

Aquatic Fauna - Biological Survey
Ten Mile Brook Dam Margaret River

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Correct citation:

Lawrence¹, C., Hugh¹, C., Larsen¹, R., Ledger², J. and Vercoe², P. (2010). Aquatic Fauna - Biological Survey. Ten Mile Brook Dam Margaret River. Fisheries Research Contract Report No. 22. Department of Fisheries, Western Australia. 16p.

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1.0 Background

Ten Mile Brook Dam has a surface area of 275 Ha and receives water from a 10 km² catchment (Figure 1). It has a total storage capacity of 1,691 ML. At the time of this study the dam was 89% capacity (1,511 ML), however as recently as June 2009 water storage was as low as 663 ML (39% capacity).



Figure 1. Ten Mile Brook Dam (Figure courtesy of Water Corporation).

In order to improve the drinking water supply to the town of Margaret River, the Water Corporation is investigating the option of pumping bore water into Ten Mile Brook Dam. This would remove the need to transfer water from the Margaret River into the dam.

However, the critically endangered Margaret River marron is endemic to this region. This endangered species can only be distinguished from the more common “smooth” marron by DNA fingerprinting.

In addition, all the endemic freshwater crayfish in this region are restricted to this global biodiversity hotspot. The region also has the highest proportion of endemic freshwater fishes of all the major Australian Drainage Divisions, with 80% of the freshwater fishes from the southwest of Western Australia are found nowhere else. Many of these native fish populations are fragmented and some species are listed as vulnerable to extinction.

2.0 Need

The proposed pumping of bore water into Ten Mile Brook Dam may have a negative, positive or negligible impact upon aquatic biodiversity.

A biological survey is required to:

- 1) Determine the presence or absence of endemic fish and crayfish species in Ten Mile Brook Dam prior to the pumping of bore water.
- 2) Determine if there are issues or species of concern in Ten Mile Brook Dam, particularly those listed as critically endangered or vulnerable, prior to pumping bore water.
- 3) Determine if the marron species in Ten Mile Brook Dam are either the critically endangered “hairy” marron (*C. tenuimanus*), the commonly found and widespread “smooth” marron (*C. cainii*) or hybrids between the two species.
- 4) Determine aquatic macroinvertebrate composition in Ten Mile Brook Dam and calculate the biological condition index of the water body prior to the pumping of bore water.
- 5) Provide a baseline biological survey from which the impact (negative or positive) of pumping bore water into Ten Mile Brook Dam can be determined by future biodiversity monitoring.

3.0 Objectives

Develop and implement a biodiversity survey program *before* bore water pumping to identify possible effects on aquatic fauna, particularly critically endangered and vulnerable species.

Develop and implement a biodiversity survey program *before* bore water pumping to provide base line data to quantify the impact and changes of the proposed pumping of bore water on aquatic fauna during similar surveys *after* construction.

Provide a final project report to Water Corporation on the aquatic fauna of Ten Mile Brook Dam.

4.0 Planned outcomes

- 1) Detailed description of fish and crayfish fauna (species biodiversity and abundance) in the Ten Mile Brook Dam.
- 2) Identify any potential risks to critically endangered or vulnerable species of pumping bore water into Ten Mile Brook Dam.
- 3) Establish a standardized sampling program to monitor changes in biodiversity before and after pumping bore water into Ten Mile Brook Dam.

5.0 Extension of results

- Extension of results, as a final report, to Water Corporation, water management authorities and aquatic system researchers in other Australian States.
- Development of monitoring plans and strategies for future Water Corporation, research and activities.

6.0 Methods

6.1 Field survey

The Field survey was undertaken in late September 2009 and followed standard sampling procedures used by the Department of Fisheries throughout the state to monitor aquatic biodiversity.

The survey was conducted by establishing a field laboratory (camper trailer containing microscopes, electronic balances, generator, lab bench, dissecting equipment, tagging equipment, lab equipment, live holding tanks, pumps, aerators and Engel freezer for DNA tissue samples) at a suitable location recommended by Water Corporation staff. From this central location on the bank of Ten Mile Brook Dam, sampling equipment (traps, plankton nets, fyke nets, gill nets) were transported and set throughout the dam using a 3.6 m punt with an electric powered outboard motor (Min Kota).

