Zoologischer Anzeiger 261 (2016) 70–81

Contents lists available at ScienceDirect

Zoologischer Anzeiger

journal homepage: www.elsevier.com/locate/jcz

Zoologischer Anzeiger

On the origin of the recent herpetofauna of Sicily: Comparative phylogeography using homologous mitochondrial and nuclear genes

Matthias Stöck[a,*,1], Gianni Grifoni[b,1], Nabil Armor[c,d], Ulrich Scheidt[e], Alessandra Sicilia[f], Nicola Novarini[g]

[a] Leibniz-Institute of Freshwater Ecology and Inland Fisheries (IGB), Müggelseedamm 301, D-12587 Berlin, Germany
[b] University of Lausanne, Department of Ecology and Evolution (DEE), Biophore, CH-1015 Lausanne, Switzerland
[c] Biotechnology Institute of Monastir, Research Unit Genetics, Biodiversity and Bio-Resources Development, 5000 Monastir, Tunisia
[d] Ecology and Biogeography of Vertebrates (EPHE), CEFE CNRS, UMR 5175, 1919 Route de Mende, 34293 Montpellier Cedex 5, France
[e] Naturkundemuseum Erfurt, Grosse Arche 14, D-99084 Erfurt, Germany
[f] University of Palermo, Dipartimento di Biologia Animale, Via Archirafi,18, I-90123 Palermo, Italy
[g] Museo di Storia Naturale di Venezia, Santa Croce 1730, I-30125 Venezia, Italy

ARTICLE INFO

Article history:
Received 30 August 2015
Received in revised form 30 October 2015
Accepted 31 October 2015
Available online 4 November 2015
Corresponding Editor: Alexander Kupfer.

Keywords:
Africa
Italian Peninsula
Sicily
Divergence time
mtDNA
Nuclear DNA
Herpetofauna
Island biogeography

ABSTRACT

In contrast to oceanic, continental islands are expected to show less diversification and endemism and thus phylogeographic signatures of multiple colonization events from adjacent continents due to episodic connections by sea level changes. In order to test this situation for the herpetofauna of Sicily, we here focus on three amphibian and four reptile species-groups and investigate their phylogeographic relationships across the Sicily and Messina straits, where Plio-Pleistocene marine transgressions shortened the distances between (or connected) Sicily, North Africa and/or the Italian (Apennine) Peninsula. Using a multi-species, multi-marker phylogeographic approach (mitochondrial cytochrome *b*; 16S rDNA, nuclear intron of tropomyosin), we apply Bayesian and Maximum Likelihood phylogenetic methods and haplotype networks to examine the phylogenies, and to estimate divergence times from molecular data using the program BEAST. We recognize three colonization patterns: (i) Plio-Pleistocene colonization of Sicily from North Africa for the skinks *Chalcides chalcides* (1.8 Mya) and *Chalcides ocellatus* (0.61 My), (ii) Pleistocene colonization from the Italian Peninsula for the anurans *Pelophylax* spp. (0.81 Mya) and *Bufo bufo* (late Pleistocene), and (iii) recent (late Pleistocene to Holocene), natural or man-mediated out-of-Africa dispersal for the anuran *Discoglossus pictus* and out-of-Africa human introduction for the gekkonid lizards *Tarentola mauritanica* and *Hemidactylus turcicus*. The Sicilian herpetofauna shows phylogeographic signatures as typical of continental islands, with limited diversification and endemism. Colonization by terrestrial amphibians and reptiles from adjacent continents appears shaped by interactions of the active geo-marine history along with species' ecology and human intervention, including a widely neglected faunal contribution from Africa. On some small islands and in Tunisia, we found isolated local populations significant for conservation. Our results underline how only multispecies approaches involving ecologically diverse taxa are able to reveal the complexity of faunal contributions to large continental islands like Sicily.

© 2015 Elsevier GmbH. All rights reserved.

1. Introduction

In contrast to oceanic ones, continental islands are expected to show less diversification and endemism because they represent environmental and biological detachments of the mainland due to episodic connections by sea level changes (Williamson, 1981; Whittaker and Fernandez-Palacios, 2007; Cody, 2006; Johnson et al., 2012; Fabre et al., 2012). However, for specific, relatively old (continental) islands, origin and/or timing of colonization by organisms are of high interest to understand their biogeographic history and the underlying evolutionary processes.

