BIOACCUMULATION MONITORING 1999 ANNUAL REPORT FOR FLETCHER CHALLENGE PAPER ALBURY

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1.0 INTRODUCTION

Fletcher Challenge Paper, Albury operates a 2100 ML irrigation storage dam (Lake Ettamogah) for tertiary treated wastewater. The Lake has been stocked with fish obtained from commercial hatcheries. The Murray-Darling Freshwater Research Centre was contracted to assess bioaccumulation of metals in these fish, as part of a long-term monitoring program.

2.0 AIMS

To undertake biological and chemical monitoring of FCP's wastewater discharged to the River Murray in accordance with proposals developed in consultation with EPA NSW and NSW Fisheries.

Environment Protection Authority New South Wales Licence No.001272; Section W11:
"A final summer survey of fish from the holding pond/winter storage dam shall be conducted to determine long term bioaccumulation of metals compared with fish of the same species from other local sources."

The null hypothesis tested "that fish living in wastewater are not different to those living in other local freshwater environments."

3.0 METHODS

Fish (golden perch (*Macquaria ambigua*) and silver perch (*Bidyanus bidyanus*)) from three different sources were used to assess bioaccumulation of metals in muscle tissue.

Ten three year old Silver Perch were purchased from Murray Cod Hatchery at Wagga Wagga in December 1998. Nine Golden Perch of mixed age were collected by anglers from the Murray River near Albury, downstream of Lake Hume between December 1998 and February 1999. Three Silver Perch and one Golden Perch were netted from FCP's Lake Ettamogah near Albury in December 1998.

The fish were anaesthetised and killed using benzocaine™. Total length, total weight and gonad weights were measured. The length and weight data were used to calculate two condition factors for each fish:

Index of Condition = Weight (g) x 10^5/Length^3 (mm)

Gonadosomal Coefficient = Gonad weight/(Total weight/gonad weight)

Ref: Clesceri *et al.* 1989

Pivot tables were used to then compare condition factors between source, species and sex.

Data from Lake Ettamogah fish collected in February 1998 (documented in the previous Annual Report) were also compared. These data are referred to as Ettamogah 1997 (summer of 1997) to differentiate them from the December 1998 samples.

Approximately 20g of muscle tissue (with skin and scales attached) were removed from behind the head of each fish. The sample was then frozen, freeze-dried, homogenised and then acid digested in preparation for assay of metals (Al, As, Cd, Cu, Fe, Mn, Pb, and Zn) by ICP at Australian Government Analytical Laboratories (AGAL). The resulting data, (expressed as mg/kg of muscle tissue), were then compared by source using scatter plots (mean ± standard deviation).

Otoliths were removed and sent to the Victorian Department of Natural Resources' Central Ageing Facility at the Marine and Freshwater Research Institute, Queenscliff, where age estimates were made without any prior knowledge of the date of capture or fish sizes. The ages were then adjusted using as birthdate the 1st January and the respective date of capture to group the fish into their correct cohorts.

4.0 RESULTS

Fish from Lake Ettamogah were observed to be robust and healthy at the time of capture (no skin lesions parasites or fin damage).

There was little difference overall in the mean (± standard deviation) physiological condition factors (Index of condition (IOC) and Gonadosomal Coefficient (GC)) between fish from each of the sources (Table 1). The mean IOC of the Hatchery fish was slightly lower than that of the fish from the other three sources, possibly due to their containment. Female fish from all sources demonstrated greater IOC than males, a reflection of sampling during the spawning season when the ovaries of females are ripe with eggs and may comprise up to one quarter of their body weight. The mean GC for female fish were greater than that of male fish except for the single Hatchery female, which although the same age and size as the males was sexually immature.

The mean concentrations of Al, As, Cd, Cu, Fe, Pb, Mn and Zn in muscle tissue are depicted in Figure 1. Iron, zinc and aluminium were the most abundant. Arsenic, cadmium and lead the least abundant. Overall, the metal concentrations were no different between the four sources, generally falling within similar ranges, except Manganese, Copper and Iron which were slightly elevated in the 1998 Lake Ettamogah fish compared with the controls. However, these were still lower than those recorded in the previous year (King 1998).

