

Chapter 1: General Introduction

The taxonomic status of the genus *Macrobrachium* Bate, 1868 belonging to Superclass Crustacea: Class Malacostraca: Superorder Eucaridea: Order Decapoda: Suborder Pleocyemata: Infraorder Caridea: Superfamily Palaemonioidea: Family Palaemonidae: Subfamily Palaemoninae: Genus *Macrobrachium* (Bowman and Abele 1982). Genus *Macrobrachium* is a highly diverse group of decapod crustaceans throughout tropic and warm temperate area of the world, have originated from marine ancestors and subsequently migrated towards freshwaters in more than one wave, hence its members are known to inhabit the entire range of habitats from purely marine areas to inland hill streams and impounded waters (Tiwari 1955, Shokita 1979, Jalihal et al. 1993). To date, approximately 210 species are recognized (Short 2004); the high number of species is probably still an underestimate of the genus's diversity; and there are numerous yet undescribed cryptic species (Chace and Bruce, 1999; Wowor and Choy 2001, Cai and Ng 2002, Cai et al. 2004, Short 2004).

The life cycle of *Macrobrachium*, as the many crustacean species, consists of egg, planktotrophic larva, post-larval and the adult phases. *Macrobrachium* can be ecologically separated into two groups: most species are widely distributed and require a certain saline concentration (i.e., 10‰~35‰) to complete their larval (planktonic stage) development, as euryhaline species. Euryhaline species have the spawning and diurnal migration between freshwater and estuary; others are land-locked species, with limited distributions and complete their entire life cycle in freshwater (Holthuis 1950, Johnson 1973, Shokita 1979). As waves of *Macrobrachium* migrated towards freshwaters, the prawns gradually evolved several adaptive features during the process of acclimatization to freshwater environments, like an increase of egg size, reduction in egg numbers, large-sized benthic larvae, and severance of salinity dependence for metamorphosis. With abbreviated larval development, the prawn benefits by reducing both the number of larval stages and the duration of the larval period, which in turn increase the

survival percentage of larvae (Shokita, 1979, Jaliha et al. 1993). The abbreviated development of larvae in land-locked species was suggested to be a result of selective pressures for becoming established in freshwater environments (Shokita 1979, Magalhães and Walker 1988), and is a multiple convergent phenomenon overriding phylogenetic relationships even above the generic level (Magalhães and Walker 1988). On the contrary, Pereira and Garcia (1995) suggested that since primitive palaemonids like *Troglocubanus*, *Palaemonetes* and *Pseudopalaemon*, possess abbreviated development, which could be considered a primitive trait, and the abbreviated development took place early in the origin of the family Palaemonidae, rather than being a recent process.

The taxonomy and phylogenetic inferences have until recently been based exclusively on comparisons of external morphological characters. The rostrum and the second pereiopod, two main morphological features for species identification, are often highly variable within species (Holthuis 1950, Johnson 1973). Because of the conservativeness of morphological characteristics, much debate has surrounded the systematic relationships of many species within this group (Holthuis 1950 1952, Johnson 1973, Pereira 1997). Beside the species identify, some species groups were proposed based on morphological similarities (synapomorphy) (Holthuis 1950, Johnson 1973). The phylogenetic significance of these groupings remains to be tested. Apart from their taxonomy, the phylogenetic affinities among freshwater prawns are poorly understood. Based on the morphological characters, Pereira (1997) carried out the first phylogenetic study on the family Palaemonidae. Results from his cladistic analysis are thus in need of being further tested using more taxa, and by applying more characters, including genetic data. In recent years, Murphy and Austin (2002 2003 2004 2005) published a series of results for the phylogeny of *Macrobrachium* species based on the mitochondrial DNA fragment of the large subunit (16S) rRNA gene. Their studies showed some interesting results and led to the generic clarification of some local species.