The geological past and biogeography of the Mediterranean and its continental islands was first imprinted by the final Miocene period, the Messinian (7.25–5.33 Mya), which isolated the Mediter-

* Corresponding author at: Leibniz-Institute of Freshwater Ecology and Inland Fisheries (IGB), Müggelseedamm 301, D-12587 Berlin, Germany. Tel.: +49 30 64 181 629, fax: +49 30 64 181 663.
E-mail address: matthias.stoeck@igb-berlin.de (M. Stöck).
[1] These authors contributed equally to the paper.

http://dx.doi.org/10.1016/j.jcz.2015.10.005
0044-5231/© 2015 Elsevier GmbH. All rights reserved.

ranean Sea from the Atlantic and created salt lakes and deserts, serving as "near-land-bridges" (Krijgsman et al., 1999; lower sea level facilitating faunal exchanges) for terrestrial fauna between Africa, Europe and most islands ("Messinian crisis"; Krijgsman et al., 1999). Messinian exchange ended with a dramatic flooding through the Strait of Gibraltar (5.33 Mya; Garcia-Castellanos et al., 2009), leading to the probably oldest formation of islands in the recent Mediterranean. However, during the following Plio-Pleistocene glacial cycles, sea level changes (Rohling et al., 1998) brought North-African and European coasts closer again (Thiede 1978; Krijgsman et al., 1999), with episodes of potential faunal exchange on islands.

The resulting complex Messinian and Plio-Pleistocene phylogeographic relationships of North-African and Southwest-European (Iberian) faunas have been widely studied (e.g. Busack, 1986; Carranza et al., 2006b; Fromhage et al., 2004; Busack and Lawson, 2008). In contrast, phylogenetic history and biogeography and their timing in Sicily were much less examined. From early Pliocene to early Pleistocene, Sicily consisted of two islands (equivalent to modern north-central and south-eastern portions; van der Geer et al., 2010; Guglielmo and Marra, 2011). The Strait of Sicily was formed at the end of the Messinian, when the last well-documented connection between African mainland and a landmass that became part of Sicily was flooded. While Sicily and Tunisia are at present approximately 140 km apart, Pleistocene sea levels of −120 m (Thiede, 1978; Rohling et al., 1998; Dorale et al., 2010) have brought African and Sicilian coasts closer than ~50 km. "Stepping stone islands" (Stöck et al., 2008a) may have facilitated terrestrial animals in overcoming this sea barrier. This geological history has been documented by close faunal relationships in co-occurring taxa between N-Africa (from now: N-Africa) and Sicily from single animal species groups (e.g. Zangari et al., 2006; Giovannotti et al., 2007; Stöck et al., 2008a; Kornilios et al., 2010; Carranza et al., 2008), and between the Italian Peninsula (from now "IP") and Sicily (Giovannotti et al., 2007; Colliard et al., 2010; Stöck et al., 2008a,b; Kindler et al., 2013; and refs. therein). Several other hypotheses on the colonization patterns of Sicily remained partly speculative; mostly based on taxonomic assignments and limited palaeontological data (cf. SIB, 2011). However, comparative molecular-based estimations of times of divergence with sufficient samples sizes from N-Africa, Sicily and the IP are scarce, and for Sicily no multispecies study on the timing of the origins of the terrestrial herpetofauna has been undertaken. In doing so, we expect to find single or multiple dispersals across the Strait of Sicily (out-of-Africa) and/or the region that formed the Italian (Apennine) Peninsula (out-of-Italy). Assuming no overseas dispersal, relationships could be explained by three major scenarios: (i) a Messinian land-connection (>5.3 Mya); (ii) post-Messinian (=Plio-Pleistocene) "near-land-bridges" or "stepping stone islands" at low sea levels (<5.3 Mya to 10 kya); (iii) recent (Holocene) human introductions (<9 kya). As typical of continental islands, for any given species, scenarios might also be more complex, involving both Plio-Pleistocene out-of-Africa and Pleistocene out-of-Italy events, as shown for green toads (Stöck et al., 2008a; Colliard et al., 2010).

2. Material and methods

2.1. Sampling

In order to examine potentially complex patterns, we use homologous mtDNA and nuDNA markers from three terrestrial amphibian and four reptile 'species-groups' (term used throughout the paper to circumscribe more or less closely related taxa without implications on taxonomy or depth of relationships). For several of these species-groups we analysed a greater number of samples from different, and often more, localities in the target region than previous studies. Sampling was undertaken to fulfill four major prerequisites: species-groups should (i) be represented in all three regions of interest (Italian mainland, Sicily and N-Africa) or at least on Sicily and one of the other two, (ii) display different ecologies and susceptibilities to man-mediated introduction, expected to reflect a spectrum of colonization histories of Sicily; (iii) include as many sampling localities as possible, especially on Sicily, and (iv) include samples from the islands in the Strait of Sicily, if present.

