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Effects of manganese on juvenile mulloway (Argyrosomus japonicus) cultured in water with varying salinity – implications for inland mariculture.

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Abstract

The effects of 5 mg/L of dissolved manganese on juvenile mulloway at salinities of 5, 15 and 45 ppt were determined by comparing their survival, growth and blood plasma and organ chemistry with those of fish grown at the same salinities without manganese addition. Survival of mulloway at 45 ppt in the presence of 5 mg/L of manganese (73 ± 13%) was significantly lower than all other treatments, which achieved 100% survival. Those fish grown in water without manganese exhibited rapid growth, which was not affected by salinity (SGR = 4.05 ± 0.29%/day). Those fish grown at 5 ppt and 45 ppt in the presence of manganese lost weight over the two week trial (SGR -0.17 ± 0.42 and -0.44 ± 0.83%/day, respectively), whilst those at 15 ppt gained some weight (SGR 1.70 ± 0.20%/day). Manganese accumulated in the gills, liver and muscle of the fish and significant differences in blood plasma chemistry were observed. Blood plasma sodium and chloride of fish exposed to manganese were significantly elevated in hyperosmotic salinity (45ppt) and depressed at hyposmotic salinity (5 ppt) compared with unexposed fish at the same salinity; consistent with manganese causing apoptosis or necrosis to chloride cells. We did not, however, observe any gill epithelial damage under light microscopy. Blood plasma potassium was significantly elevated at all salinities in the presence of manganese and liver potassium and glycogen reduced. These findings are consistent with manganese interfering with carbohydrate metabolism.

Keywords: manganese, inland saline aquaculture, mulloway Argyrosomus japonicus, carbohydrate metabolism.
1. Introduction

Salt is a natural part of the Australian landscape, having been deposited by rainfall, wind and dust over thousands of years (Zalizniak et al., 2006). In many arid and semi-arid areas of southern Australia, human activities including land-clearing and irrigation have led to rising groundwater tables, dissolving stored salts from the soil and bringing them to the surface (Ghassemi et al., 1995; Lambers, 2003). The impacts of such secondary salinisation are many and include the loss of farm production and revenue, the degradation of rural infrastructure and recreational facilities, the salinisation of rivers and associated impacts on irrigation and drinking water (Williams, 2001).

As the salt in the Australian landscape is of marine origin, the culture of marine organisms has considerable potential as an adaptive use for saline groundwater (Doupé et al., 2003). Despite its marine origin, however, important deviations in ionic composition occur in saline groundwater compared to seawater (Partridge et al., 2008). These deviations depend primarily on the nature of the groundwater systems and the regolith with which they are associated (Mazor and George, 1992). One common deviation in Australian saline groundwater is an elevated concentration of manganese relative to seawater. A recent survey of 315 saline water sources in the wheatbelt of Western Australia, for example, found dissolved manganese concentrations as high as 83 mg/L, with an average value of 3.1 mg/L (George, unpublished data). Sources of dissolved manganese include anaerobic reduction of manganese oxides or the pedogenic weathering of manganese-bearing rock (Howe et al., 2004; Hardie et al., 2007).
Manganese is an essential nutrient for all organisms, including fishes (Underwood, 1977; Knox et al., 1981; Gatlin and Wilson, 1984; Maage et al., 2000) and plays critical physiological roles as a constituent of several metalloenzymes and as a coactivator of many other enzymes (Underwood, 1977; Cossarini-Dunier et al., 1988). Excess concentrations of manganese, however, can have negative effects including the disruption of sodium balance (Gonzalez et al., 1990), the impairment of calcium uptake and mineralisation (Reader et al., 1988), impacts on carbohydrate metabolism (Nath and Kumar, 1987; Barnhoorn et al., 1999), effects on immune response (Cossarini-Dunier et al., 1988; Hernroth et al., 2004) and neurotoxicity (Newland, 1999; Gunter et al., 2006). Of those studies on fishes, only freshwater fishes have been investigated, presumably due to the susceptibility of freshwater sources to anthropogenic sources of manganese pollution. Seawater contains a very low concentration of manganese (0.002 mg/L) (Spotte, 1992) and there appears to be no documented studies on the effect of elevated manganese on marine or estuarine fishes. Unpublished data from our laboratory collected during bioassays investigating the aquaculture potential of saline groundwater sources suggested that an elevated concentration of manganese may have a chronic effect on the growth of mulloway (Argyrosomus japonicus).

