Hydrobiologia **210**: 145–160, 1991.
W. D. Williams (ed.), Salt Lakes and Salinity.

Salinity tolerances of four species of fish from the Murray-Darling River system

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Key words: salinity, Murray-Darling, fish, tolerances, Australia

Abstract

The salinity tolerances of four species of small fishes native to the Murray-Darling river system were measured. Slow acclimation LD_{50}s were 43.7 ± 1.7 g L^{-1} for *Craterocephalus stercusmuscarum* Gunther, 38.0 ± 1.1 g L^{-1} for *Hypseleotris klunzingeri* (Ogilby), 58.7 ± 0.9 g L^{-1} for *Retropinna semoni* (Weber), and 29.8 ± 0.7 g L^{-1} for *Melanotaenia splendida* (Castelnau). The salinity tolerance of *M. splendida* was also measured by direct transfer, providing an estimated LD_{50} (infinite exposure time) of 20.8 g L^{-1}, ~70% of the slow acclimation value. Results suggest that at least adults of the species studied are under no threat from present or foreseeable salinities in the Murray River.

Introduction

The fish fauna of the Murray-Darling river system is depauperate by world standards; only 25 species are present, well below the world average for other rivers of similar size (Walker, 1983). Many species, such as the flat-headed galaxias (*Galaxias rostratus* Klunzinger), are found almost exclusively in the basin, and some, such as the two-spined blackfish (*Gadopsis bispinosus* Sanger), are both endemic and endangered (Lloyd & Walker, 1986). Most native species have declined in range and abundance since 1960 (Cadwallader, 1978) and their continued survival is of concern. A potential threat to survival is posed by those human activities within the basin causing a gradual increase in river salinities (Engineering and Water Supply Department, 1978; Mackay, Hillman & Rolls, 1988). Since 1978, salinities in the lower South Australian Murray have averaged ~0.5 g L^{-1}, with a mean annual salt increment of ~0.007 g L^{-1} yr^{-1} (Mackay *et al.*, 1988).

The effects of increased river salinities on the native flora and fauna have not been fully ascertained. Salinity tolerance testing has previously been confined to commercially important species such as the callop (*Macquaria ambigua* Richardson) and yabbie (*Cherax destructor* Clark). Yet, smaller species of fishes and crustaceans are just as likely to be important to the river ecosystem.

This study aims to extend our knowledge of salinity tolerance to include four small fish species considered important in the riverine food web, namely:

(1) *Craterocephalus stercusmuscarum* Gunther (Atherinidae);

(2) *Hypseleotris klunzingeri* (Ogilby) (Eleotridae);

(3) *Retropinna semoni* Steindachner (Retropinnidae);

(4) *Melanotaenia splendida* (Castlenau) (Melanotaeniidae).

These fishes include the most common small species in the Murray river. In the main, this study addressed their tolerance to slow increases in salinity, but to gain some idea of the relationship between slow acclimation and direct transfer experiments, direct transfer testing was carried out on one species, *M. splendida*.

Methods

Specimens were collected from the Murray River, near Mannum (35° S, 139° E), in August and September, 1987. For all species, fish were captured in three distinct size classes. As the largest size class comprised less than 1% of captures, only the smallest two classes were used in experiments.

Fish were maintained for two weeks in 100 L aquaria at 20 °C before experimentation. During this time and experiments, fish were fed a variety of food including dried flake food, live and frozen brine shrimp, and live *Drosophila* (fruit flies) and *Tubifex* (worms). Feeding presented no problems, except for *C. stercusmuscarum*, which refused anything but small, live food.

Marine aquarium salts ('Instant Ocean') were used in all experiments. These provide a better approximation of Murray salinity than sodium chloride, which constitutes only about 60% of ions in the South Australian Murray (Mackay *et al.*, 1988).

Glass aquaria (60 × 45 × 30 cm) were used for experiments. Washed river sand (4 L) was added to each tank as a substrate. Tanks were initially filled with 50 L of distilled water mixed with marine aquarium salts at a concentration of 0.30 g L^{-1} and were fitted with Ehiem 2007 internal power filters, which recirculated water at about 200 L h^{-1}. They were maintained in a temperature controlled room at 20 °C, and lit with a 12/12 hour light/dark cycle.

Each species was tested separately, but without replication, using two tanks (one experimental, one control). Twenty five fish were randomly selected for each tank. Fish were allowed two days to acclimatize before experimentation began. Salinities were then increased in the experimental tanks by adding marine aquarium salts each 24 hours at a rate of 2 g L^{-1} day^{-1}. Mortality was measured 24 hours after salt addition and counted against current salinity. Whether fish were in obvious distress was also noted. The fish were then fed; whether they accepted food was recorded. Dead fish were removed, and water lost by evaporation replaced with distilled water.