The East Asia has a larger landmass within the tropics and subtropics, has a longer history of overland connection with tropical areas having rich species pools (Hamilton 1983, Guo 1998). Its freshwater fish fauna represent one of the richest ichthyofauna in the Sino-Indian region, which is in agreement with its geographical complexity (Koltelat 1989, Banarescu 1991). East Asia also supports high species diversity of plant taxa (Guo 1998, Qian and Ricklef 2000). The climatic changes and geographic heterogeneity have played major roles in the diversification and speciation of East Asia's biota and is a superior continental model for studying increases in regional diversity through allopatric speciation (Qian and Ricklef 2000).

Previous studies of the freshwater prawns in East Asia comprised regional species surveys or zoogeographic distributions (Yu 1936, Dai 1984, Liang 1986, Hwang and Yu 1982 1983, Hayashi 2000a b c). There are about 37 species found and there is a relatively high level of endemism. Shokita (1979) inferred the speciation and origin of land-locked freshwater prawns based on prawns from the Ryukyus, and discussed the biogeography of genus *Macrobrachium* with special reference to larval dispersal (Shokita 1985). Some population studies of *M. nipponense* were discussed based on allozyme variations and reproductive traits (for review see Mashiko and Numachi 2000), but little work has been carried out on the phylogenetic relationships of freshwater prawns in East Asia.

For the molecular systematic study of this freshwater prawn *Macrobrachium*, I attempted, using mtDNA markers, to investigate the phylogeny and evolution of land-locked species of the genus *Macrobrachium*, and to elucidate the phylogenetic relationships of East Asian species.

In phylogeography and genetic structure study of the land-locked prawn *M. asperulum*. The geographical distribution and phylogenetic relationships of animal mtDNA variants reflect both contemporary gene flow and biogeographical events in the history of a species (Avise 1994). A complex history of geological events and climate has shaped current phylogeographical patterns (Hewitt 1996, Avise 2000). The land-locked species with abbreviated larval

development showed limited dispersal ability compared to the euryhaline species. The spatially disconnected nature of river systems confers unique phylogeographical constraints on freshwater fauna (Avice 2000). The dispersal of freshwater species between river drainages is normally extremely limited and is dependent on direct connections between drainage basins, such as drainage re-arrangements, short-term connections between drainages, and sea-level changes (Bilton et al. 2001). For this reason, the distribution of freshwater fauna is more likely to reflect historical events than will that of terrestrial species (Kotlik and Berrebi 2001). Historical phylogeographical analyses of freshwater species permit strong inferences regarding the biotic and geological evolution of a region (Lundberg 1993, Berminham and Martin 1998).

Taiwan is a subtropical island, located between the Eurasian continent and the Philippine Sea basin. The gradual uplifting of the north-south extension of the Central Mountain Range (CMR) as well as lesser mountain ranges and plateaus have produced prominent watersheds and act as dispersal barriers for the freshwater fauna in Taiwan (Tzeng 1986). The cyclic separation and rejoining of Taiwan and the Asian mainland caused by the sea level changes due to Pleistocene glaciations provided opportunities for dispersal of taxa between the two areas (Shen 1997, Ota 1998). The distribution patterns of freshwater fauna on this island have certainly been influenced by geological events and climatic oscillations. Therefore, Taiwan provides an excellent natural laboratory to study biogeographical hypotheses of contemporary phylogeographical patterns.

The freshwater prawn, *Macrobrachium asperulum* (von Martens 1868), is a land-locked species inhabiting a wide range of water bodies. The prawn is an abundant freshwater macroinvertebrate species, and has a pan-island distribution in Taiwan (Hwang and Yu 1983). Because of its life cycle, gene flow between different river systems is limited and stronger genetic subdivisions among populations are expected in situations in which dispersal is limited (Shokita 1985, de Bruyn et al. 2004). Due to its wide distribution and limited capacity for

dispersal, *M. asperulum* can be considered an interesting subject for use in evaluating historical events on population structures of freshwater species. In this study, we used mtDNA markers to examine the phylogeographical patterns and genetic structure of *M. asperulum* in Taiwan. This study attempts to infer the potential roles of geographical events and historical climatic on the phylogeographical patterns and genetic structures of freshwater species of Taiwan.