The aim of this study was therefore to investigate the effects of an elevated dissolved manganese (II) concentration (5 mg/L) on the survival, growth, pathology, and blood plasma and organ chemistry of mulloway. Mulloway is a temperate species considered a suitable candidate for inland saline aquaculture based on its euryhalinity, well-documented hatchery production techniques, reasonably rapid growth rates and a
temperate habitat compatible with the climatic zones where most secondary salinity in Australia occurs (Partridge et al., 2008). Because of the wide range of groundwater salinities found throughout Australia, the effect of elevated manganese in this trial was investigated across the range of salinities that mulloway are known to tolerate, namely 45, 15 and 5 ppt.

2. Materials and Methods

At each of the three salinities investigated (45, 15 and 5 ppt), the effects of an elevated manganese concentration (5 mg/L) were compared against a control (0 mg/L) in triplicate, flow-through 180 L tanks at 23°C over a period of two weeks. Five juvenile mulloway (9.4 ± 0.2 g) were stocked into each tank with flowing seawater (33 ppt), and then acclimated to their respective salinity over a period of three days. Once the target salinities had been reached, fish were acclimated to the experimental conditions for a further five days, then anaesthetised and reweighed. Prior to transferring fish back to their respective tank, those tanks randomly assigned to a treatment of 5 mg Mn/L were supplemented with laboratory grade MnCl₂·6H₂O by dissolving the required quantity of salt into 1 litre of tank water. Water flowed through each tank at 8 L/hour from header tanks containing the treatment water. Header tanks were emptied and cleaned daily and new treatment water prepared by dissolving the required quantity of MnCl₂·6H₂O into 1 L of water of the appropriate salinity before mixing this into the header tank. Water of 45 ppt was prepared by the addition of artificial seasalt (Ocean Nature®; Aquasonic Pty Ltd, Wauchope, Australia) to seawater, whilst 5 and 15 ppt water were prepared by diluting seawater with dechlorinated tap water. Seawater (33 ppt) was sourced from a marine bore at 18
This water is filtered to 1 µm and is free of particulate and dissolved organic carbon. Fish were fed daily to satiety and the amount of food consumed was recorded. Water quality parameters including temperature, dissolved oxygen and pH were measured daily. Dissolved manganese was measured four times throughout the trial in all tanks using inductively coupled plasma atomic emission spectroscopy (Vista AX CCD Simultaneous ICP-AES; Varian, Mulgrave, Australia) after filtering water samples to 0.45 µm (Whatman GFC).

At the completion of the two week trial, all fish were anaesthetised and weighed. Blood was immediately taken from the caudal vessels of three fish per replicate and pooled for the determination of plasma sodium, chloride and potassium using an ion-specific electrode analyser (Vetlyte, IDEXX, USA). Samples of dorsal muscle, liver and gills were taken for analysis of their ionic composition. These samples were freeze-dried, weighed to determine water content, then ground and digested in a combination of hydrogen peroxide, concentrated nitric acid and hydrochloric acid at 120°C. Digests were subsequently analysed for manganese, sodium, potassium, calcium, magnesium and iron via ICP-AES.

Samples of gill and liver were preserved in 10% formalin prepared in seawater, before embedding in paraffin then cutting 5 µm sections. Gill and liver sections were stained with haematoxylin and eosin (H&E) and liver sections with the periodic acid-Schiff (PAS) method for glycogen.

2.1. Statistical Analysis
Growth was expressed as specific growth rate (SGR) using the following equation:

\[
SGR\%/day = \left( \frac{\ln(W_f) - \ln(W_i)}{\text{Time (days)}} \right) \times 100
\]

Where \(W_f\) and \(W_i\) were the final and initial wet weights of the fish, averaged over all fish in a replicate. Feed intake was calculated as a percentage of the fish’s initial body weight.

The effects of dissolved manganese concentration and salinity on survival, specific growth rate, feed intake, and blood plasma and organ chemistry were compared using two-way ANOVA followed by a post hoc comparison of group LSD (Least Square Difference) means using Tukey’s HSD (Honestly Significant Differences) test. Statements of statistical significance refer to the 0.05 level. Arcsine transformations were performed where necessary to normalise data prior to ANOVA.

