos en un escenario de población aislada y endogámica. En este trabajo hemos seleccionado una de estas poblaciones, situada en una parcela de 248 m² situada en el campus de la Universidad de Oviedo. Esta parcela de estudio se aisló del resto en 1985, a partir de la construcción de varios edificios universitarios y la pavimentación de dos calles alrededor de la misma. Entre 2008 y 2011 capturamos 210 salamandras que fueron marcadas con marcas PIT, visitando la parcela periódicamente hasta el otoño de 2013 para localizar los ejemplares marcados y para estimar el N_c . Por otra parte, 58 de estos animales fueron genotipados usando 15 microsatélites para calcular el N_e mediante cuatro métodos diferentes que emplean estimadores a partir de una sola muestra. Los análisis de marcaje-recaptura estimaron una población media de 92 ± 5 ejemplares adultos ($n \pm SE$) para el periodo 2008 a 2013 (IC del 95%: 83 a 103). El análisis genético estimó un N_e entre 56 y 80 adultos (95% IC: 40 - 110), según el método utilizado. Este estudio nos ha permitido comparar los valores de N_c y N_e y los resultados muestran estimaciones similares, ligeramente inferior en N_e . Por otra parte, este estudio revela la existencia de una población inesperadamente elevada para una parcela tan pequeña, poniendo de manifiesto la utilidad de análisis demográficos para comprender mejor la dinámica de la población y el estado de conservación de las poblaciones naturales.

Comparing mark-recapture and genetic estimates of population size in an urban Fire salamander (*Salamandra salamandra*) population from the city of Oviedo (North of Spain)

The loss and fragmentation of natural habitats caused by human activities is widely considered one of the major threats to biodiversity. In the city of Oviedo, fire salamander populations remain isolated from the eighth century after the construction of buildings and defense walls, while others were isolated in more recent times in association with urban growth. Their survival is possible because this species evolved viviparity (birth of fully metamorphosed terrestrial juveniles) in the Pliocene-Pleistocene, which allowed them to persist in small patches in the city where the presence of water bodies is very scarce or even totally absent. The special characteristics of these urban populations make the city of Oviedo a great field laboratory to study population dynamics and compare different methods of analysis of fragmented populations. Our goals are to: 1) calculate census size (N_c) and compare with effective population size (N_e) estimates; and 2) discuss the potential of different methods to accurately estimate N_e in a scenario of isolated and inbred populations. We selected one of these populations, situated in a plot of 248 m² on the University of Oviedo campus. This study plot was isolated in 1985 after the construction of several university buildings and two paved roads around the plot. Between 2008 and 2011, 210 fire salamanders were captured and marked with PIT tags and the plot was visited periodically until the fall of 2013 to locate all tagged individuals. Moreover, 58 of these animals were genotyped using 15 microsatellites to calculate N_e using four different methods that use single-sample estimators. Mark-recapture analysis estimated an average adult population of 92 ± 5 individuals ($n \pm S.E.$) in the period 2008-2013 (95% CI: 83-103). Genetic analysis estimates an N_e of ca. 56-80 adults (CI: 40 - 110) depending on the method used. This study allowed us to compare N_c and N_e values and the results show similar estimates, slightly lower in N_e . Moreover, this study unveils an unexpectedly large population in an extremely small patch, highlighting the utility of demographic analyses to better understand the population dynamics and conservation status of natural populations.

Salamandra salamandra, urban populations, census size, effective population size, comparative methods.

(P70) **Actualización histórica del conocimiento de las comunidades de galápagos del municipio de Vitoria-Gasteiz**

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Los primeros trabajos llevados a cabo para conocer la situación de las comunidades de galápagos en el municipio de Vitoria-Gasteiz comenzaron ya en 2001 y se han llevado a cabo de forma sistemática desde 2008. Desde este año se lleva a cabo una campaña intensiva anual en Salburua, con el objetivo de conocer la dinámica de las poblaciones autóctonas del Parque y llevar a cabo el control de exóticas, acompañada de una campaña complementaria en otras zonas del municipio. Durante estas campañas, todos los ejemplares de especies exóticas fueron retirados del medio. En el presente trabajo se muestra un resumen de las capturas de galápagos exóticos y galápagos autóctonos realizadas a lo largo de este periodo en todo el ámbito del municipio. Los datos aquí mostrados incluyen todos los individuos capturados durante estos trabajos, así como cualquier otro ejemplar recogidos por los servicios de guardería del Anillo Verde o entregados por visitantes y usuarios a las administraciones.

A lo largo de estos años se han identificado 54 ejemplares de galápago leproso y dos de galápago europeo, y se han extraído del medio 49 galápagos exóticos de diferentes taxones, predominantemente *Trachemys scripta*. El número de capturas de nuevos ejemplares mostró un importante incremento a partir de 2008, al incrementarse notablemente el esfuerzo de muestreo, y ha sufrido importantes variaciones inter-anales desde entonces, oscilando entre 1 y 14 para los autóctonos y entre 4 y 14 para los exóticos. La mayor parte de las capturas se llevaron a cabo en el humedal de Salburua, de acuerdo con su mayor esfuerzo de muestreo, seguido por el meandro muerto de Aramangelu. Dentro del humedal de Salburua, la balsa de Arkaute resulta de mayor relevancia para el galápago leproso, mientras que la abundancia de exóticos parece ser mayor en Betoño. La razón de sexos de los galápagos leprosos no resultó significativamente diferente de 1 (1,2 machos por hembra), mientras que la de galápagos exóticos estuvo fuertemente sesgada hacia las hembras (3,3 hembras por macho). El rango de tamaños y edades resultó muy amplio, de 50 a más de 226 mm de espaldar para los galápagos leprosos y de 102 a 282 para *Trachemys scripta*. Cabe destacar la captura de un juvenil de 50 mm en Salburua, lo que permite confirmar por primera vez la reproducción de esta especie en el humedal.

Historical account of the knowledge on pond turtle communities in Vitoria-Gasteiz municipality

The first works carried out on the status of pond turtle communities in Vitoria-Gasteiz municipality began back in 2001 and have continued in a systematic way since 2008. From then on, an annual intensive capture campaign is being carried out in Salburua, aiming at knowing the dynamics of native populations and at controlling exotic pond turtle species, together with an ancillary campaign in other areas of the council. All exotic individuals captured in these campaigns were extracted from the environment. Here we present a summary of native and exotic pond turtle captures along this time period in the whole municipality. The data shown include all the captured individuals plus any other individual captured by wardens of the Green Ring or taken by visitors and users to municipally administration.