Species-groups selected for analyses (distributions: Figs. 1–3) were painted frogs (*Discoglossus pictus* Otth, 1837, Anura, Discoglossidae); common toads (*Bufo bufo* (Linnaeus, 1758) and *B. spinosus* Daudin, 1803, Anura, Bufonidae); Palearctic water frogs (*Pelophylax* kl. *hispanicus* (Bonaparte, 1839), *P. bergeri* (Günther, 1985) and *P. saharicus* (Boulenger in Hartert, 1913), Anura, Ranidae); ocellated skinks (*Chalcides ocellatus* (Forskål, 1775), Squamata, Scincidae); three-toed skinks (*Chalcides chalcides* (Linnaeus, 1758), Squamata, Scincidae); Turkish geckos (*Hemidacylus turcicus* (Linnaeus, 1758), Squamata, Gekkonidae); and Moorish wall geckos (*Tarentola mauritanica* (Linnaeus, 1758), Squamata, Gekkonidae). Samples, were collected from multiple localities in N-Africa, Sicily, the IP and on Mediterranean islands in the Strait of Sicily (Pantelleria, Linosa, Lampedusa and Malta), or obtained from scientific collections (Table S1). Published sequences (GenBank) were used for outgroups (Table S1 for details on specimens examined, and museum vouchers; File S3 for all sequence data and alignments).

2.2. DNA extraction, amplification, cloning and sequencing

Genomic DNA was extracted from ethanol preserved muscle tissue, tail tips or frozen buccal swabs using the DNeasy Blood & Tissue Kit (Qiagen). Two mitochondrial markers were amplified in all species (Table 1). In amphibians, we PCR-amplified most of the mitochondrial cytochrome *b* (from now: cyt-b) using primers MVZ15-L_AmphCytb/Ptacek2-H (94 °C, 7 min, denaturation; cycle [94 °C, 40 s, denaturation; 46 °C, 30 s, annealing; 72 °C, 90 s, extension] 40 times; 72 °C, 10 min, final extension; Moritz et al., 1992). For lizards, cyt-b was amplified with primers Rep_rGlu-1L/Rept_rcytb-1H (94 °C, 1 min, denaturation; cycle [94 °C, 30 s, denaturation; 48 °C, 60 s, annealing; 72 °C, 60 s, extension] 38 times; 72 °C, 5 min, final extension). We further amplified the 16S rDNA, for amphibians using primers 16Sar-L/16Sbr-H (PCR: 95 °C, 3 min, denaturation; cycle [94 °C, 45 s, denaturation; 55 °C, 45 s, annealing; 72 °C, 60 s, extension] 35 times; 72 °C, 5 min, final extension; Kocher et al., 1989); for reptiles with primers L2606/H3056 (PCR: 96 °C, 2 min, denaturation; cycle [94 °C, 30 s, denaturation; 55 °C, 45 s, annealing; 72 °C, 90 s, extension] 38 times; 72 °C, 5 min, final extension; Kocher et al., 1989).

We chose a nuclear marker, which could be commonly amplified in all amphibian and reptile species and that was expected to provide appropriate phylogeographic signals: intron 5–6 of nuclear *alpha*-tropomyosin (Friesen et al., 1999). To amplify this marker, we used primers of Friesen et al. (1999); "frog version" for amphibians, "bird version" for *Chalcides*; we restricted analyses for geckos on mtDNA). PCR conditions for "frogs" were adapted: 95 °C, 1:30 min; cycle [94 °C, 30 s; 59.0°, 30 s; 72 °C, 45 s] 30 times; 72 °C, 5 min (Stöck et al., 2008a); PCR conditions for reptiles were: 94 °C, 1:30 min; cycle [94 °C, 30 s; 55°, 30 s; 72 °C, 45 s] 30 times; 72 °C, 5 min.

Mitochondrial PCR products were sequenced directly. Nuclear amplicons were cloned using the pGEM®easy system (Promega) as described (Stöck et al., 2008a). To identify both alleles in potential heterozygotes prior to sequencing, 1 μl of DNA from 10 positive colonies (clones) was re-amplified (primers M13F/M13R), products

Download English Version:

https://daneshyari.com/en/article/2790490

Download Persian Version:

https://daneshyari.com/article/2790490

Daneshyari.com