Tanks used for direct transfer experiments were set up in the same way as those for slow acclimation experiments. Experimental salinities were 0.03, 10, 20, 22.5, 25 and 30 g L^{-1}. Five adult *M. splendida* were placed directly into each tank. Mortalities were measured after 6, 112, 24, 48 and 96 hours. After taking control mortalities into account, LD_{50} values were calculated for each exposure time, and LD_{50}s (infinite exposure time) estimated as described by Green (1965).

Results

Table 1 and Fig. 1 summarize results of the slow acclimation experiments.

Craterocephalus stercusmuscarum. The range of salinities at which mortalities occurred was 36–50 g L^{-1}. The dose-mortality curve (Fig. 1) produced an LD_{50} of 43.7 ± 1.7 g L^{-1} (Table 1). Individuals began to show signs of distress and stopped feeding at salinities > 30 g L^{-1}. At salinities > 44 g L^{-1}, most specimens swam in an uncoordinated fashion and were unable to maintain

Table 1. Results of slow acclimation experiments.

Species	LD_{50} (g L^{-1})	S.E.
C. stercusmuscarum	43.7	± 1.7
H. klunzingeri	38.0	± 1.1
R. semoni	58.7	± 0.9
M. splendida	29.8	± 0.7

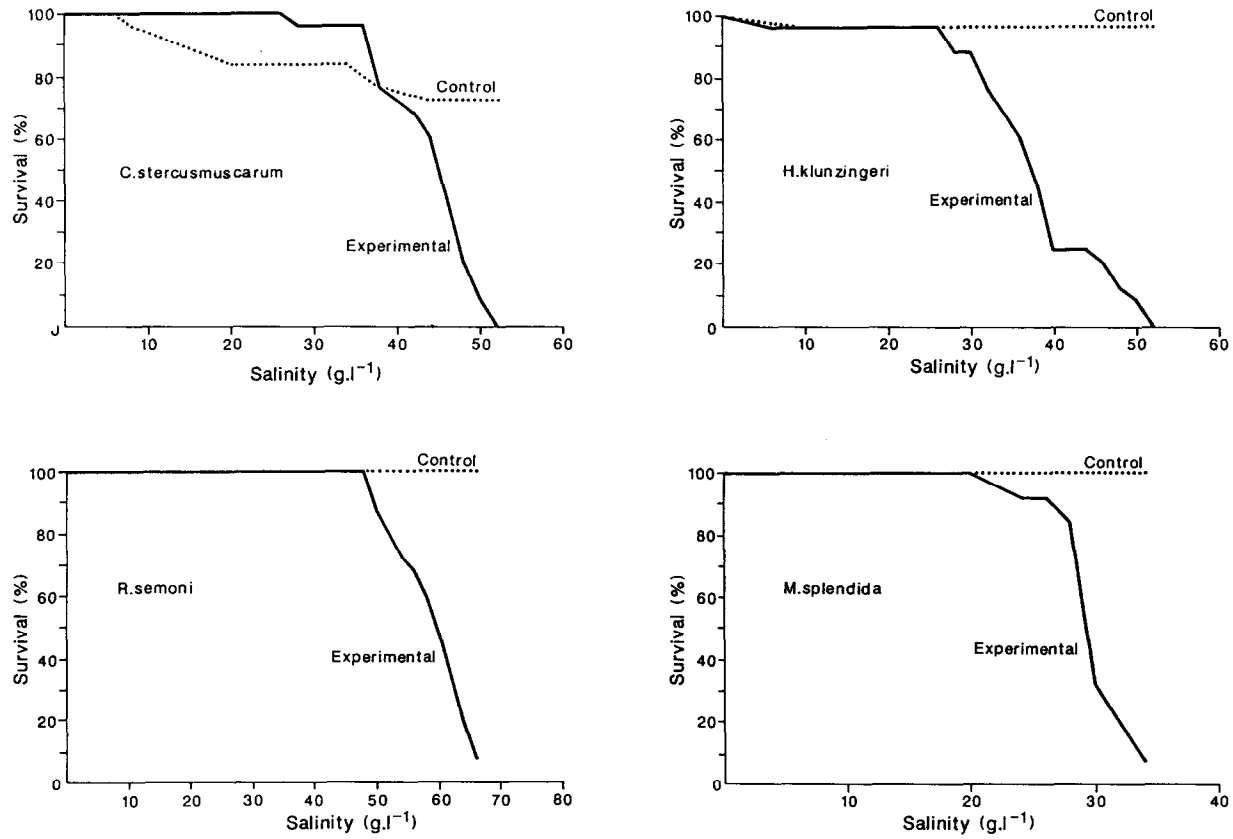

Fig. 1. Salinity vs survival in slow acclimation experiments.

balance properly. Fish in the control tank (0.30 g L^{-1}) seemed to suffer a higher initial mortality rate than those in the experimental tank (Fig. 2), but this was not statistically significant.