3. Results

3.1. Water Quality

Actual dissolved manganese concentrations were slightly less than the target value of 5 mg/L (Table 1). Despite concentrations between treatments differing by no more than 0.15 mg/L, the differences were found to be significant (\(P < 0.01\)), with the concentration at 15 ppt (4.13 ± 0.01 mg/L) being significantly lower than at 5 ppt (4.28 ± 0.01 mg/L) and 45 ppt (4.24 ± 0.01 mg/L). Manganese concentration in the control treatments were below the detectable limit of 0.01 mg/L.
Water pH (Table 1) was significantly affected by salinity ($P < 0.001$), but not manganese concentration ($P = 0.24$), or the interaction of manganese and salinity ($P = 0.23$). Water pH was significantly higher at 45 ppt (8.13 ± 0.01) than at 15 ppt (8.01 ± 0.01) and 5 ppt (8.03 ± 0.01).

3.2. Survival, Growth, Food Intake and Conversion Ratio

Survival of mulloway over the two week experimental period was 100% in all treatments except 45ppt-5mg Mn/L, with a survival of 73 ± 13%; significantly lower than all other treatments (Table 1).

At all salinities, those fish exposed to manganese grew at a significantly slower rate than those without manganese (Table 1). Two way analysis of variance revealed that salinity did not affect SGR ($P = 0.09$) but both manganese concentration ($P < 0.0001$) and the interaction of manganese concentration and salinity did ($P = 0.03$). Those mulloway grown in the presence of manganese at salinities of 5 and 45 ppt lost weight over the 2 week period (SGR -0.17 ± 0.42 and -0.44 ± 0.83%/day, respectively), whereas those at 15 ppt gained some weight (1.70 ± 0.20%/day). Those mulloway grown in the control treatments grew rapidly with an average SGR of 4.05 ± 0.29%/day (pooled across three salinities).

Food intake was affected by both salinity ($P = 0.02$) and manganese concentration ($P < 0.0001$), but not their interaction ($P = 0.63$) (Table 1). The post hoc comparison of group LSD means showed that those fish at 45 ppt ate significantly less food than
those at 15 ppt and those fish exposed to 5 mg/L of manganese at all salinities ate
significantly less food than those in the control treatments.

Due to mulloway’s loss of weight at 5 and 45 ppt in the presence of manganese, we
were unable to calculate food conversion ratios for these fish. Fish grown in the
presence of manganese at 15 ppt had a significantly higher (poorer) FCR (1.55 ± 0.22)
than those cultured at the same salinity without manganese addition (0.81 ± 0.06).

3.3. Histology

No epithelial damage to the gills was observed in histological sections. PAS-positive
cells in the liver were clearly more abundant in manganese exposed fish at all
salinities, however, quantification could not be conducted due to a lack of uniform
distribution within sections.

3.4. Blood plasma ionic composition

Concentrations of the electrolytes in the blood plasma of mulloway are shown in
Figure 1. Blood plasma potassium and chloride were significantly affected by
manganese concentration (P<0.0001 and 0.0073 for K and Cl, respectively), salinity
(P = 0.04 and <0.0001, respectively) and the interaction of these terms (P = 0.006 and
<0.0001, respectively). Blood plasma sodium concentration was not affected by
manganese concentration (P = 0.27) but was affected by salinity (P = 0.0005) and the
interaction of manganese concentration and salinity (P = 0.002).
Because of the significant interaction terms, we conducted one-way analysis of variance, comparing blood plasma ionic concentrations between fish exposed and not exposed to manganese at each salinity. Blood plasma sodium was significantly greater in fish exposed to manganese at 45 ppt ($P = 0.002$) but significantly lower in those exposed at 5 ppt ($P = 0.04$). There was no difference in blood plasma sodium concentrations between exposed and unexposed fish at 15 ppt ($P = 0.11$). The same pattern can be seen with blood plasma chloride at the various salinities and manganese concentrations, i.e. a significantly greater chloride concentration in manganese exposed fish at 45 ppt compared with unexposed fish ($P = 0.006$), with the opposite effect at 5 ppt ($P = 0.016$) and no significant difference at 15 ppt ($P = 0.51$). Blood plasma potassium was significantly higher in exposed fish compared with unexposed fish at all salinities ($P < 0.02$).

In order to test the hypothesis that mulloway were osmoregulating well in the absence of manganese across the range of salinities tested, one-way analyses of variance were also conducted on the electrolyte data from unexposed fish. These ANOVAs revealed that salinity had no significant effect on blood plasma sodium ($P = 0.97$), chloride ($P = 0.13$) or potassium ($P = 0.69$).