The dam was divided into sampling grids and three zones, each representing approximately 30% of the dam. The northern Zone 1 contained the water off-take for Ten Mile Brook Dam, marked by a white float. This required a minimum 15 m exclusion to prevent fouling of the off-take with sampling gear. Zone 2 consisted of the central third of the dam. Zone 3 consisted of the upper, southern third of Ten Mile Brook Dam.

The biological survey was conducted over 5 days using a variety of methods to target different organisms.

6.2 Freshwater crayfish

Standardized trapping was used to determine species biodiversity, density, abundance and population structure of freshwater crayfish in Ten Mile Brook Dam. This involved setting a

grid pattern of traps (approx 40 m apart) covering the entire water body to survey distribution and abundance of freshwater crayfish. The traps were set late each afternoon and retrieved the next morning. The bait used was a marron pellet. This is because it is pesticide free, water stable, low in nutrients and does not leach nutrients into the waterbody.

DNA fingerprinting techniques developed by Department of Fisheries and The University of Western Australia (UWA) were used to distinguish between “hairy”, hybrid and “smooth” marron to determine if the waterbody contained the critically endangered “hairy” marron. In the field laboratory tissue samples were obtained by removing a rear leg from 104 marron. The legs were immediately prepared, labelled and frozen in the field laboratory. The tissue samples were transported back to UWA for genotyping. A profile was generated for each animal using Random Amplified Polymorphic DNA (RAPD) Polymerase Chain Reaction (PCR). Samples from marron of known gender were genotyped as a control to confirm that any variation was not the result of heterogametic difference. The primer used in this investigation (AL-14) was characterised in a previous study at the University of Western Australia. PCR was performed in a final volume of 10 μ L made up of 1x PCR buffer (Fisher Biotech), 2.0mM MgCl₂ (Fisher Biotech), 200mM dNTP's (Fisher Biotech), 0.5 U Taq polymerase (Fisher Biotech), 0.4mM AL-14 primer (Geneworks), 0.5 μ L of DNA template and high pure water. Amplification was performed in an Eppendorf thermo-cycler according to the conditions described by Fabri *et. al.* (1995): 7mins at 94C followed by 40 cycles of 45 secs at 92C de-naturation, 1 min at 36C annealing, 2 min at 72C extension, and a final single cycle of 72C for 5 minutes. RAPD PCR amplicons were visualised by staining with ethidium bromide after electrophoresis through a 1.5 – 2% agarose gel; genotypes were scored *de visu*.

6.3 Freshwater fish

Gillnets, traps and fyke nets were used to determine species biodiversity, abundance and population structure of freshwater fish in Ten Mile Brook Dam.

Traps were set using the same procedure and bait as reported above for sampling marron, but using a smaller mesh trap. Traps were set late each afternoon and pulled the next morning.

Fyke nets were placed at two locations (two nets per location) in the southern and central Zones, where there was water inflow and therefore likely fish migration. Fyke nets were set each day and inspected every 24 hours.

Unbaited gill nets (1, 1.5, 2, 2.5, 3, 4” str mesh) were set in each zone every afternoon and pulled the next morning.

6.4 Abundance of native fish and freshwater crayfish

Abundance of native fish and freshwater crayfish was determined using CMRR (capture mark release recapture). This involved marking all animals collected (using a tail punch for crayfish and injecting a VIE tag into each fish) (Figure 2). The marked fish and crayfish were then released back in the waterbody. Subsequent recaptures of marked and unmarked fish and crayfish were then used to calculate the abundance of each species in Ten Mile Brook Dam using the Petersen-Lincoln index.



Figure 2. Tagged fish for abundance estimate using CMRR.

While awaiting release all native fish and crayfish were maintained in holding tanks (Figure 3). This process was repeated daily to provide sufficient replication for a robust statistical analysis of abundance.



Figure 3. Tagged fish in holding tank prior to release.

All native fish and crayfish were released back into Ten Mile Brook Dam at the conclusion of the field survey.

6.5 Macroinvertebrates

Invertebrates were sampled by sweeping a plankton net (30 cm x 25 cm) horizontally through 10 m of water at four locations (North, South, East and West). The species composition and abundance of invertebrates was determined on-site using field laboratory equipment and dissecting microscopes. This data was used to calculate the biological condition index of Ten Mile Brook Dam based upon invertebrate water quality sensitivity ratings.