Along these years we have identified 54 Spanish and two European pond turtles, and we extracted from the environment a total of 49 exotic terrapins belonging to several taxa, mostly *Trachemys scripta*. The number of individuals captured for the first time increased notably since 2008, as a result of an increased sampling effort, and has shown ample annual variability since then, varying from 1 to 14 for the native turtles and from 4 to 14 for the exotic ones. Most captures were carried out in the Salburua wetland, according to its higher sampling effort, followed by the Aramangelu oxbow. Within Salburua wetland, the Arkaute pond was the most important area for Spanish pond turtle, while exotic pond turtles were more abundant at Betoño. Sex ratio of Spanish pond turtle did not differ from 1 (1.2 males per female), while that of exotic terrapins was severely biased towards females (3.3 females per male). Size and age of captured turtles varied greatly, from a carapace length of 50 to more than 226 mm for Spanish pond turtle and from 102 to 282 for *Trachemys scripta*. Remarkably, we captured a young individual of 50 mm in Salburua, which confirms for the first time the reproduction of this species in this wetland.

Mauremys leprosa, *Emys orbicularis*, *Trachemys scripta*, Vitoria-Gasteiz, communities.

(P71) **Dinámica poblacional y crecimiento del galápago leproso *Mauremys leprosa* en el humedal de Salburua**

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Desde el año 2009 se ha llevado a cabo un seguimiento anual de la población de *Mauremys leprosa* del humedal de Salburua, aprovechando las labores de extracción de galápagos exóticos. El seguimiento se lleva a cabo por medio de al menos una campaña anual de trampeo de 10 días de duración en las balsas de Arkaute y Betoño. Los individuos capturados son medidos, sexados y pesados y finalmente marcados individualmente por medio de transponders o marcas SP.

El número de capturas ha sufrido importantes variaciones a lo largo de estos años, con 14 capturas en 2009 y mucho menores para los años posteriores. El bajo número de capturas de estos últimos años imposibilitó la obtención de estimas de abundancia robustas por medio de modelos de poblaciones cerradas, aunque esto sí fue posible para 2009 y 2010, en que la población estimada osciló entre 8 (error típico: 1,36) y 16 (et: 7,53) individuos. La supervivencia no varió a lo largo de los cinco años de muestreo, con una supervivencia anual estimada de 0,8 (et: 0,11; modelo de poblaciones abiertas de Cormack-Jolly-Seber). Sin embargo, la tasa de crecimiento de la población (estimada a partir de un modelo de Pradel) varió entre 0,51 (et: 0,11) y 2,66 (et: 1,29). Basándonos en la biología de la especie, la elevada variación de este parámetro apunta a la violación de alguna de las asunciones del modelo. Probablemente, la superficie cubierta por la especie (y por tanto, la proporción de la población total que resulta capturable cada año) varía en función del nivel de agua del humedal y de movimientos de larga distancia de los individuos. De hecho, nuestros datos de telemetría muestran desplazamientos de varios kilómetros que permitirían conectar el humedal con el curso del Zadorra. El número de hembras recapturadas fue inferior al de machos y no permitió realizar el análisis de crecimiento para este sexo. Asumiendo que el crecimiento de la especie sigue una curva de von Bertalanffy, la tasa estimada para los machos fue de 0,026, por lo que la ecuación $L_2 = 231 - (231 - L_1)e^{-k(t_2 - t_1)}$ describe adecuadamente el crecimiento anual de los machos de *M. leprosa*. Esta tasa de crecimiento es muy reducida si la comparamos con los datos de otros autores para ésta y otras especies afines.

Population dynamics and body growth of the Spanish pond turtle *Mauremys leprosa* in the Salburua wetland

Since 2009 we have carried out an annual monitoring of the population of *Mauremys leprosa* in the Salburua wetland, taking advantage of the campaigns for the extraction of exotic pond turtles. This monitoring consisted of at least one annual 10-day trapping session in the Arkaute and Betoño ponds. All captured individuals were measured, sexed and weighed, and finally individually marked with passive transponders or SP marginal marks.

The number of captures has suffered important variations along years, with a value of 14 for 2009, and much lower values for the following years. The low number of captures made impossible to obtain a robust abundance estimate for these years, although this was possible for 2009 and 2010, when the estimated population size varied from 8 (standard error: 1.36) to 16 (se: 7.53). Survival did not vary along these five years, with an annual estimate of 0.8 (et: 0.11; Cormack-Jolly-Seber open population models). However, population growth rate (estimated by a Pradel model) varied from 0.51 (se: 0.11) to 2.66 (se: 1.29). Based on the biology of the species, the high variability of this parameter points to a violation of model assumptions. Probably, the area covered by the species (and hence, the proportion of total population available to be captured) varies with water level of the wetland and with long-distance movements of the individuals. In fact, our radiotracking data show displacements of several kilometers that would imply connecting Salburua with the Zadorra River. Recaptured females were scarce and not enough for analyzing individual body growth for this sex. Assuming that body growth for this species follows a von Bertalanffy curve, the growth rate for males was estimated to be 0.026, and hence the equation $L_2 = 231 - (231 - L_1)e^{-k(t_2 - t_1)}$ properly describes annual growth of males of *M. leprosa*. This growth rate is much lower to that reported by other authors for this and related species.

Spanish pond turtle, *Mauremys leprosa*, Salburua, Zadorra River, population dynamics, body growth.

(P72) Estado de uma população de cágado-de-carapaça-estriada num sistema de charcos no Sudoeste de Portugal: 11 anos de dados

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Em Portugal, o cágado-de-carapaça-estriada, *Emys orbicularis*, é uma espécie ameaçada que é localmente abundante em sistemas de charcos localizados ao longo da plataforma costeira. Esta preferência de habitats é um factor importante de ameaça, uma vez que as zonas costeiras são tipicamente sujeitas a uma elevada pressão urbanística. Com efeito, verificou-se um forte declínio no número de charcos no Sudoeste Alentejano desde os anos 90, com a destruição de cerca de 56% do número total de charcos existentes em 1991, principalmente devido à intensificação agrícola que se fez sentir na região. Desde 2009, a Associação Nacional de Conservação da Natureza, Quercus, tem vindo a promover uma acção de gestão de um sistema de charcos de carácter temporário no Sudoeste Alentejano para a conservação do cágado-de-carapaça-estriada, localizado numa propriedade privada dedicada predominantemente à criação de gado. A população de cágados da área tem sido regularmente amostrada desde 2003. Um total de 595 capturas, totalizando 205 indivíduos, foram já realizadas. Neste estudo, analisou-se a evolução de parâmetros da população frequentemente utilizados como indicadores do estado das populações de tartarugas, incluindo a razão entre sexos, a estrutura de tamanhos, a estrutura etária, os efectivos populacionais e as densidades. A estimação dos efectivos populacionais foi baseada em métodos de captura-marcação-recaptura, considerando uma variabilidade na probabilidade de captura ao longo do tempo e entre indivíduos. Uma vez que o esforço de amostragem foi mais intenso e durante um período mais longo em 2003 e 2013, a maioria dos testes estatísticos realizados focaram-se em comparações entre estes anos. Não foram encontradas diferenças significativas entre 2003 e 2013 na razão entre sexos, na proporção de juvenis e na proporção de recém-nascidos. No que se refere aos tamanhos, apenas foi detectado um decréscimo significativo nas dimensões corporais de machos. Segundo as estimativas dos efectivos populacionais, houve um aumento da população entre 2003 e 2013, de 109.5 indivíduos (I.C. 95%, 82.7-166.1) para 135.2 indivíduos (I.C. 95%, 106.9 – 190.0), apesar da elevada sobreposição dos intervalos de confiança a 95%. As estimativas de densidade variaram entre os 46.6 indivíduos por hectare em 2003 e os 57.5 indivíduos por hectare em 2013. Em conclusão, a população do cágado-de-carapaça-estriada encontra-se estável com tendência para um pequeno aumento dos efectivos populacionais.

