

## Is a Living Fossil about to Go Locally Extinct? No Mitochondrial Genetic Variation in Horseshoe Crab Juveniles *Tachypleus tridentatus* at Haomeiliao Nature Reserve, Budai, Taiwan

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Habitat destruction caused by landfill projects, breakwater deposition, and fishery port construction has led to the loss of 55% of the natural coastline of Taiwan, particularly intertidal areas of the west coast (CPAMI 2007). As a consequence, significant declines in the populations or local extinctions of certain species are expected, although the extents of these impacts are poorly understood. In an effort to promote coastal conservation measures, the tri-spine horseshoe crab, *Tachypleus tridentatus*, was selected as a "flagship species" to study the distribution and population genetics. Surveys conducted between 2003 and 2007 indicated the existence of one small population in the Haomeiliao Nature Reserve (23°22'01"N, 120°07'49"E) of the Longgong River, which represents the only nursery ground for the horseshoe crab along the west coast of Taiwan (Yang et al. unpubl. data). However, the Longgong River estuary was blocked by expanding construction of Budai Harbor in 2004 (Fig. 1). Genetic data revealed by the mitochondrial AT-rich region (369 bp in full length, Yang et al. 2007) and cytochrome *c* oxidase subunit I (COI) (598 bp in partial length, Folmer et al. 1994) indicated that both nucleotide ( $\pi$  %) and haplotype diversities (*HD*) of the Budai population (CB) in 2007 were 0. This unexpected genetic homogenization is worse than those horseshoe crab populations that are isolated within natural embayments resulting in decreased genetic diversity due to restricted gene flow and the bottleneck effect (Fig. 2) (Pierce et al. 2000, Yang et al. 2007). This was further supported when the small population size at Budai was compared to the density of *T. tridentatus* juveniles in Hsiashu, Kinmen in 2005 and 2006 which averaged 0.169 individuals/m<sup>2</sup> based on annual transect surveys (Hsieh and Chen 2009), but the density was too low to be detected at Budai even based on monthly transect surveys throughout 2007 (Yang et al. unpubl. data). Loss of genetic variability and disruption of current flow by harbor construction and other developmental projects near the Haomeiliao Nature Reserve and along the west coast of Taiwan indicate that this remnant population is not sustainable and may be about to become extinct. This might be the case, not only for *T. tridentatus*, but also for other aquatic assemblages along the west coast of Taiwan. <http://zoolstud.sinica.edu.tw/Journals/48.6/737.pdf>

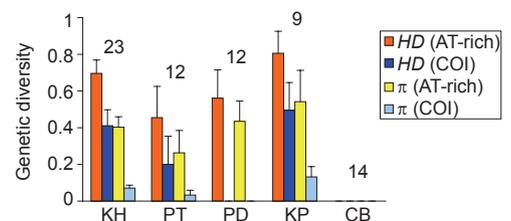
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**Fig. 1.** Longgong Estuary (23°22'01"N, 120°07'49"E) (A) and a juvenile horseshoe crab *Tachypleus tridentatus* (B) on a mudflat at Budai. (A) Budai Harbor is located to the north of and blocks the estuary. The red arrowhead indicates the nursing ground and sampling site of horseshoe crab juveniles.



**Fig. 2.** Genetic diversity of the mitochondrial AT-rich region and cytochrome *c* oxidase subunit I (COI) of *Tachypleus tridentatus* around Taiwan. Numbers of individuals are listed above the bars. Abbreviations of sampling sites are as follows: KH, Hsiashu at Kinmen; PT, Tiexianwei in the Penghu Is.; PD, Dongwei in the Penghu Is.; KP, Pengjiayu in Keelung; CB, Budai in Chiayi County. Numbers above the bars indicate the sample size. The PT population in a natural embayment has lower nucleotide diversity which is only higher than the CB population with 0 genetic diversity (both *HD* and  $\pi$ ). AT-rich region data from KH, PD and PT are from Yang et al. (2007); others are presented in this study.

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