6.6 Water chemistry

Water samples (5 x 1L) were collected from the dam and frozen on site for chemical analysis.

7.0 Results

7.1 Freshwater crayfish biodiversity

Two species of freshwater crayfish were found in Ten Mile Brook Dam, marron and gilgies.

Marron (*Cherax cainii/tenuimanus*)

Marron collected from Ten Mile Brook Dam ranged from 6-570 g in weight. The average weight (mean weight \pm SD) was 116 ± 111 g. Male marron (mean weight \pm SD = 128 ± 114 g) were on average, larger than female marron (mean weight \pm SD = 76 ± 83 g).

The population distribution is bimodal and skewed towards smaller individuals (Figure 4).

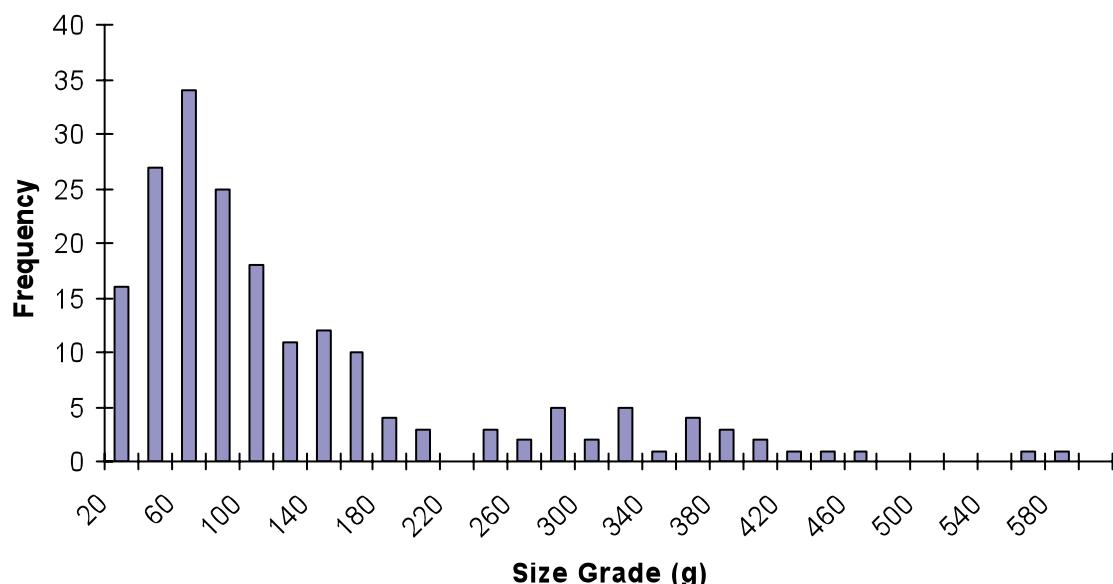


Figure 4. Size frequency distribution of the marron population in Ten Mile Brook Dam.

Ten Mile Brook dam contains approximately 1037 marron (95% confidence interval = 1014 – 1044 marron). This indicates that the dam contains a relatively low total biomass of approximately 120 kg marron (95% confidence interval = 118 – 121 kg).

Gilgie (*Cherax quinquecarinatus*/*Cherax crassimanus*)

Only two Gilgies (13 and 41 g) were collected from Ten Mile brook Dam.

7.2 Freshwater crayfish genetics

RAPD PCR analysis of marron DNA resulted in an array of differently sized amplicon fragments for each animal. An amplicon of approximately 850 base pairs shows clear and considerable size variation and in approximately 30% of samples two bands were visible of approximately this size (Figure 5). Both male and female control animals were genotyped to confirm that this was not the result of heterogametic amplification. Analysis of 104 marron samples resulted in at least five different genotypes after scoring this allele (Figure 5: 1 - 5). When samples that shared a common allele of this size were grouped, analysis of minor bands allowed further genetic subdivision. As a result it can be concluded that the marron population of Ten Mile Brook Dam is heterogenous and does not consist of any one dominant species of marron, rather it is a pool of hybrid animals.

