

# Do large molecular sequence divergences imply an early closure of the Isthmus of Panama?

Bacon et al. (1) “reject a 3.5 Ma assumption” for the closure of the Central American Seaway, a conclusion that echoes earlier studies that established that the “bulk [of marine ‘geminate’ or ‘transisthmian’ species on opposite sides of the Isthmus] were split at some point during the long period of geological upheavals associated with the rising Isthmus,” ~15–2.8 Ma (2). However, Bacon et al. (1) also argue that large DNA sequence divergences between some transisthmian lineages reveal that the Isthmus geographically isolated marine species as early as 24 Ma.

Although Bacon et al. (1) consider the impact of random error on their results, they ignore several well-known sources of error that will systematically bias their results toward overrepresentation of older divergences that were not Isthmus-driven. The critical assumption used by Bacon et al., that the temporal distribution of divergence times across the Isthmus reflects the tempo of speciation driven by the Isthmus, is naive to the fact that as many as 70% of Caribbean taxa were lost to extinction after seaway closure (ref. 2 and references therein). The loss of one member of a geminate pair will cause the erroneous assignment of the next most closely related species as a sister-species, but with an older molecular divergence. High rates of Caribbean origination may have preserved the age of some transisthmian speciation in molecular phylogenies (2), but without the fossil record it remains impossible to distinguish

relicts of extinction from species formed by the Isthmus (2, 3). Similarly, incomplete taxon sampling has led to incorrect assignments of geminates that are not actually sister-taxa, including at least one “false” geminate pair (4) mistakenly included by Bacon et al. (1). Much of the data used by Bacon et al. are from studies that were not focused on testing geminate relationships. However, the bias toward finding ancient divergences that is evident in the few studies that have addressed sampling issues should give pause to biogeographers that take patterns of diversity in molecular phylogenies at face value.

Most of the studies considered by Bacon et al. (1) were also based on a single (mtDNA) locus, which often leads to overestimates of species’ divergence times because gene divergences usually predate population divergences. The variance around single locus estimates caused by stochastic lineage sorting cannot be assessed without multilocus data. For recently diverged snapping shrimp geminates, mtDNA and multilocus divergences (the latter oddly not considered by Bacon et al.) over the last 10 Ma are uncorrelated, indicating that most of the variance in mtDNA divergence times over this time period is probably explained by sorting of ancestral polymorphisms (5). For bivalve geminates, the addition of a nuclear locus to mtDNA data cut geminate divergence times by more than 50%, from 15 Ma (mtDNA) to 4.8 Ma (multilocus) for one pair (4). Clearly, the timing of these splits

and the magnitude of this error will introduce a significant source of bias into analyses that rely on single-gene divergences as a proxy for population divergences.

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**1** Bacon CD, et al. (2015) Biological evidence supports an early and complex emergence of the Isthmus of Panama. *Proc Natl Acad Sci USA* 112(19):6110–6115.

**2** Lessios HA (2008) The great American schism: Divergence of marine organisms after the rise of the Central American Isthmus. *Annu Rev Ecol Syst* 39(1):63–91.

**3** Marko PB, Jackson JBC (2001) Patterns of morphological diversity among and within arcid bivalve species pairs separated by the Isthmus of Panama. *J Paleontol* 75(3):590–606.

**4** Marko PB, Moran AL (2009) Out of sight, out of mind: High cryptic diversity obscures the identities and histories of geminate species in the marine bivalve subgenus *Acar*. *J Biogeogr* 36(10):1861–1880.

**5** Hurt C, Anker A, Knowlton N (2009) A multilocus test of simultaneous divergence across the Isthmus of Panama using snapping shrimp in the genus *Alpheus*. *Evolution* 63(2):514–530.

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