Conservation status of the European pond turtle in a pond system in the SW Portugal: 11 years of data

In Portugal, the European pond turtle, *Emys orbicularis*, is an endangered species that is locally abundant in pond systems along the coastal platform. This is a matter of concern, since coastal zones are typically subjected to a strong urbanization pressure. In fact, a strong decline in the number of ponds in SW Alentejo, Portugal, have been already taking place since the nineties, with a destruction of almost 56% of the total existing ponds in 1991, mainly due to the agricultural intensification in the region. Since 2009, the National Association for Nature Conservation, Quercus, has been promoting a management action for the conservation of *E. orbicularis* in a system of interconnected temporary ponds located in SW Alentejo in a private property mainly dedicated to livestock. The turtle population living in this area has been regularly surveyed since 2003. A total of 595 captures, totaling 205 individuals, were performed since then. In the present study we aimed to analyze the evolution of several population parameters, most often used as indicators of turtle population status, including sex ratio, size structure, age structure, population estimate and density. Population estimates were based on capture-recapture methods that considered both a temporal and an individual variability in the capture probabilities. Since the sampling effort was more intense and longer in 2003 and 2013, most statistical tests focused in comparisons between these two years. No significant differences between 2003 and 2013 were found in the sex ratio, the proportion of juveniles and the proportion of hatchlings. Regarding size, a significant difference was only found for males that showed an overall decrease in size. A near significant increase in the size of juveniles was also found. According to population size estimates, there was an increase from 2003 to 2013 from 109.5 (95% C.I. 82.7-166.1) to 135.2 individuals (95% C.I. 106.9 – 190.0), although there is a wide overlap of the 95% confidence intervals. Density estimates varied between 46.6 individuals per hectare in 2003 to 57.5 individuals per hectare in 2013. In conclusion, according to the studied population's parameters, the population of the European pond turtle seems to be stable in the study area, possibly with a tendency for a small increase in population size.

Age structure, *Emys orbicularis*, population size, sex ratio, size structure.

(P73) Diagnosis de la població de galápagu europeo (*Emys orbicularis*) en la Sèquia Major de Vila-Seca, Tarragona, Catalunya

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La Sèquia Major de Vila-seca es una de las últimas zonas húmedas de Tarragona donde aún habita el galápagu europeo. Este Espacio de Interés Natural (EIN) tiene una superficie de 16,6 ha y se halla en una comarca muy humanizada y profundamente transformada. Este entorno de alta frecuentación, contaminación de las aguas y fuerte presión urbanística, hizo menguar la presencia del quelonio, referenciada desde los años 90. Los primeros estudios de 1998 estimaron la población en una veintena de ejemplares. En 2008 se marcaron 16 ejemplares y en 2010 una última prospección arrojó datos muy negativos, próximos a la desaparición de la especie, cuando sólo se capturaron tres quelonios. En 1999, un estudio de determinación genética de las poblaciones de *Emys orbicularis* en Catalunya, incluyó la población de la Sèquia Major en el haplotipo V, *E. o. galloitalica*.

En 2013 el Departamento de Agricultura, Ganadería, Pesca, Alimentación y Medio Natural de la Generalitat de Catalunya hizo una prospección sistematizada para evaluar el estatus de la especie. Entre los meses de abril y septiembre se colocaron en el conjunto de lagunas, canales y lagos artificiales del campo de golf 15 estaciones de muestreo de 10 trampas cada una durante una semana (150 nasas, 105 días de prospección). Se capturaron 53 galápagos; cada ejemplar se identificó individualmente con microchip, se fotografió (caparazón y plastrón), y se tomaron los datos biométricos básicos (peso, anchura, altura máxima y longitud recta del caparazón). De los 53 animales, 27 eran machos, 19 hembras, cinco juveniles y dos crías. El tamaño medio de las hembras (curva escapular, CE) fue de 152 mm (rango 86-185) y el peso medio de 450 g (rango 108-686); los machos mostraron una CE media de 142,1 mm (rango 107-168) y un peso medio de 319,3 g (rango 150-554). Sólo se capturó un ejemplar nacido en 2012 y otro 2013, que medían 47 y 30,5 mm respectivamente. Las recapturas reiteradas de algunos ejemplares (30 ejemplares fueron recapturados, con un total de 83 recapturas, y un único ejemplar fue recapturado hasta 11 veces) evidenciaron desplazamientos por todo el complejo de canales y lagunas, con una distancia máxima comprobada entre puntos de captura de 651,23 m.

Los resultados obtenidos sugieren que el aislamiento artificial del espacio mediante una valla ha resguardado en cierta forma a la especie de agresiones directas e indirectas, lo que ha permitido una lenta recuperación poblacional, aunque aún existen amenazas que hay que minimizar, como la protección de las puestas frente a predadores e inundaciones, el control de tortugas alóctonas y el aislamiento poblacional. Para los próximos años, además de continuar con los muestreos, se radiomarcarán hembras adultas para localizar las zonas de puesta y se crearán pasillos naturales entre los lagos para facilitar la dispersión de individuos y flexibilizar el área de distribución ya que la población de *E. orbicularis* solo ocupa 23,8% de la zona disponible.

Diagnosis of the populations of European pond turtle (*Emys orbicularis*) in Sèquia Major de Vila-Seca, Tarragona, Catalonia

The Sèquia Major de Vila-Seca is one of the last wetlands in Tarragona where the European pond turtle is still present. This Space of Natural Interest (EIN) covers an area of 16.6 ha and is located in a highly humanized and deeply transformed region. This environment of high attendance, water pollution and strong urban pressure, contributed to diminishing the presence of the turtle, referenced from the 90s. Early studies in 1998 estimated the population size in about 20 animals. In 2008, 16 specimens were found, and a final survey in 2010 showed very negative data, next to the demise of the species, when only three turtles were captured. In 1999, a study of genetic determination of populations of *Emys orbicularis* in Catalonia included the population of Sèquia Major in the haplotype V, *E. o. galloitalica*.

