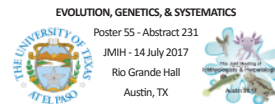


From the Floor, to the Canopy: Comparative Phylogeography of Two Sympatric Chameleon Species in Central Africa's Albertine Rift

Daniel F. Hughes^{1,2}, Wilber Lukwago³, Mathias Behangana³, Michele Menegon⁴, J. Maximilian Dehling⁵, Jan Stipala⁶, Colin R. Tilbury⁷, Krystal A. Tolley^{7,8}, Chifundera Kusamba⁹, and Eli Greenbaum^{1,2}

¹Department of Biological Sciences, ²UTEP Biodiversity Collections, University of Texas at El Paso, El Paso, Texas, USA; ³Makerere University, Kampala, Uganda; ⁴Museo Tridentino di Scienze Naturali, Trento, Italy; ⁵Universität Koblenz-Landau, Koblenz, Germany; ⁶University of Exeter, Cornwall, United Kingdom; ⁷University of Stellenbosch, Stellenbosch, South Africa; ⁸South African National Biodiversity Institute, Cape Town, South Africa; ⁹Centre de Recherche en Sciences Naturelles, Lwiro, République Démocratique du Congo



Introduction

The Albertine Rift (AR) is one of the most diverse highland regions in continental Africa, yet it is not well known whether taxa with overlapping ranges share diversification patterns.

Kinyongia adolffriderici and *Rhampholeon boulengeri* are generally sympatric and share habitats in the AR, yet they differ in microhabitat use and foraging behavior. *Rhampholeon boulengeri* is largely terrestrial and forages on the forest floor, whereas *K. adolffriderici* is mostly arboreal and forages high in the forest canopy.

We investigated the relative roles of environmental and taxon-specific factors in influencing spatiotemporal patterns of genetic diversity in two sympatric chameleon species.

Methods

Samples and sequencing. Forty-six samples of *R. boulengeri* and 14 samples of *K. adolffriderici* were collected during surveys in the AR from 2008–2016, including topotypic material. Two mitochondrial (16S, ND2) and one nuclear (RAG-1) gene were sequenced using standard protocols.

DNA data. Chromatographs were interpreted in SEQMAN PRO and alignments were made using MUSCLE in MESQUITE. We made minor manual adjustments in MACCLADE. We used PHASE in DNASP to phase haplotypes for RAG-1. The phased sequences were used in species-tree analyses.

Gene trees. Maximum-likelihood analyses were conducted with the GTRGAMMA model in RAXML using the rapid bootstrap algorithm with 1,000 replicates. Bayesian-inference analyses were conducted in MRBAYES and run for 20 million generations sampling every 1,000 with 25% burn-in. We used PARTITIONFINDER to establish the best model of evolution. The concatenated data set included nearly all species per genus and analyses were run on the CIPRES Science Gateway.

Species trees and divergence dating. We used *BEAST in the program BEAST to estimate species trees for focal taxa. We used BEAST to estimate divergence dates with an uncorrelated log-normal relaxed clock model and a Yule prior. We included all chameleon genera and 22 squamate taxa plus *Sphenodon*. Fossil calibrations were placed on nine nodes and secondary calibrations on five nodes. We analyzed five concurrent runs of 200 million generations sampled every 20,000. All runs were combined using LOGCOMBINER with 10% burn-in.

See Hughes et al. (2017) for detailed methods

Results

1. Gene trees

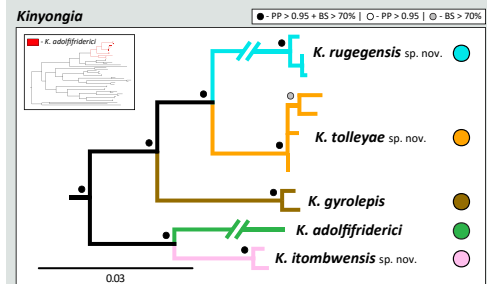


Figure 1. Maximum-likelihood phylogeny of *Kinyongia adolffriderici* from the Albertine Rift, Central Africa.

2. Dated species trees

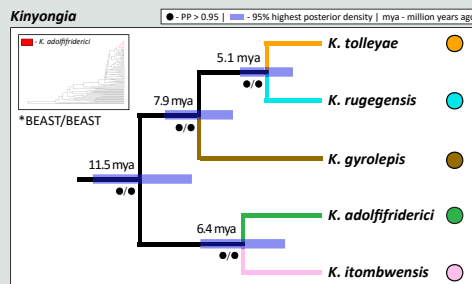


Figure 3. Species tree with mean diversification dates for the *Kinyongia adolffriderici* species complex.