### 3.5. Organ Chemistry

Exposure to manganese resulted in significantly higher concentrations of this metal in the gills, muscle and liver of the fish (Table 2). The manganese content of the gills and muscle were both affected by water manganese concentration ($P < 0.0001$ for both organs), salinity ($P = 0.001$ and 0.01, respectively) and the interaction ($P = 0.0008$ and...
0.015, respectively). Despite fish exposed to manganese at 45 ppt showing the poorest performance in terms of survival and growth, gill and muscle manganese concentrations at this salinity were lower than at the other salinities; significantly more so than at 5 ppt. Liver manganese concentration was not significantly affected by salinity (P = 0.65) or the interaction of salinity and manganese concentration (P = 0.70), but was significantly affected by water manganese concentration (P < 0.0001).

There were no clear trends in other organ chemistry parameters with salinity or manganese concentration, with the exception of the potassium content of liver, which was significantly lower in those fish reared in the presence of manganese (P < 0.0001) (Table 2). Salinity (P = 0.30) or the interaction of manganese concentration and salinity (P = 0.41) did not affect liver potassium.

4. Discussion

Dissolved manganese had a significant, detrimental effect on the growth of juvenile mulloway at all salinities tested, with this effect being greater at salinities of 5 and 45 ppt. Dissolved manganese only affected survival at the highest salinity.

Our finding of equivalent survival, growth and blood plasma electrolyte concentrations across the control salinity treatments demonstrates that mulloway are capable of efficient osmoregulation across this salinity range. Mulloway’s ability to tolerate 5 ppt has been described previously (Fielder and Bardsley, 1999), however this appears to be the first published data on their performance at a salinity greater than seawater.
The poor growth performance of fish exposed to manganese was concomitant with a significantly lower food intake. Intakes in the control treatments were similar to that recommended for mulloway of similar size by Collett et al. (2008). Food intake of manganese-exposed fish was approximately half this rate. Similar inhibitory effects of excess manganese on appetite have been described for pigs, cows and sheep (Cunningham et al., 1966; Underwood, 1977). The significantly higher (poorer) FCR in fish exposed to manganese at 15 ppt, compared with unexposed fish at this same salinity, indicates that the reduced growth of manganese exposed fish was contributed to by poor feed utilisation efficiency as well as reduced food intake.

The mode of action of manganese toxicity to fishes has not been well defined. Nyberg et al. (1995) suggested that its toxic action is probably similar to other metals whereby it is deposited onto the gills, causing physiological disruption to iono- and or osmo-regulatory functions. Manganese has been shown to deposit on the gills of lobsters (Baden et al., 1990), however, we found no evidence of precipitation of manganese on the gill surface or damage to the gill epithelium. Although chemical analysis of gills in the current study revealed a high concentration of manganese, we are unable to determine whether this was precipitated on the gill surface, within the gills filaments or associated with the cartilaginous gill arches, which have been proposed as a long term fixation site for manganese (Adam et al., 1997).

Rouleau et al. (1995) suggested that uptake of manganese probably occurs mainly via the gills and is then transported by the blood to various organs and tissues, where it accumulates most in those tissues rich in mitochondria such as liver, pancreas and
kidney. In their study, Rouleau et al. (1995) found that manganese accumulation in freshwater brown trout was greatest in the liver, then gills, then muscle. In our study, manganese concentration was greatest in the gills, followed by the liver then muscle. Our findings are consistent with Capelli et al. (1987) who found higher concentrations of (naturally occurring) manganese in the gills of marine Atlantic bonito, *Sarda sarda*, compared with muscle and liver. These differences in the major target organs for manganese accumulation suggest that different uptake mechanisms occur between freshwater and marine species. These differences may be caused by differences in metal speciation between freshwater and seawater as described in further detail below.

It is well documented that manganese uptake and toxicity in freshwater fishes increases with decreasing pH (Bendell-Young and Harvey, 1986; Rouleau et al., 1996) and with decreasing freshwater hardness (Reader et al., 1988; Gonzalez et al., 1990; Adam et al., 1997; Stubblefield et al., 1997; Reimer, 1999). At low pH, particularly at pH values below 7, a greater proportion of manganese in freshwater exists as aqueous, toxic Mn(II), whereas high pH values favour the oxidation of insoluble and inert colloidal oxides (Florence et al., 1992; Rouleau et al., 1996; Zaw and Chiswell, 1999). Although there is a lack of data on the effects of pH on manganese speciation in seawater, our findings are consistent with this freshwater model, given that the pH of 45 ppt water in the current study was significantly higher than at 15 and 5 ppt, and that uptake of manganese at this salinity was significantly lower than at the lower salinities. However, although the difference in pH was significant, it amounted to less than 0.12 pH units and the pH at all salinities was well above the ‘critical’ range of 6 - 7 described by Rouleau et al. (1996). It seems unlikely, therefore, that differences in pH among salinity treatments had a major...
effect on manganese uptake. As we discuss in further detail below, we believe the far
greater differences in salinity between treatments is responsible for the observed
differences in toxicity.