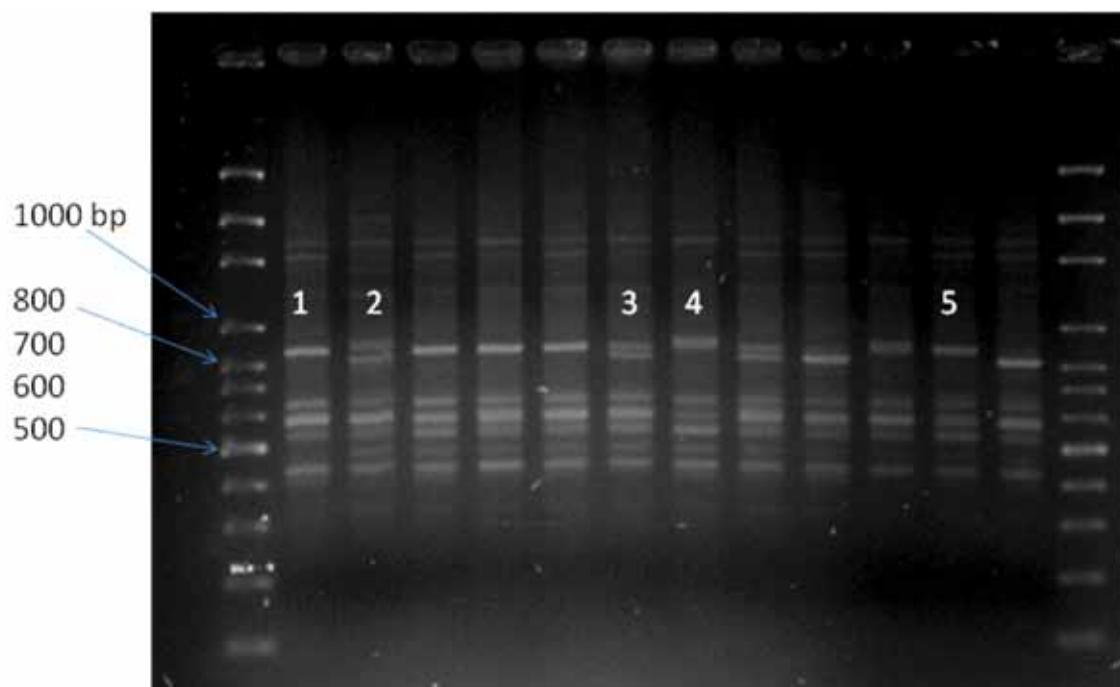


Figure 5. Agarose gel of AL-14 RAPD PCR amplicons. Lane 1 and 14: 100bp molecular size marker. Lanes 2 to 13: DNA profiles generated from Ten Mile Brook Dam marron. Lanes labelled 1 to 5 highlight five different genotypes based on the fragment/fragments slightly larger than 800bp.

7.3 Freshwater fish biodiversity

Two species of native freshwater fish were found in Ten Mile Brook Dam. The Western Minnow, (*Galaxias occidentalis*) (Figure 6), and Nightfish, (*Bostokia porosa*) (Figure 7).



Figure 6. Western Minnow (*Galaxias occidentalis*).



Figure 7. Nightfish (*Bostokia porosa*).

Western Minnow (*Galaxias occidentalis*)

Western Minnows collected from Ten Mile Brook Dam ranged from 45 – 130 mm in length. The average length was $70 \text{ mm} \pm 12 \text{ SD}$.

The population of Western Minnows in Ten Mile Brook Dam follows a normal distribution (Figure 8).