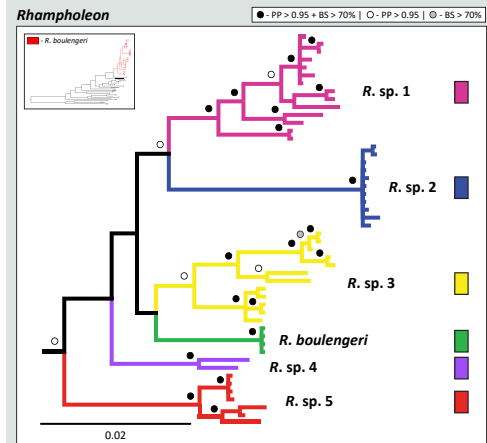


Figure 2. Maximum-likelihood phylogeny of *Rhampholeon boulengeri* from the Albertine Rift, Central Africa.

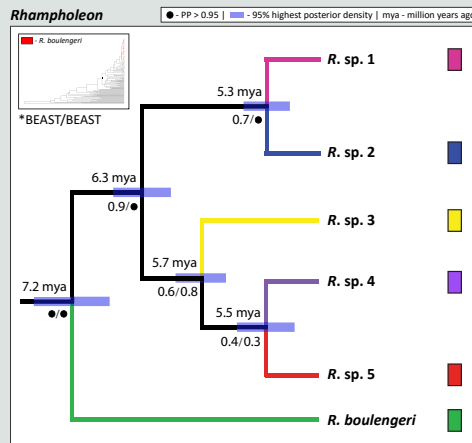


Figure 4. Species tree with mean diversification dates for the *Rhampholeon boulengeri* species complex.

3. Phylogeography

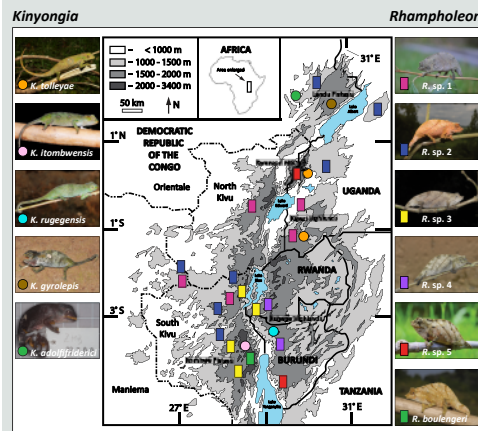


Figure 5. Map of the Albertine Rift showing sampled populations for *Kinyongia* (circles) and *Rhampholeon* (squares).

4. Elevational zonation

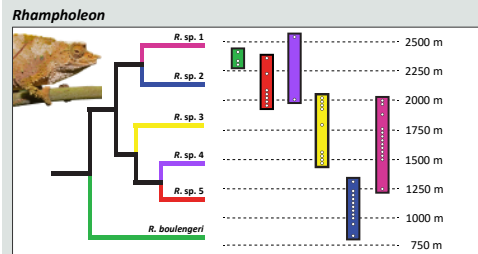


Figure 6. Species tree with elevational zonation of six *Rhampholeon* species from the Albertine Rift, Central Africa.

Conclusions

- Three new species of forest chameleons (*Kinyongia*) were formally described from the AR (Hughes et al., 2017) and five new species of pygmy chameleons (*Rhampholeon*) await description from the AR (Hughes et al., in review).
- The Miocene was identified as an important period for diversification of chameleons in the AR, which is linked to the fragmentation of African forests over this period (Kissling et al., 2012).
- Diversification patterns for *Kinyongia* were consistent with the model of allopatric speciation driven by forest fragmentation in the Miocene.
- Diversification events at the genus level for *Rhampholeon* generally fit the vicariance-driven speciation model, however, it does not fully explain the rapid radiation across the Miocene–Pliocene boundary in the *R. boulengeri* complex with several species distributions that span traditional biogeographic barriers in the AR.
- Several species in the *R. boulengeri* complex are sympatric but not syntopic because they occur in largely non-overlapping elevational zones for which adaptations to novel physical factors (e.g., temperatures or climatic niches) likely initiated processes leading to parapatric speciation.
- We found that the Gradient Speciation Hypothesis (i.e., parapatric speciation via niche differentiation) is more appropriate to explain diversification in the *R. boulengeri* complex.
- Traditional diversification scenarios are inadequate to explain the high levels of cryptic diversity we found in *R. boulengeri* and *K. adolffriderici*, which differ in ecomorphology.
- Comparative approaches to phylogeography are needed to assess both environmental and ecological factors promoting species diversity.

References

Hughes, DF, et al. 2017. Integrative taxonomy of the Central African forest chameleon, *Kinyongia adolffriderici* (Sauria: Chamaeleonidae), reveals underestimated species diversity in the Albertine Rift. *Zoological Journal of the Linnean Society* (published online - 20 May 2017).

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Kissling, WD, et al. 2012. Cenozoic imprints on the phylogenetic structure of palm species assemblages worldwide. *PNAS* 109: 7378–7384.