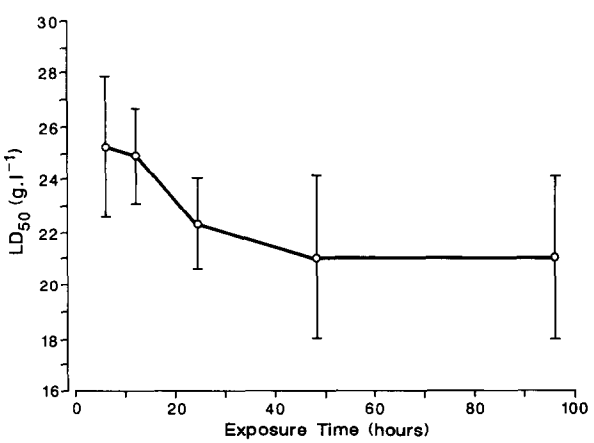

Fig. 2. M. splendida: LD_{50} (salinity) vs exposure time in direct transfer experiment. Vertical bars = standard error.

Nevertheless, overall health of fish in the experimental tank appeared noticeably better than of fish in the control tank when test salinities were between 5 and 25 g L^{-1}.

Hypseleotris klunzingeri. The range of salinities at which mortalities occurred in the experimental tanks was 26–50 g L^{-1}. The dose-mortality curve (Fig. 1) produced and LD_{50} of 38.0 ± 2.1 g L^{-1} (Table 1). Only one death occurred in the control tank. Fish began to show signs of distress and stopped feeding at salinities > 30 g L^{-1}. At salinities > 40 g L^{-1}, they seemed to have difficulty in coordination.

Retropinna semoni. Deaths occurred at salinities between 50 and 66 g L^{-1}. The dose mortality curve (Fig. 1) produced an LD_{50} of 58.7 ± 0.9 g L^{-1} (Table 1). No deaths occurred in the control tank. Specimens showed no signs of

distress until death; specimens continued to swim and feed normally up to 100% mortality.

Melanotaenia splendida. Deaths occurred at salinities between 22 and 36 g L^{-1}. The dose-mortality curve (Fig. 1) produced an LD_{50} of 29.8 ± 0.7 g L^{-1} (Table 1). No deaths occurred in the control tank. Specimens showed no obvious signs of distress during testing, although at salinities > 20 g L^{-1}, they were clearly less active than those in the control tank. Fish continued to feed throughout testing.

The direct transfer experiments on *M. splendida* produced LD_{50} values for 6-96 hours. Figure 2 plots LD_{50} versus exposure time, and Fig. 7 log LD_{50} versus exposure time. Transforming the data indicated in this figure to log LD_{50} and exposure time^{-1} provides a straight line relationship described by $y = 1.3235 + 0.54309$ ($r^2 = 0.816$). Here the y-intercept, viz. the LD_{50} at infinite exposure time, is 20.8 g L^{-1}, a value of 69.8% of the slow acclimation one (29.8 g L^{-1}).

As indicated, specimens of *M. splendida* (and *R. semoni*) seemed not adversely affected by high salinity until the point of death. To gain some idea of the ability of these fishes to withstand prolonged high salinity, no more salt was added in the slow acclimation experiments when only two specimens remained. In each case, the specimens continued to feed and behave normally until the experiment was terminated after one week. By contrast specimens of *H. klunzingeri* and *C. stercusmuscarum* at high salinities showed varied stress symptoms – such as resting on the substrate or at the surface, ceasing to feed, or swimming abnormally. For ethical reasons, these species were not tested for prolonged tolerance.

Discussion

The salinity tolerances of the species studied appear unusually high for freshwater fishes. Only known salt lake dwellers such as some *Cyprinodon* and *Fundulus* species (e.g. Martin, 1969; Teegavarapu, 1977) have been found to have tolerances as high as those of *C. stercusmuscarum* and *R. semoni*. We note that blood osmolarity in most marine and freshwater teleosts is equivalent to a salinity of 10–12 g L^{-1}; little variation is found worldwide (Parry 1966; Holliday, 1971). Fishes adapted solely to fresh waters (waters of salinities < 3 g L^{-1}) generally cannot regulate plasma ion levels when external osmolarity rises above blood osmolarity. Most cyprinids, for example, die at external salinities between 14 and 20 g L^{-1} (e.g. Geddes, 1979; Threader & Houston, 1983). Death is due to abnormal ion ratios of the body fluids, resulting in neuromuscular malfunction and dehydration (Holliday, 1971).

