

Analysis of connectivity in the Skunk Clown Fish using a combination of microsatellite and mitochondrial genetic markers

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Like most coral reef associated organisms, *Amphiprion akallopisos* (Skunk Clown Fish), which lives in mutualistic symbiosis with two species of giant sea anemones, is unable to migrate from one coral reef to another during its adult life stage. Dispersal between reefs in the patchy coral reef environment is limited to an approximately two-week long pelagic larval stage. Because small pelagic larvae are extremely difficult to track in the open ocean, population genetics is a useful technique to evaluate gene flow and connectivity between populations of coral reef associated organisms. Insight into connectivity between populations of coral reef associated organisms is vital for a correct spacing of Marine Protected Areas, needed for the management and conservation of coral reefs. *Amphiprion akallopisos* has a disjunct distribution, occurring in the Western Indian Ocean (WIO) and the Eastern Indian Ocean (EIO), separated by more than 4,500km of open sea. This study aims to estimate levels of gene flow between these WIO and EIO populations and, on a smaller geographical scale, to reveal connectivity between populations within the WIO. A combination of mitochondrial and nuclear genetic markers is used for this approach. This, by providing information drawn from several parts of the genome, reduces genetic sampling error. But it also provides us with the opportunity to compare data from a relatively slow evolving genetic marker (Control Region of the mitochondrial DNA) with data from fast evolving microsatellite markers, revealing more recent information on gene flow. Using a 337 base pair long sequence of the Control Region, a first analysis was performed on 263 samples of *A. akallopisos* individuals from 17 different sites in both the WIO (Kenya, Tanzania, and Madagascar) and the EIO (Indonesia). Strong population structure ($\phi_{st} = 0.28$; $P < 0.001$), and strong differentiation ($\phi_{ct} = 0.61$; $P < 0.001$) was encountered between the EIO and the WIO populations, indicating very low gene flow. Within the WIO, low but significant population structure ($\phi_{st} = 0.016$; $P < 0.01$) was observed, but no clear genetic break could be detected. Selective neutrality tests (Tajima's D and Fu's Fs) indicate a derogation from selective neutrality in the WIO, and suggest this population underwent a rapid expansion in the past (raggedness index $r = 0.068$; $P = 0.82$). During the coming months, the dataset will be extended with samples of individuals from 5 to 10 different sites along the Mozambican coast, providing an improved geographic coverage for the WIO population. Furthermore, a total of 38 microsatellite markers described in literature for species belonging to the genus *Amphiprion* are tested in *A. akallopisos*.