Metals in the fish from the hatchery were the least variable and those from Lake Ettamogah in the summer of 1997 were the most variable. Between the two Lake Ettamogah sampling periods (summer 97 and summer 98) the concentrations of As, Cu and Zn were significantly

different (t = 2.18, t = 2.35, t = 2.9 respectively where t crit = 2.17). Although the older fish contained more As and Zn, but less Cu compared with the younger fish, the concentrations of the former were still similar or less than the concentrations from the hatchery and river fish,. Overall, the concentrations of metals in the older fish tended to approach those of the "control" fish from the River and Hatchery.

Ageing data including images of the sectioned otoliths are included in Appendix 1. Silver Perch from Lake Ettamogah were three y/o old females. Golden Perch from the River were between three and fifteen years old (males 3 - 4 y/o and females 3 - 15 y/o). The one male Golden Perch from Lake Ettamogah was one year old. In Table 2, the physical measures are compared for three y/o old fish from each source. The mean index of condition was greatest for the Lake Ettamogah Silver Perch (females) and least for the Wagga Hatchery Silver Perch (primarily males), whilst their liver body mass ratio's were the same. A comparison of the gonadosomal coefficient is of little value due to the disparity of sex and species for the different sources. Of the ten Hatchery Silver Perch only one was female, whereas all three of the Lake Ettamogah Silver Perch were female. Three out of nine fish from the River were females but they were all Golden Perch.

5.0 DISCUSSION

The Lake Ettamogah and River fish were healthy and in spawning condition. The Hatchery fish were much smaller than Lake Ettamogah fish at the same age and the sole female was sexually immature. Fishing in Lake Ettamogah was abandoned after only a small number of fish were captured due to concern over capture of non target species.

Metals in the Stocked Fish (Lake Ettamogah) fell within an acceptable range compared with the two "controls" (Hatchery and River). Of the metals assayed the National Health and Medical Research Council specifies maximum residue limits in edible fish tissue for the heavy metals cadmium (0.2 mg/kg) and lead (1.50 mg/kg) (NHMRC 1988). The Lake Ettamogah fish assayed lower than the controls for both cadmium and lead, and all were below the NHMRC limits.

The greater index of condition for the Lake Ettamogah fish may be realistic and it would have been useful to compare the Lake Ettamogah data with the Murray River data in further detail. However, the comparison of small differences in physiological measures for the three y/o fish may be confounded by differences attributable to species and sex. Only Golden Perch were captured from the River. Despite an almost even proportion of males to females, the females tended to be older than the males and only one female could be included in the three y/o comparison. Further more, all of the three y/o fish caught in Lake Ettamogah were female Silver Perch and only one of the Hatchery Silver Perch was female.

Sexual maturity for both species is size (length) related (Golden Perch - 325 mm and 397 mm, and Silver Perch - 215 mm and 312 mm for males and females respectively) (Mallen – Cooper *et al.* 1995). These differences are also reflected in the approximate minimum age for mature fish reported by the same authors: Silver Perch - males 3 years and females 5 years; Golden Perch- males - 2 years and females - 4 years.). The immaturity of the female Silver Perch from the hatchery can be attributed to her small size. Gonads of mature females comprise a greater proportion of their body mass compared with males during spawning season and contribute significantly to their greater overall size. Although meaningful comparisons may be limited by differences of species and sex for the three sources, sexual maturity and size of the Silver Perch living in FCP's Lake Ettamogah compares favourably with similar data from the literature for wild stock.

There was no evidence of harm or significant bioaccumulation of the metals assayed in fish growing in FCP's wastewater at Lake Ettamogah.

6.0 COMMUNICATION

A research paper on the role of bacteria in manganese bioaccumulation was published in an international journal (King *et al.* 1999).

7.0 REFERENCES

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8.0 FIGURES

9.0 TABLES

Table1: Pivot Table Comparing the Physiological Condition Factors of Fish of different Sex and Species from Different Freshwater Sources

Index of Condition

	Female						**Male**						**Overall**	
	Golden Perch		*Silver Perch*		**Female Overall**		*Golden Perch*		*Silver Perch*		**Male Overall**		**for each Source**	
Source	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev
Ettamogah 97	1.71	-	2.43	0.36	2.31	0.43	1.69	-	1.81	0.09	1.78	0.09	2.1	0.43
Ettamogah 98	-	-	2.17	0.22	2.17	0.22	1.64	-	-	-	1.64	-	2.04	0.32
Hatchery	-	-	1.4	-	1.4	-	-	-	1.32	0.21	1.32	0.21	1.33	0.2
River	2.12	0.33	-	-	2.12	0.33	1.76	0.13	-	-	1.76	0.13	1.92	0.29
Overall for each sex & species	2.04	0.34	2.23	0.43	2.16	0.4	1.73	0.12	1.44	0.29	1.55	0.28	1.81	0.45