In 2013 the Department of Agriculture, Livestock, Fisheries, Food and Natural Environment of the Generalitat de Catalunya made a systematic survey to assess the status of the species. Between April and September, 15 sampling stations with 10 traps each were placed in the group of lagoons, canals and artificial lakes in the golf court for a week (150 pots, 105 days prospecting). We captured 53 turtles, identified each animal individually with a microchip, photographed them (carapace and plastron), and took basic biometric data (weight, width, maximum height and straight carapace length). Of the 53 animals, 27 were males, 19 females, five juveniles and two hatchlings. The average size of females (shoulder curve, SC) was

152 mm (range 86-185) and the average weight was 450 g (range 108-686); males showed an average SC of 142,1 mm (range 107-168) and an average mass of 319,3 g, (range 150-554). We only capture one individual born in 2012 and another born in 2013, measuring 47 and 30.5 mm, respectively. Repeated recaptures of specimens (30 specimens were recaptured for a total of 83 recaptures, and one specimen was recaptured up to 11 times) showed movements around the whole complex of canals and lagoons, with a maximum proven distance between points of capture of 651.23 m.

These results suggest that the artificial isolation of the space by a fence has somehow protected the species from direct and indirect aggression, allowing a slow population recovery. There are still threats to be minimized, such as the protection of nests from predators and flooding, and the control of non-native turtles and population isolation. In the coming years, in addition to continue samplings, adult females will be radio-tracked to locate nesting areas, and natural corridors among the lakes will be created to ease the dispersion of individuals and to widen the distribution area of the species in this space, as the population of *E. orbicularis* occupies only 23.8% of the available area.

Pond turtle, mark-recapture, sex ratio.

(P74) Situación de la población de tortuga mediterránea (*Testudo hermanni hermanni*) en la sierra de Llaberia, Tarragona, Cataluña

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La población de tortuga mediterránea (*Testudo hermanni hermanni*) encontrada el año 2005 en las faldas del cerro de "La Miloquera", dentro del término municipal de Marçà, Priorat, Tarragona, en el Sur de Cataluña, presenta todos los indicios de provenir de sueltas incontroladas, por su cercanía al núcleo urbano. Para determinar si procedían de sueltas recientes o se trataba de una subespecie no propia de Cataluña, durante el año 2007 se realizó una caracterización genética de la población. El resultado del estudio fue que todas las tortugas de la población de Marçà analizadas presentan un genoma mitocondrial que se corresponde al de *Testudo hermanni hermanni*. La población de Marçà se estimó, en 2005, en un total de unos 235 individuos. Desde entonces no había habido un plan de seguimiento establecido para determinar el estado poblacional, la dispersión de la especie en la zona o la viabilidad de dicha población.

En el año 2013 el gobierno de Cataluña junto con el ayuntamiento de Marçà inició un plan de seguimiento que comienza a vislumbrar el estado real de dicha población y su dispersión dentro del territorio, y que debe macar las pautas a seguir para la conservación de la especie en la zona. Después de seis censos y diferentes encuentros de individuos por el campo, el número de ejemplares censados y marcados en la zona es de 26 tortugas, 17 de las cuales se encontraron durante los períodos de censo mientras que las nueve restantes han sido encontradas por diferentes causas en la zona. Después del marcado con microchips y tomados los respectivos datos, los animales son liberados de nuevo. Respecto a la proporción de sexos y la clase de edad, de los 27 individuos, nueve eran juveniles y el resto, 17, correspondían a individuos adultos (65%), de los cuales nueve son hembras y ocho machos. De todos los ejemplares se tomaron medidas biométricas, peso, curva escapular, anchura del plastrón, longitud y anchura del caparazón y altura máxima. El peso medio de las hembras es de 639,9 g (rango 84-1328 g) y la longitud media es de 129,4 mm (rango 55-210 mm); en los machos, la media de peso es de 486,57 g (rango 172-654 g) y la longitud media es de 117,75 mm (rango 72-150 mm).

Population status of Mediterranean Tortoise (*Testudo hermanni hermanni*) in Serra de Llaberia, Tarragona, Catalonia

The population of Mediterranean tortoise (*Testudo hermanni hermanni*) found in 2005 in the hills of "The Miloquera", municipality of Marçà, Priorat, Tarragona, in southern Catalonia, shows all signs of coming from uncontrolled release, due to its proximity to the urban core. To determine whether they came from recent releases or whether it was a subspecies not from Catalonia, a genetic characterization of the population was performed in 2007. The result of the study was that all turtles of the population of Marçà that were analyzed had a mitochondrial genome that corresponds to *Testudo hermanni hermanni*. The population size of Marçà was estimated in 2005 to be about 235 individuals. Since then, there had not been a monitoring plan to determine the population status, the dispersion of the species in the area, or the viability of this population.

In 2013 the government of Catalonia and the town council of Marçà initiated a monitoring plan that is beginning to show the real status of the population and their dispersion within the territory, and which should set the guidelines to follow for the conservation of the species in the area. After six censuses and different findings of individuals, the number of specimens counted and marked in the area is 26 turtles, 17 of which were found during the censuses periods, while the remaining 9 were found by different causes in the area. After microchipping and taking biometric data, animals are released back. As for the sex ratio and age classes, of the 26 individuals, nine were juveniles and the remaining 17 corresponded to adult individuals, out of which nine were females and eight males. All biometric measures, weight, shoulder curve, plastron width, carapace length and width, and maximum height were taken from all individuals. The average weight of females is 639.9 g (range 84-1328 g), and the average length is 129.4 mm (range 55-210 mm); in males, the weight average is 486.57 g (range 172-654 g), and the average length is 117.75 mm (range 72-150 mm).

Mediterranean tortoise, population status, microchipped, sex ratio.

(P75) *Gallotia galloti insulanagae*, el lagarto del Roque de Fuera de Anaga, Tenerife, España

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Gallotia galloti insulanagae es una subespecie que vive exclusivamente en el Roque de Fuera de Anaga (Tenerife, Islas Canarias). Se aportan datos sobre su biometría, dinámica poblacional y abundancia. Así mismo se incluye una descripción del medio donde vive y de los recursos tróficos de los que dispone.

Esta subespecie fue descrita en el año 1985, se diferencia de la del norte de Tenerife por su melanismo y mayor tamaño. Su hábitat se caracteriza por la baja diversidad vegetal y la gran presencia de aves marinas. En diferentes campañas se han realizado censos para estimar su población, calculada en 350 ejemplares para todo el Roque, con una densidad aproximada de unos 200 individuos por hectárea. Esta densidad es bastante superior a la de otros lagartos que viven en ambientes similares. A pesar de su aislamiento y aparente escasez de recursos, el estado general de la población parece adecuado, presentando además una proporción de machos a hembras próxima a uno.

***Gallotia galloti insulanagae*, the “Roque de Fuera de Anaga” lizard (Tenerife, Spain)**

Gallotia galloti insulanagae is a subspecies inhabiting exclusively the islet of “Roque de Fuera de Anaga” (Tenerife, Canary Islands). Here we provide data on biometry, population dynamics and abundance. We also include a description of the environment where it lives and trophic resources available.

This subspecies was described in 1985. It differs from that of northern Tenerife because of its melanism and larger size. Its habitat presents a low floral diversity and an abundant presence of marine birds. We have conducted census to estimate the population size in several campaigns. The population size is estimated in 350 specimens on the whole “Roque”, with an approximate density of 200 individuals per hectare. This density is higher than that observed for other lizards inhabiting similar environments. In spite of its isolation and apparent resource scarcity, the general status of the population is adequate. Furthermore, the sex ratio is close to one.

