

From Antarctica or Asia? New colonization scenario for Australian-New Guinean narrow mouth toads suggested from the findings on a mysterious genus *Gastrophrynoides*

Abstract

Background: Microhylidae is a geographically widespread family of anurans. Although several extensive molecular analyses have attempted to elucidate their subfamilial relationships, and correlate these with Mesozoic and Cenozoic continental drifts, consensus has not been reached. Further, generic level relationships have not been well investigated in some microhylid subfamilies, and therefore subfamilial affiliations of some genera are still unclear. To elucidate the phylogenetic positions of two mysterious Asian genera, *Gastrophrynoides* and *Phrynella*, and to better understand the trans-continental distributions of microhylid taxa, we performed molecular phylogenetic and dating analyses using the largest molecular dataset applied to these taxa to date.

Results: Six nuclear and two mitochondrial genes (approx. 8 kbp) were sequenced from 22 microhylid frog species representing eight subfamilies. The maximum likelihood and Bayesian analyses could not fully elucidate the subfamilial relationships, suggesting a rapid radiation of these taxa between 85 and 66 million years ago. In contrast, generic relationships of Asian microhylines were generally well resolved. Conclusion: Our results clearly showed that one of two problematic Asian genera, *Phrynella*, was nested in the clade of the Asian subfamily Microhylinae. By contrast, *Gastrophrynoides* occupied the most basal position of the Australian-New Guinean subfamily Asterophryinae. The estimated divergence of *Gastrophrynoides* from other asterophryine was unexpectedly around 48 million years ago. Although a colonization scenario via Antarctica to the Australian-New Guinean landmass has been suggested for Asterophryinae, our finding suggested a novel colonization route via Indo-Eurasia.