As the LD_{50}s of all species tested were over 29 g L^{-1}, it seems that they can survive in strongly hypertonic media. The implications are that these species or their immediate ancestors have been subjected to high salinities in inland waters in the relatively recent past, and/or that they have inherited salinity tolerance from their marine or estuarine ancestors.

With regard to the former suggestion, present conditions in the river are very different to those under which its fish fauna evolved. Construction of locks and weirs has apparently resulted in an increase in the *mean* salinity, but a decrease in the *range* of salinities found in the river proper. Since 1978, salinities of up to 21 g L^{-1} have been recorded in tributaries of the river which receive drainage from irrigation areas; however, salinity in the river itself now rarely exceeds 1.2 g L^{-1} (Mackay *et al.*, 1988). Before impoundment, the river was known to degrade during very dry seasons to a series of pools with salinities up to 6 g L^{-1} (Mackay *et al.*, 1988), and seawater was known to move some 100 km upstream. If such conditions have been recorded in the last 200 years, it seems probable that even greater extremes of salinity occurred during the evolution of the river's fish fauna.

With regard to the latter suggestion, only a handful of fishes is known to have a long evolutionary history in fresh water in Australia: only *Scleropages, Neoceratodus* and *Lepidogalaxias* species. Most species appear to have been derived from relatively recent marine ancestors and a few even have marine congeners

(McDowall, 1981). All species of native fish found in the Murray-Darling fall into the second category, and *a priori* therefore could be expected to retain at least moderate salinity tolerance.

Previous investigations of Murray-Darling macrofauna have also indicated high salinity tolerances. Thus, adult callop (*Macquaria ambigua*) have been found to tolerate seawater (~ 36 g L^{-1}) (Langdon 1987), while the yabbie (*Cherax destructor*) has an LD_{50} of 29.9 g L^{-1} (Mills & Geddes, 1980). The fact that both vertebrate and invertebrate groups display such tolerance supports the notion that the river fauna evolved in conditions of at least intermittently high salinity. Moreover, some of the species investigated, as well as close relatives, are known to occur in saline waters elsewhere. For example, specimens of *C. stercusmuscarum* and *H. klunzingeri* have been recorded from Victorian inland waters of up to 8.8 g L^{-1} salinity (Chessman & Williams, 1974), whilst *Craterocephalus eyrsii* (Steindachner) has been collected from waters of 110 g L^{-1} in Central Australia (Glover, 1982). Populations of *R. semoni* have been observed in estuaries and individuals have been captured in salt lakes (Merrick & Schmida, 1984).

M. splendida has not been reported from saline waters, and can tolerate salinities only as high as 17.8 g L^{-1} (Beumer, 1979). There are, in fact, few recorded observations of *Melanotaenia* species in saline waters. *M. splendida* can therefore be considered as the species most intolerant to salinity. Even so, current South Australian river salinities (~ 0.3–1.2 g L^{-1}) are far below the range of salinities at which mortalities occurred in our experiments (22–36 g L^{-1}); adult *M. splendida* can scarcely be seen to be under threat at current levels of river salinity.

In conclusion, this study suggests that present salinities in the South Australian Murray are at the lower parts of the tolerance ranges for adults of the species tested. Only in one tributary of the Murray, Barr Creek, where salinities of up to 21 g L^{-1} have been measured, could salinity pose a threat.

Extreme caution should be used in extrapolating laboratory tests on adult fish to the entire life cycle of the species. For example, it is possible that the fry of *R. semoni* have a salinity tolerance well below adult tolerance, as the Retropinnidae are from an anadromous lineage. Thus, the effects of salinity on eggs and fry of these species are still uncertain, although a preliminary study on the eggs and fry of *M. splendida* suggests that their 24 hour LD_{50}s are over 10 g L^{-1} (Williams, 1987). To ascertain fully the effect of increasing Murray salinities on flora and fauna, careful monitoring of natural populations and ecosystem studies are necessary, as well as laboratory investigations of the sort described here, but with greater replication.

Acknowledgements

This research formed part of a thesis for the degree of B. Sc. (Hons) by the senior author (M.D.W.) under the supervision of W.D.W. We sincerely thank Mr Brian Pierce and Mr Lance Lloyd for help and advice in the capture and maintenance of fish, and Dr Keith Walker for statistical advice and the supply of computer programs. W. D. W. acknowledges the support of the Australian Water Research Advisory Council during the period when this paper was written.

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