Gallotia, abundance, biometric data, isolation.

(P76) Nuevos datos sobre capturas accidentales de tortugas laúd (*Dermochelys coriacea* Vandelli, 1761) obtenidos por el IEO en el marco del Programa de Observadores a bordo en las pesquerías españolas de palangre de superficie de túnidos y especies afines en el Mediterráneo occidental y el Golfo de Cádiz (NE Atlántico)

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La conservación de las especies marinas altamente migratorias requiere de acciones coordinadas entre expertos y administraciones sustentadas en la mejor información científica disponible en cada país, en particular la relacionada con el uso de hábitats y la incidencia sobre la especie de las principales pesquerías desarrolladas en su área de distribución. La tortuga laúd tiene una distribución geográfica amplia llegando hasta las frías aguas de Alaska en el norte. Las regiones del Golfo de Cádiz y el Mediterráneo occidental son habitualmente ocupadas por ejemplares de tortuga laúd de poblaciones atlánticas pertenecientes a distintas Regional Management Units (RMU). En nuestra área de estudio, las tortugas tendrían origen tanto americano (Atlantic Northwest RMU) como africano (Atlantic Southeast RMU).

El Golfo de Cádiz y el Mediterráneo occidental reúnen importantes pesquerías potencialmente peligrosas para la especie, considerada globalmente vulnerable (A2bd, según criterio del MTSG de la IUCN) y que podrían afectar a ejemplares de las dos RMU señaladas. Coordinado por el Centro Oceanográfico de Málaga, el Programa de Observadores a bordo del IEO en las pesquerías españolas de palangre que pescan túnidos y afines en el Mediterráneo occidental y Golfo de Cádiz, ha permitido desde 1999 la recopilación sistemática y científica de la información sobre las capturas accidentales de las especies objetivo, además de otras incidentales como tiburones, tortugas marinas, cetáceos y aves capturadas por las flotas españolas.

Entre 1999 y 2012 se ha observado un total de ocho capturas accidentales de tortuga laúd en el Mediterráneo occidental (0.001014 ejemplares / 1000 anzuelos), lo cual contrasta con las 3940 capturas observadas de tortuga boba (0.499 ejemplares / 1000 anzuelos). El aparejo de pesca que concentró la mitad de las capturas observadas de tortuga laúd fue el palangre de superficie dirigido al atún rojo (*Thunnus thynnus*), mientras que en el caso de la tortuga boba el aparejo con mayor incidencia de capturas fue el palangre de superficie dirigido al atún blanco (*Thunnus alalunga*). Estas diferencias se explican por las profundidades y preferencias de hábitat de ambas especies y los hábitats que explota cada tipo de aparejo.

En este poster se representan los resultados de las capturas incidentales de tortuga laúd durante el periodo de ejecución del Programa de Observadores (1999-2012), contribuyendo al mejor conocimiento de las pesquerías que afectan a la especie y del área de distribución y uso de hábitat de *Dermochelys coriacea* en aguas del sur de la Península Ibérica y Mediterráneo occidental.

New data on incidental captures of leatherback (*Dermochelys coriacea* Vandelli, 1761) obtained by the IEO in the framework of the on board Observers' Program in long-line Spanish fisheries targeting tuna and tuna-like species in the Western Mediterranean and Gulf of Cadiz (NE Atlantic)

Conservation of highly migratory marine species needs coordinated efforts among experts and authorities supported on the best available scientific information in each country, particularly related to the use of habitats of key species and the impact on non-target species of the fisheries developed in their range. The leatherback has the wider geographical distribution reaching the cold waters of Alaska to the north. The regions of the Gulf of Cadiz and Western Mediterranean are usually occupied by specimens of Atlantic leatherback populations belonging to two different Regional Management Units (RMU). Leatherback in our study area would have American (Atlantic Northwest RMU) or African origin (Atlantic Southeast RMU).

The Gulf of Cadiz and Western Mediterranean gather important fisheries potentially dangerous for this species, which is considered globally vulnerable (A2bd, according to MTSG-IUCN criteria) and could affect individuals of the two mentioned RMU. Coordinated by the IEO Oceanographic Centre of Malaga, the on board Observers' Program in long-line fisheries targeting tuna and tuna-like species in the Western Mediterranean and Gulf of Cadiz has enabled since 1999 the systematic and scientific collection on catches of target species, as well as other incidental species like sharks, sea turtles, sea mammals and birds captured by Spanish fleets.

Between 1999 and 2012 a total of eight leatherbacks were incidentally caught in the Western Mediterranean Sea (0.001014 specimens / 1000 hooks), which contrasts with 3940 observed catches of loggerhead turtles (0.499 specimens / 1000 hooks). The fishing gear that concentrated half of the observed leatherback catches was the surface long-line targeting Bluefin tuna (*Thunnus thynnus*), while in the case of the loggerhead, the higher incidence of catches was the surface long-line targeting Albacore (*Thunnus alalunga*). These differences are explained by the different depths and habitat preferences of both sea turtle species and the habitats exploited by each fishing gear.

This poster presents results of the incidental capture of leatherbacks occurred during the implementation of the Observer Program (1999-2012) contributing to a better understanding of the Spanish fisheries affecting the species and the range and use of habitat by *Dermochelys coriacea* in

Leatherback, incidental captures, Mediterranean, Gulf of Cadiz.

(P77) **La Red de Rescate de Fauna Marina: análisis de resultados de tres años de funcionamiento sobre tortuga boba *Caretta caretta***

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En 2012 la Generalitat de Catalunya puso en marcha la Red de Observación y Rescate de Fauna Marina. Hasta ese momento, la gestión de los varamientos de fauna se había basado en un contrato plurianual con una ONG. Con el paso de los años, por una serie de razones que incluían una pobre obtención de datos y en un contexto de crisis que empujó al gobierno a trabajar con recursos propios, el modelo evolucionó hacia una red profesional que incluía la policía ambiental.

Esta red oficial está liderada por técnicos del Servicio de Biodiversidad y Protección de los Animales (BiPA) de la Dirección General del Medio Natural y Biodiversidad. Este núcleo forma la unidad de guardia del BiPA, que coordina y lidera otras unidades participantes: el Cuerpo de Agentes Rurales (CAR), el Servicio de Espacios Naturales Protegidos y los Parques Naturales del Delta de l'Ebre, Montgrí, Illes Medes i Baix Ter, Aiguamolls de l'Empordà y Cap de Creus y otros técnicos presentes en las unidades costeras del Gobierno: los Servicios Territoriales. El protocolo cuando se da un varamiento incluye el primer aviso al centro de emergencias 112 o al CAR, quienes avisan a la unidad del BiPA para que actúe. En el caso de ser necesaria la hospitalización, o si la necropsia de animales fallecidos recientemente es factible, se cuenta con la participación puntual de otras instituciones y/o empresas privadas.

