

**Population Fragmentation in the Murray Hardyhead
Craterocephalus fluviatilis McCulloch, 1912 (Teleostei:
Atherinidae): Ecology, Genetics and Osmoregulation**

by

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Abstract

Population fragmentation is a common symptom of the decline of species, including freshwater fishes. It occurs naturally, but has also proliferated in response to human interventions that increase the prevalence and intensity of isolating barriers and events. In regulated rivers, for example, fish are affected by the loss of connectivity between habitats that is associated with hydrological changes. The process has evolutionary consequences by limiting gene flow, reducing genetic diversity and rendering the isolates vulnerable to local environmental changes.

Comparative studies of related species may help to elucidate the causes and consequences of fragmentation. For example, they may identify habitat features that influence the spatial separation of congeneric species. An opportunity for such a study arises with small fishes (Atherinidae) in the intensively-regulated River Murray, south-eastern Australia. Whereas the unspecked hardyhead *Craterocephalus stercusmuscarum fulvus* is widespread and abundant, the Murray hardyhead *C. fluviatilis* has a patchy distribution and is listed as ‘endangered’ by the International Union for Conservation of Nature and ‘vulnerable’ under the Australian *Environment Protection and Biodiversity Conservation Act 1999*. These two species rarely cohabit, implying that they could be separated by particular habitat characteristics.

In the past, several species of *Craterocephalus*, including *C. fluviatilis* and the Darling River hardyhead *C. amniculus*, have been regarded as *C. eyesii sensu lato*. The taxonomic separation of *C. s. fulvus* has been confirmed, but some doubt remains about the relationship of *C. fluviatilis* and *C. amniculus*. This issue needs resolution to ensure that appropriate targets are set for conservation.

This study is a comparative investigation of the aforementioned species. It was designed (1) to identify the habitat characteristics that influence the distribution and abundance of *C. fluviatilis* and, given that salinity emerged as a key factor, (2) to explore the biological implications of salinity through a comparative study of osmoregulation in *C. fluviatilis* and *C. s. fulvus*, (3) to determine whether the osmoregulatory responses of population isolates of *C. fluviatilis* differ at varying salinities, and (4) to evaluate the genetic population structure of *C. fluviatilis*, confirm its taxonomic separation from *C. amniculus* and identify genetic ‘management units’ for conservation.

Field sampling showed that *C. fluviatilis* is confined mainly to saline waters (0.4-20‰), whereas *C. s. fulvus* is absent from salinities >7‰. Comparisons were made of osmoregulation in these two taxa over a salinity range of 0.03-85‰, with additional reference to the small-mouth hardyhead *Atherinosoma microstoma*, a related estuarine species that tolerates salinities >94‰. The three species all are euryhaline, although the osmoregulatory ability of *C. s. fulvus* falters above about 35‰ salinity. *C. fluviatilis* is a better osmoregulator than *A. microstoma* at salinities <1‰, but both species tolerate hypersaline conditions (85‰).

Osmoregulation was compared in *C. fluviatilis* from two isolated populations in different salinity regimes (Wyngate: 0.4-1.5‰, Disher Creek: c. 1.0-45‰) to determine whether they show related phenotypic differences. Fish from both populations remained healthy at salinities from 5-65‰. The Disher Creek population maintained a significantly lower blood osmotic concentration than the Wyngate population at salinities ≤1‰, suggesting that there is a physiological difference between them.

The genetic population structure of *C. fluviatilis* and its taxonomic distinction from *C. amniculus* were investigated using complementary allozyme and mtDNA markers. This confirmed that *C. fluviatilis* is genetically distinct from its sister taxon, *C. amniculus*. It also identified several genetically-defined ‘management units’ as a framework for future conservation. Further, it revealed that *C. fluviatilis* in habitats downstream of Lock 1 on the Murray (274 km from the river mouth) displays a genetic signature indicating introgression with *C. amniculus*. Clearly, these findings have implications for the conservation of *C. fluviatilis*. For example, isolates can be prioritised for protection, and re-introduction programs can be modified accordingly.

The findings may be applied to other freshwater fish, especially populations of closely-related species subject to salinisation or other stressors, and they may also contribute toward understanding of the factors and processes underlying rarity and fragmentation. It is clear that salinity can be a significant factor in population fragmentation, and that closely-related species with similar ranges may be segregated by differences in osmoregulatory ability.

Declaration

This thesis contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

I give consent to this copy of my thesis being made available for photocopying and loan, subject to the provisions of the *Copyright Act 1968*.

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