In freshwater, decreases in metal toxicity with increasing hardness have been
attributed to the ability of calcium (and to a lesser extent magnesium) to compete with
toxic metals for ligand sites on the gill surface, which reduces the permeability of the
paracellular pathways to these metals (McDonald et al., 1989). In seawater, the effects
of increasing salinity on metal ion toxicity appear to be more complicated and
dependent on how the salinity affects metal ion speciation. The freshwater model
implies that the higher calcium and magnesium concentrations in seawater should
provide even greater protection than hard freshwater and that this protection should
increase with increasing salinity. Although supporting data are lacking for manganese,
the uptake by fishes of certain other metals, including copper and lead, is inversely
proportional to salinity (Somero et al., 1977; Jezierska and Witeska, 2006) and the
uptake of manganese by salt water mussels (*Mytilus edulis*) has been shown to follow
this trend (Struck et al., 1997). There are examples, however, where metal
bioavailability can increase with salinity. For example, when freshwater containing
relatively non-toxic aluminium colloids mixes with seawater, the calcium and
magnesium in seawater can displace the aluminium and thereby increase the
concentration of toxic, low molecular mass cationic aluminium species (Bjerknes et
al., 2003; Teien et al., 2006). Our data showing decreasing manganese concentration
in gills and muscle with increasing salinity are consistent with the former model,
however more research is required to determine the effect of salinity (and pH) on
manganese speciation.
Our data also suggest, however, that despite reduced uptake at 45 ppt, toxicity at this salinity was greater compared with lower salinities. We suggest that the greater toxicity of manganese at higher salinity, despite its reduced uptake, is due to the effects of manganese on osmoregulation.

Gonzalez et al. (1990) found that freshwater brook char, Salvelinus fontinalis, exposed to 600 mg/L of dissolved manganese experienced a 40% reduction in plasma sodium concentration. Studies have also shown that copper exposure induces hyponatremia in freshwater fishes caused by apoptosis and necrosis of chloride cells (Dang et al., 2000). Given that mulloway in hyposmotic salinities face the same osmotic challenges as freshwater fishes; our findings of hyponatremia in mulloway at 5 ppt are consistent with these studies and suggest that manganese may also induce apoptosis and necrosis of chloride cells. Mulloway held at hyperosmotic salinity (45 ppt) face the opposite osmotic challenges to those in hyposmotic salinity and their chloride cells are responsible for the elimination (rather than uptake) of monovalent ions (Evans et al., 2005). Our data showing elevated blood plasma sodium and chloride in manganese-exposed fish at 45 ppt is therefore consistent with manganese causing apoptosis and necrosis of chloride cells, as the loss of such cells would result in reduced elimination of these ions. Seemingly inconsistent with this theory of manganese causing increased apoptosis and necrosis of chloride cells is the fact that we did not observe any histological changes in H&E sections. Cells dying from apoptosis, however, remain intact, and Dang et al (2000) only observed these changes using immunocytochemistry and electron microscopy.
Given that 15 ppt is closer to iso-osmotic than the other two salinities, uptake and elimination of monovalent ions will be less at this salinity and the effects of chloride cell apoptosis not as detrimental as at the more extreme salinities tested. Indeed the differences in plasma sodium and chloride concentrations between exposed and unexposed fish were smaller and statistically non-significant at this salinity, compared with 5 and 45 ppt. We therefore suggest that the reduced osmotic demands at 15 ppt are the likely reason these fish tolerated manganese exposure better than at 5 and 45 ppt. We cannot rule out the possibility that the greater tolerance for manganese exposure at a salinity of 15 ppt was due to the small, but significantly lower manganese concentration at this experimental treatment, compared to the 5 ppt and 45 ppt treatments. There are, however, two reasons why we believe that this lower manganese concentration in the 15 ppt treatment was not biologically significant. First, there was a reduction of only 0.15 mg/L at this treatment, 3% of the target value of 5 mg/L. Second, there was no relationship between manganese concentration and fish survival and growth rate in the three replicates within treatments.