A fecha de hoy, 2 años y medio después del despliegue, se han realizado 240 actuaciones, más del doble de lo que venía siendo habitual en los últimos años. Este aumento se puede atribuir a la implementación de la red sobre el territorio y a la intervención directa de la administración, agilizando el registro de varamientos. Además, el nuevo sistema de seguimiento y recogida de datos implica un importante despliegue ya que cada vez que se detecta un varamiento por lo menos una patrulla del CAR se desplaza al sitio de varamiento y se realiza la biometría y toma de fotografías de todos los ejemplares, así como el registro de posibles impactos, heridas, etc.

Gracias al análisis de los restos encontrados se ha podido establecer un patrón de distribución anual ligado a la temperatura del agua y se ha realizado el análisis biométrico de los ejemplares varados en Cataluña. La mayoría de los animales rescatados (n=240) son restos de tortugas boba (144 ejemplares, 63% de los rescates) que se han encontrado principalmente en el hemidelta sur del Ebro, en la comarca del Monstà (66%). Once de estas tortugas se rescataron vivas y después de unos meses hospitalizadas, seis de ellas han vuelto al mar y una falleció a los pocos días del rescate.

El protocolo de funcionamiento actual está ayudando a entender lo que sucede con las tortugas marinas en Cataluña y regiones vecinas de Valencia y ha establecido una base viable desde donde mirar hacia adelante hacia una mayor protección de estas especies en peligro de extinción.

The Marine Fauna Rescue Network: analysis of results of three years of operations on loggerhead turtle *Caretta caretta*

In 2012 the Government of Catalonia launched the Marine Fauna Observation and Rescue Network. Until then, management of strandings was based on a multi-year contract with an external NGO. Over the years, for a number of reasons including former poor data collection and a context of crisis that pushed the government to work with its own resources, the model evolved towards a professionalized network involving the environmental police.

This official network is led by wildlife experts of the Biodiversity and Animal Protection Service (BiPA) at the General Directorate of Environment and Biodiversity. This core group forms the guard unit of BiPA which coordinates and leads other participating units: Rangers Service (CAR), Protected Natural Sites Service, Natural Parks of Delta de l'Ebre, Montgrí, Illes Medes i Baix Ter, Aiguamolls de l'Empordà and Cap de Creus and other technicians widespread in the coastal units of the Government, the so called Regional Services. Procedure when facing a stranding includes first-notice to the emergency call center 112 or CAR, who then alert BiPA unit to proceed. This unit coordinates actions and solves possible doubts. If hospitalization is required, or necropsy of recently deceased animals is feasible, the network counts on the occasional participation of other institutions or private companies.

As of today, 2 ½ years after deployment, we have carried out 240 rescues, more than double the usual numbers in recent years. This increase can be attributed to the implementation of the network on the territory and to direct intervention of the administration, streamlining the stranding record. In addition, the new system of monitoring and data collection

involves a major deployment since whenever a stranding is detected at least one ranger patrol goes to the site and both biometrics and pictures of all specimens are taken, as well as registration of potential impacts, injuries, etc.

Thanks to analysis of the remains found, we have been able to establish a pattern of annual distribution linked to water temperature and performed the biometric analysis of the specimens stranded in Catalonia. Most animals (n=240) are remnants of loggerhead turtles (144 specimens, 63% of rescues) that have been found mainly in the south of the Ebro hemidelta, in the region of Montsià (66%). Eleven of these turtles were rescued alive and hospitalized; after a few months, six of them have returned to the sea and one died a few days after rescue.

Current operating protocol is helping to understand what happens with sea turtles in Catalonia and neighbour Valencia regions and has established a feasible basis from where to look forward towards a better protection of these endangered species.

Loggerhead turtle, marine fauna rescue network.

(P78) Protocolo de cría en cautividad de galápago europeo

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En la mayoría de tortugas y sobre todo en el género *Emys* existe una clara relación entre el hombre y la extinción de sus poblaciones, en su mayoría debido a la alteración, fragmentación o destrucción de sus hábitats. Cuando una población se extingue o se encuentra en una situación muy próxima, como es el caso de algunos de los núcleos poblacionales de galápago europeo (*Emys orbicularis*) distribuidos a lo largo de la península Ibérica, se puede tratar de recuperarla mediante el refuerzo o reintroducción de ejemplares criados en cautividad. Son éstos los casos en los que la cría en cautividad queda justificada. En la presente comunicación explicamos las vicisitudes de todo el proceso, mostrando algunos resultados claves adquiridos tras años de experiencia en diversos centros de reproducción en cautividad (ver lista de afiliaciones). La etapa de cría y recría, alimentación, estereotipias de estrés, mortalidad y tasas de crecimiento son algunos de los conceptos que se desarrollan en el póster. El protocolo de cría que presentamos ha sido desarrollado con el objetivo principal de incrementar las tasas de crecimiento en el mínimo tiempo posible y disminuir la tasa de depredación de los juveniles liberados en proyectos de reforzamientos poblacionales. De este modo se obtienen ejemplares que a los ocho meses de edad presentan una condición física equivalente a la de los ejemplares de dos años criados en condiciones naturales.

Captive breeding protocol of the European pond turtle

In the main quelonian species and especially, in the gender *Emys*, there is a clear relationship between humans and population extinctions associated with alteration, fragmentation or destruction of habitats. When a population becomes extinct or approaches to extinction, like some European pond turtle populations distributed along the Iberian Peninsula, we can try to recover it through reinforcement or reintroduction of captive-bred specimens. In these cases, captive breeding programs are justified. In our communication we explained the vicissitudes of the entire process, showing some key results acquired after years of experience in various captive breeding centers (view list of affiliations). The head-starting, feeding, stress, stereotypies, welfare, mortality, growth rates are some of the developed concepts. The captive breeding protocol has been developed with the main objective to increase growth rates in a short time and decrease predation on juveniles released in reinforcement projects. Thus, we obtained eight month-old, captive-bred individuals with similar physical condition as two year-old individuals grown in natural conditions.

European pond turtles, captive breeding, population reinforcements, breeding, raising, incubation.

(P79) Reprodução em cativeiro de *Emys orbicularis*: Ferramenta de sucesso para o seu reforço populacional?