Gonodosomal Coefficient

	Female						**Male**						**Overall**	
	Golden Perch		*Silver Perch*		**Female Overall**		*Golden Perch*		*Silver Perch*		**Male Overall**		**for each Source**	
Source	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev	Mean	St Dev
Ettamogah 97	0.010	-	0.058	0.028	0.050	0.032	0.010	-	0.020	0.000	0.018	0.005	0.037	0.029
Ettamogah 98	-	-	0.086	0.044	0.086	0.044	0.002	-	-	-	0.002	-	0.065	0.055
Hatchery	-	-	0.007	-	0.007	-	-	-	0.053	0.029	0.053	0.029	0.048	0.031
River	0.103	0.085	-	-	0.103	0.085	0.007	0.005	-	-	0.007	0.005	0.05	0.072
Overall for each sex & species	0.084	0.084	0.062	0.038	0.070	0.057	0.007	0.005	0.045	0.029	0.031	0.030	0.047	0.047

Table 2: Physiological Measures for Three Year Old Fish from Wagga Hatchery (WH), Murray River (MR) and Lake Ettamogah (LE)

Sample Id	Perch Species	Sex	Age (years)	Total Length (mm)	Total Weight (g)	Gonad Weight (g)	Liver Weight (g)	IOC *	GS **	Liver:Body Mass Ratio
WH98-1	Silver	M	3	270	300.0	11.42	1.91	1.524	0.040	0.006
WH98-2	Silver	M	3	271	300.0	13.69	2.23	1.507	0.048	0.007
WH98-3	Silver	M	3	292	300.0	29.04	2.58	1.205	0.107	0.009
WH98-4	Silver	M	3	278	200.0	10.05	3.14	0.931	0.053	0.016
WH98-5	Silver	M	3	300	400.0	4.22	5.83	1.481	0.011	0.015
WH98-6	Silver	M	3	272	300.0	8.85	3.34	1.491	0.030	0.011
WH98-7	Silver	F	3	278	300.0	1.99	3.99	1.396	0.007	0.013
WH98-8	Silver	M	3	295	350.0	12.74	2.64	1.363	0.038	0.008
WH98-9	Silver	M	3	310	350.0	21.18	4.08	1.175	0.064	0.012
WH98-10	Silver	M	3	294	300.0	23.51	3.22	1.181	0.085	0.011
	mean			**286.00**	**310.00**	**13.67**	**3.30**	**1.325**	**0.048**	**0.011**
	std dev			**13.98**	**51.64**	**8.55**	**1.13**	**0.196**	**0.031**	**0.003**
MR98-1	Golden	M	3	385	900.0	11.54	15.07	1.577	0.013	0.017
MR98-2	Golden	M	3	380	1000.0	12.05	15.52	1.822	0.012	0.016
MR98-4	Golden	M	3	350	800.0	3.00	16.07	1.866	0.004	0.020
MR98-6	Golden	F	3	450	1700.0	4.46	31.96	1.866	0.003	0.019
	mean			**391.25**	**1100.00**	**7.76**	**19.66**	**1.783**	**0.008**	**0.018**
	std dev			**42.11**	**408.25**	**4.70**	**8.21**	**0.139**	**0.005**	**0.002**
LE98-1	Silver	F	3	370	1000.0	106.90	12.16	1.974	0.120	0.012
LE98-2	Silver	F	3	470	2500.0	233.83	30.60	2.408	0.103	0.012
LE98-4	Silver	F	3	470	2200.0	79.03	15.16	2.119	0.037	0.007
	mean			**436.67**	**1900.00**	**139.92**	**19.31**	**2.167**	**0.087**	**0.010**
	std dev			**57.74**	**793.73**	**82.51**	**9.89**	**0.221**	**0.044**	**0.003**

***Index of Condition: K = W*10^5/L^3**

****Gonadosomal Coefficient = Gwt/(Twt-Gwt)**

10.0 APPENDIX 1:

Ageing Data for Golden and Silver Perch
from
Fletcher Challenge Paper's Lake Ettamogah (Ref. prefix: LE)
Murray River Albury (Ref. prefix : MR)

Conducted by the Central Ageing Facility
Marine and Freshwater Research Institute
Queenscliff
Victoria

For
The Murray Darling Freshwater Research Centre