

Divergence dates for planktic foraminiferal cryptic species estimated from multi-genes

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Accurate estimation of divergence dates advances understanding of organismal evolution and assesses the effects of climatic and geological conditions on patterns of speciation and geographic distribution among organisms. Planktic foraminifera is one of the model organisms for divergence time estimation due to good fossil records, which are applicable to give time constraints during the dating analysis. In recent molecular phylogenetic studies, multiple genetic types have been found in a single morphospecies. In present study, we focused on a species *Pulleniatina obliquiloculata*, which mainly distribute in the subtropical-tropical water of the Indo-Pacific Oceans, having three genetic types (types I, IIa, and IIb). Intriguingly, these three types show longitudinal clines in frequencies within a narrow latitudinal range in the Indo-Pacific Warm Pool (IPWP) area. We estimate divergence time of these three genetic types corresponding with geological events that generated the oceanic circulation system in the IPWP area.

Firstly, we increased sequence data, which are complete small and large subunit ribosomal DNAs (SSU, LSU), of three genetic types of *P. obliquiloculata* and two outgroup species *Neogloboquadrina dutertrei* and *Globorotalia inflata*. Each single gene data-set was applied to maximum likelihood estimation through the program multidivtime (Bayesian molecular dating using PAML). Three patterns with single-gene data-sets (SSU and LSU) and multi-gene data set (SSU + LSU) were conducted for MCMC analyses using one maximum constrain (the first appearance date of *Neogloboquadrina acostaenesis* as the common ancestral lineage for both *P. obliquiloculata* and *N. dutertrei*). Divergence ages based on multi-gene analysis were estimated with more narrow credibility intervals (CI) than single-gene analyses. Estimated ages were similar among these data-sets: divergence of the lineages *P. obliquiloculata* and *N. dutertrei*, between 10.5 and 10.1 Ma, first divergence of genetic types (I and II) between 4.0 and 3.7 Ma, and latest divergence of genetic types (IIa and IIb) between 2.0 and 1.1 Ma. Divergence time of three genetic types suggests that their longitudinal clines have been established according to development of the IPWP system: distinct water masses were formed between the Pacific and Indian-sides and subtropical gyre system was emphasized in this area. The present results indicate that changes of the oceanic circulation system impact on the geographic patterns of migration and divergence in pelagic organisms.

Keywords: planktic foraminifera, divergence time estimation, LSU, SSU