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O Cágado-de-carapaça-estriada (*Emys orbicularis*) está classificado em Portugal com estatuto de “Em perigo”, verificando-se um decréscimo acentuado no último século, tanto a nível populacional como na sua distribuição. No âmbito do projecto LIFE+*Trachemys* “Estratégias e técnicas demonstrativas para a erradicação de cágados invasores” (LIFE09 NAT/ES/000529), iniciou-se pela primeira vez em Portugal a reprodução em cativeiro desta espécie, sendo o Centro de Recuperação e Investigação de Animais Selvagens (RIAS) um dos responsáveis por esta acção. Neste sentido, torna-se fundamental perceber qual o efeito que a incubação e posterior criação em cativeiro poderá ter no fitness dos animais nascidos deste modo, o que terá importância para a sua sobrevivência na natureza após libertação. O objectivo deste estudo foi comparar a fitness de animais nascidos em cativeiro com a de animais em estado selvagem. As fêmeas grávidas foram capturadas e mantidas em instalações próprias até à postura dos ovos, que foram incubados em condições semi-naturais (numa zona exposta às condições climáticas exteriores, numa área com cerca de 50% de exposição solar, 40% de sombra, 10% de água, e alguma vegetação rasteira) em que apenas a humidade do solo foi regulada. Estudaram-se três grupos diferentes de indivíduos. Os neonatos nascidos em cativeiro durante o Inverno de 2013 (n=18), os neonatos que emergiram na primavera na natureza (n=12), e ainda os indivíduos do primeiro grupo, mas mais tarde, com cerca de seis meses de idade. Realizaram-se três testes de performance: (i) Righting reflex (tempo que o indivíduo se demora a reposicionar, quando colocado de carapaça para baixo), (ii) Corrida (avaliando o tempo de reacção para fuga e as velocidades atingidas) e (iii) Natação (velocidade atingida), sendo que cada teste foi efectuado quatro vezes para cada indivíduo. Os primeiros resultados não apontam para a existência de diferenças significativas entre os neonatos nascidos em cativeiro e no estado selvagem. Os neonatos de cativeiro com cerca de seis meses de idade aparentam condições de fitness regulares e reflexos adequados. É assim possível inferir preliminarmente que o tipo de reprodução em cativeiro realizado, pode ser de facto uma reprodução modelo, uma ferramenta de sucesso quando o intuito é aumentar populações em decréscimo numa determinada área e cuja conservação é essencial.

***Emys orbicularis* captive breeding: a successful tool for population reinforcement?**

The European pond turtle (*Emys orbicularis*) is classified, in Portugal, as Endangered (EN), suffering a strong regression in the last century, not only of its population size but also of its distribution area. For the first time in Portugal, during the LIFE+*Trachemys* “Demonstration strategy and techniques for the eradication of invasive freshwater turtles” project (LIFE09 NAT/ES/000529), there was a captive breeding program for *E. orbicularis*, being the Wildlife Rehabilitation and Investigation Centre of the Ria Formosa (RIAS) one of the responsible entities for this action. To understand the effects that the incubation and subsequent rearing may have on the fitness of turtle hatchlings incubated in captivity is an important factor to consider when evaluating their survival prospects once released back in nature. The aim of this study was to compare the fitness of the animals born under captive breeding with that of the ones born in the wild. Pregnant females were captured and maintained in proper facilities until the eggs were laid. Those were incubated in semi-natural conditions (in an outdoors enclosure, in an area with about 50% of solar exposure, 40% of shadow, 10% standing water and some shrubs) in which only the soil humidity was regulated. Three different groups of individuals were studied. First, the hatchlings born in captivity during the winter of 2013 (n=18), later the hatchlings born in nature during the 2014 spring emergence of 2014 (n=12), and finally the first group individuals, evaluated later, when they were six months old. Three performance tests were conducted: (i) Righting reflex (the time a hatchling placed on its back took to right itself), (ii) Sprinting (the time during which the hatchling reacted and escaped, as well as the velocities it reached) and (iii) Swimming (reached velocities). Each test was performed four times for each hatchling. The first results do not show significant differences between the hatchlings born in captivity and the ones born in nature. The hatchlings born in captivity with six months old appeared to have normal fitness conditions and adequate reflexes. It is therefore possible to infer that the captive breeding program can indeed be a model for further efforts, a successful tool when the aim is to reinforce populations that are decreasing in a certain area in which conservation is crucial.

Emys orbicularis, captive breeding, Life+*Trachemys*, fitness, population reinforcement.

(P80) Una “nueva”, rápida y eficiente herramienta para estudios de conservación en herpetología: el perroALBERT VILARDELL-BARTINO^{1,2,3,4}, ELI HINOJAL⁵, SANTI VIDAL⁵¹Centre de Reproducció de Tortugues de l'Albera. Santuari de la Mare de Déu del Camp, 17780 Garriguella, Girona, Spain.²Grup d'Estudi i Protecció de les Tortugues. Can Laporta, Centre Cultural de l'Albera, 17700 La Jonquera, Girona, Spain.³Amics de les Tortugues de l'Albera. Santuari de la Mare de Déu del Camp, 17780 Garriguella, Girona, Spain.⁴Departament de Ciències Ambientals Universitat de Girona. Campus de Montilivi, 17071 Girona, Spain.⁵Más que Guau. 28341 Valdemoro, Madrid, Spain.

En el año 2009 se creó la primera unidad canina de conservación de flora y fauna (UCCFF) a nivel nacional y europeo, especializada en la localización de ejemplares de tortuga mediterránea (*Testudo hermanni*). Este quelonio, además de estar en peligro de extinción (EN), es una especie muy críptica con bajas densidades poblacionales, por lo que su localización muy difícil. Después de comprobar la eficacia de la UCRT en prospecciones realizadas en estudios de impacto ambiental para proyectos de minería, los perros participan en un censo de tortuga mediterránea en la sierra de la Albera que es donde se encuentra la última población considerada de origen natural de toda la Península Ibérica. La prospección tiene lugar en una zona donde se tenían marcadas 400 tortugas de las que en los últimos ocho años de estudio solo se habían conseguido localizar cinco ejemplares adultos. La UCRT localizó en una semana de trabajo 42 ejemplares adultos en un total de 100 ha. Los mismos perros son utilizados en otro proyecto para la localización de nidos de otra especie de quelonio como es el galápago europeo (*Emys orbicularis*). En la fase de entrenamiento los perros son capaces de localizar nidos en la fase final de incubación.

Recientemente, la UCRT ha participado en un operativo policial para localizar dos ejemplares adultos de tortuga de espolones africana (*Centrochelys sulcata*) que habían sido sustraídos del Centro de Reproducción de tortugas de la Albera, y los perros encontraron el sitio donde las tenían.

Finalmente los perros de la UCRT han sido entrenados para localizar otras especies (heces y pelo) como el oso pardo (*Ursus arctus*) y el lobo (*Canis lupus*), mostrando su efectividad en prácticas conjuntas con agentes rurales de la Generalitat de Catalunya, así como también en la localización de transmisores extraviados en estudios de radioseguimiento de rapaces, con la colaboración del Grupo de Rehabilitación de la Fauna Autóctona y su Hábitat (GREFA).

Los resultados obtenidos y su eficacia, fiabilidad, versatilidad y bajo coste avalan un esperanzador futuro en cuanto a la participación de perros en numerosos proyectos de conservación.

A “new”, fast and efficient tool in herpetological conservation studies: the dog

In 2009, we established the first European Flora and Fauna Conservation Canine Unit (UCCFF), specializing in Western Hermann's Tortoises (*Testudo hermanni*). This is an endangered (EN) species that is very difficult to detect because of their low population densities and their cryptic colors. After checking the effectiveness of UCCFF in environmental impact studies for mining projects, the dogs participate in a census of tortoises in the Albera Range, where the last native Iberian population is located. The survey takes place in an area where 400 tortoises had been marked, out of which only five specimens were located in the last eight years. The UCCFF located in a week 42 adult specimens in a total of 100 ha.