Even though the fish exposed to manganese at 15 ppt performed better than those at 5 and 45 ppt, they still exhibited significantly slower growth compared with control fish at the same salinity. Blood plasma potassium was significantly elevated in these fish, suggesting that there was some disruption to their osmoregulatory capacity. The reduced feed conversion efficiency seen in these fish, compared with unexposed fish at the same salinity, may therefore be the result of the repartitioning of a greater proportion of dietary energy into osmoregulation or, alternatively, the result of impaired carbohydrate metabolism described below.
We found significant reduction in liver potassium and reduced liver glycogen in fish exposed to manganese. Manganese has been implicated in impaired carbohydrate metabolism in both fishes (Nath and Kumar, 1987; Nath and Kumar, 1988; Barnhoorn et al., 1999) and mammals (Underwood, 1977) and observations of decreased liver glycogen and hyperglycemia in manganese exposed fishes have led to the hypothesis that manganese exposure may inhibit insulin release (Nath and Kumar, 1987) in a similar fashion to that seen in cadmium exposed rats (Ghafghazi and Mennear, 1973). Although the reduced liver glycogen that we observed in manganese exposed fish is consistent with these studies, it would also be the expected response of fish that are losing weight or growing at a sub-optimum rate. The significant reduction in liver potassium of manganese exposed fish, however, is consistent with a lack of insulin, as such a deficiency inhibits potassium absorption by liver cells in rats (Fehlmann and Freychet, 1981).

Cardeilhac et al. (1979) and Cardeilhac and Hall (1977) attributed the death of the marine fishes sheepshead (Archosargus probatocephalus) and pinfish (Lagodon rhomboides) exposed to excess copper to osmoregulatory failure and specifically to an elevated concentration of blood plasma potassium. We found significantly elevated blood plasma potassium in fish exposed to manganese at all salinities. The blood plasma potassium of manganese exposed mulloway at 45 ppt was within the toxic range reported for these two species and is therefore a possible cause of death of mulloway in this treatment.
5. Conclusion

Our results demonstrate that 5 mg/L of dissolved manganese, a concentration typical of many saline groundwater sources in Western Australia, has a significant, detrimental effect on the growth, health and survival of mulloway. It appears that selecting a water source with salinity close to iso-osmotic will minimise the effects of dissolved manganese, however, further studies are required to determine the minimum concentrations at which growth is not affected. Given that this study was conducted at relatively high pH values, the negative impacts of a similar dissolved manganese concentration can be expected to be far greater in aquaculture systems or for groundwater sources which have lower values of pH.

Given that both salinity and the pH values typical of groundwater result in very slow oxidation rates of manganese, its removal from saline groundwater is more complicated than iron; another metal often found in high concentrations in saline groundwater. Significant chemical pre-treatment with strong oxidising agents, or the temporary elevation of pH to above 9, would be required to oxidise manganese prior to its physical removal; a process that will add significant cost and complexity to the delivery of water, particularly for those production systems using large volumes. Although oxidation is slow, once it does occur, precipitation of manganese oxides onto tanks, pipes and pumps is likely to be problematic and impose a significant management issue if dissolved manganese is not oxidised and removed prior to use.
6. Acknowledgements

This study was funded by Challenger TAFE, the Australian Department of Fisheries, Forestry and Agriculture and the Department of Conservation and Land Management.

7. References


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Table 1: Water quality and performance parameters for juvenile mulloway grown in the presence or absence or manganese at various salinities for two weeks.

Table 2: Organ chemistry data for juvenile mulloway grown in the presence or absence or manganese at various salinities for two weeks.

Figure 1: Concentrations of (a) sodium, (b) chloride and (c) potassium in the blood plasma of juvenile mulloway grown in the presence or absence or manganese at various salinities for two weeks. Columns within salinity sharing the same letter are not significantly different (P>0.05).
<table>
<thead>
<tr>
<th>Salinity (ppt)</th>
<th>Target Mn (mg/L)</th>
<th>Actual Mn (mg/L)</th>
<th>pH</th>
<th>Survival (%)</th>
<th>SGR (%/day)</th>
<th>Food intake (%BW/day)</th>
<th>FCR</th>
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<td>Manganese</td>
<td>Muscle (mg/kg)</td>
<td>Liver (mg/kg)</td>
<td>Potassium Liver (mg/kg)</td>
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Figure(s)

Figure (a) shows plasma Na levels (mmol/L ± SE) for two Mn concentrations (0 mg Mn/L and 5 mg Mn/L) across different salinities (5 ppt, 15 ppt, and 45 ppt). Figures (b) and (c) display plasma Cl and K levels, respectively, under the same conditions.