The same dogs are used in another project with the aim to locate nests of European pond turtle (*Emys orbicularis*). The dogs are able to locate 15 nests at the end of incubation season.

Recently, the UCCFF participated in a police operation to locate two adult African spurred tortoises (*Centrochelys sulcata*) that had been stolen from our breeding center, and the dogs found the site where they were kept.

Finally, UCCF dogs are trained to locate other species (feces and hair) as the brown bear (*Ursus arctus*) and wolf (*Canis lupus*), showing their effectiveness in joint exercises with rural agents of the Generalitat de Catalunya, and also in locating lost transmitters from radio-tracking studies of raptors in collaboration with the Native Fauna and its Habitat Rehabilitation Group (GREFA).

The results obtained and effectiveness, reliability, versatility and low cost guarantee a bright future of the trained dogs in many conservation projects.

Canine unit, conservation, detection, species, detecting dogs, conservation dogs.

(P81) “We are snake friendly”: una campaña de concienciación para mejorar la imagen de las serpientes

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Además de las habituales amenazas a la conservación que sufren los vertebrados, las serpientes tienen que enfrentarse también con su persecución directa, en aras de la “legítima defensa”, por parte de gente que se siente irrealmente amenazada. Este miedo, que a menudo se traduce en violencia, parece ser el resultado de una profunda ignorancia y falsas creencias en torno a la peligrosidad, historia natural y estado de conservación de las serpientes. La persecución humana de las serpientes es desenfrenada, así que para que cualquier esfuerzo por su conservación sea eficaz, las personas deben ser antes conscientes de su importancia y su carácter inofensivo. Así, la educación ciudadana para cambiar la actitud hacia estos animales puede ser un instrumento de conservación clave. Por otra parte, una actitud respetuosa hacia las serpientes generalmente se hace extensible a otros animales tan impopulares como ellas y, en general, a todo el medio ambiente. Con el fin de aumentar la sensibilización en torno a las serpientes, en 2013 (año chino de la serpiente) la Sociedad de Ciencias Aranzadi lanzó la campaña “We are snake friendly”, diseñada para persuadir a la gente de que no debe temer o matar a las serpientes, tratar de generar fascinación por estos reptiles y eliminar los muchos prejuicios y mitos que existen acerca de ellos. En definitiva, con el objetivo final de lograr que las serpientes sean consideradas una parte importante de nuestro patrimonio natural. Para ello se han llevado a cabo diferentes actividades: conferencias, exhibiciones de serpientes en vivo, exposiciones fotográficas, excursiones para la observación de serpientes en su entorno natural y talleres educativos para niños. La campaña inicialmente se ha centrado en “la gente del campo” (en su mayoría, agricultores y ganaderos) y niños. Nuestra experiencia ha mostrado que las serpientes son una gran herramienta para enseñar a la gente el valor de la conservación. Los niños casi invariablemente se quedan fascinados con ellas y proporcionarles (tanto a ellos como a los adultos) la oportunidad de observar, tocar e interactuar con animales que no tienen oportunidad de ver todos los días crea un ambiente idóneo para que aprendan más sobre ellos y aprecien el valor de estas especies. Además, tanto niños como adultos, en cuanto interactúan con serpientes vivas, suelen comenzar a perderles el miedo. Por último, dada su disposición y capacidad de aprender, los niños de entre ocho y doce años de edad son el público más adecuado para centrar este tipo de campañas.

“We are snake friendly”: an awareness campaign to put snakes in a good light

In addition to the usual conservation threats for vertebrates, snakes also have to face direct persecution, for the sake of “self-defense”, by people who feel unrealistically threatened by them. This fear, which often turns into violence, seems to be the result of deep ignorance and false beliefs about snake dangerousness, their natural history and conservation status. Human persecution of snakes is rampant, and people must be made aware of the importance and harmlessness of snakes to make any conservation effort effective. Thus, educating the public about snakes to change their attitude towards these animals may be a very important conservation tool. Moreover, a respectful attitude towards snakes usually becomes extensible to other unpopular animals and their natural environment.

Thus, in order to raise awareness of snakes, in 2013 (the Chinese year of the snake) Aranzadi Sciences Society launched the campaign “We are snake friendly”. It has been designed to persuade people to not fear or kill snakes by generating fascination for these reptiles and by eliminating prejudice and dispel myths about them. Finally, the ultimate goal is to encourage consideration of snakes as an important part of our natural heritage. For this purpose, we have carried out different activities: conferences, live snake displays, photographic exhibits, field trips for snake species observation in their natural environment, and children's educational workshops. The campaign has initially been focused on “country people” (mostly farmers and cattle ranchers) and children. Our experience has shown that snakes are great tools for teaching people the value of conservation. Children are almost always fascinated by snakes. Giving children (and adults) the opportunity to observe, touch, and interact with an animal they do not get to see every day creates a fantastic environment for them to learn more about and appreciate the value of these species. Moreover, both children and adults usually begin to lose their fear of these animals when they interact with live snakes. Finally, we have also come to the conclusion that given their willingness and capacity to learn, children eight to twelve years old are the most appropriate public for this kind of campaigns.

Snakes, awareness, campaign, children, education, conservation.

(P82) Situación y conservación del lagarto moteado de Tenerife, *Gallotia intermedia* en Guaza (Tenerife)

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El lagarto moteado de Tenerife, *Gallotia intermedia*, es la única de las tres especies de lagartos pertenecientes al clado "*simonyi*" que presenta dos poblaciones naturales en la isla: una en el macizo de Teno y otra en los acantilados costeros de la montaña de Guaza, donde su área de distribución se estima en unas 23,09 hectáreas. En el presente estudio se estimó el tamaño de la población de esta especie en Guaza mediante el método de marcaje-recaptura. Los datos obtenidos de las 35 sesiones de trampeo realizadas en cinco localidades muestran una densidad que varió entre 3 y 44 individuos por hectárea. De esta forma se ha estimado que la población está formada por entre 529 y 867 ejemplares. Se discuten comparativamente estas estimaciones y las obtenidas por otros autores.

Status and conservation of the Tenerife Speckled Lizard, *Gallotia intermedia* in Guaza (Tenerife)

The Tenerife Speckled Lizard, *Gallotia intermedia*, is the only out of the three species belonging to the "*simonyi*" clade that has two natural populations in the island, one in the Teno Massif and another one in the coastal cliffs of Guaza Mountain, where the distribution area is estimated in 23.09 hectares. In this study, the population size of this species in Guaza was estimated by using the mark-recapture technique. The data collected for the 35 trapping sessions, in five different locations, show a population density between 3 and 44 individuals per hectare. According to these data, the estimated population size ranges between 529 and 867 specimens. These data are confronted with those obtained by other authors

Gallotia intermedia, population density, mark-recapture, Canary Islands.



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