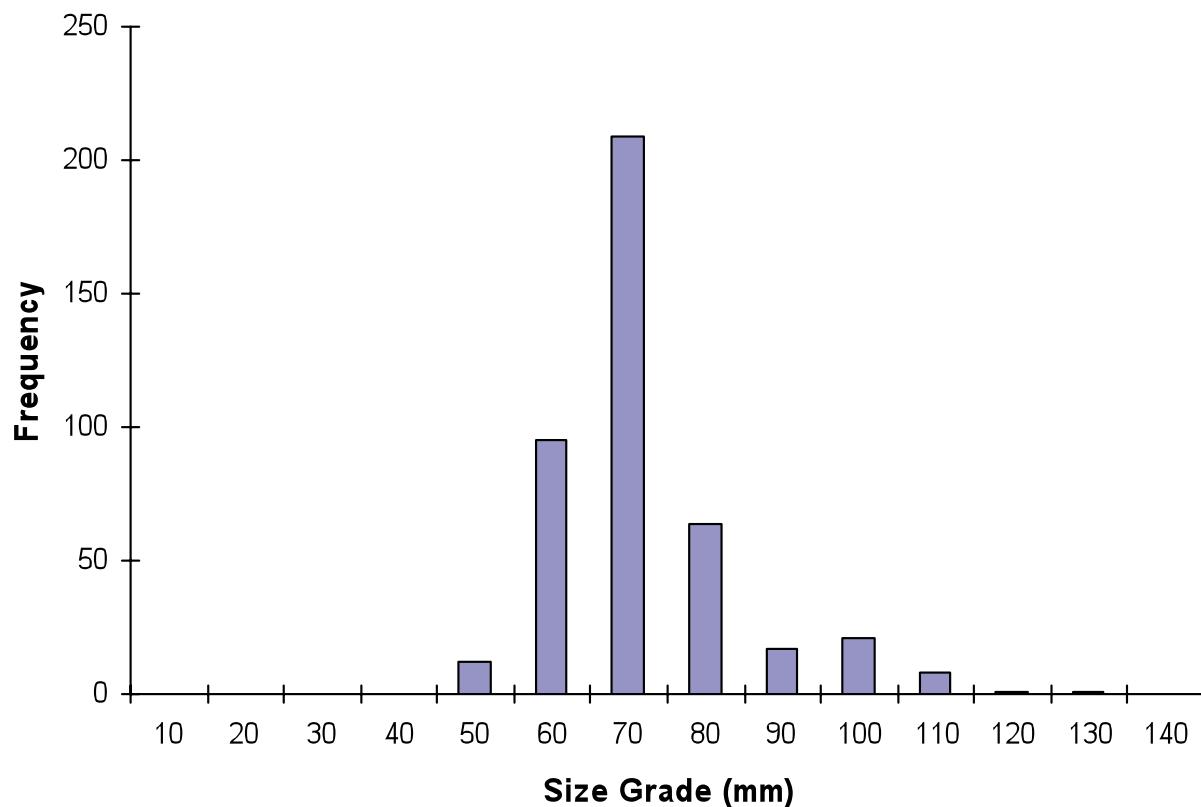


Figure 8. Size frequency distribution of Western Minnow (*Galaxias occidentalis*) population in Ten Mile Brook Dam.

Ten Mile Brook dam contains approximately 22,099 Western Minnows (95% confidence interval = 21,828 – 22,187 Western Minnows).

Nightfish (*Bostokia porosa*)

Three nightfish were collected from Ten Mile Brook Dam (mean length \pm SD = $160 \pm 20 \text{ mm}$). An unusually large Nightfish (180 mm) was collected during this survey of Ten Mile Brook Dam, this is the largest size reported for this species.

Non-native feral fish

Although both trout (*Oncorhynchus mykiss* and *Salmo trutta*) and redfin perch (*Perca fluviatilis*) were reported in Ten Mile Brook Dam in 1998 (Morgan et. al. 1998), neither were found to be present by this study in 2009.

Importantly, the study by Morgan et. al. 1998, did not find any native fish, however the native Western Minnow (*Galaxias occidentalis*) is now abundant in Ten Mile Brook Dam.

7.4 Macroinvertebrates

The dominant macroinvertebrate in Ten Mile Brook Dam is the freshwater shrimp (*Parataya* sp most likely *australiensis*) (Figure 9).



Figure 9. Freshwater Shrimp (*Parataya* sp).

The biodiversity and abundance of invertebrates is low (Table 1). This is likely to be a result of a protected, forested catchment that provides few nutrients, as would be expected for a water storage dam for human consumption.

The composition of macroinvertebrates contains both sensitive (mites) and tolerant (freshwater shrimp) (Table 1). These species are indicators of good to excellent water quality.

Table 1. Macro and Micro invertebrates collected per m³ of water from three 10 m plankton net samples.

Macroinvertebrates	No./m³	Microinvertebrates	No./m³
Freshwater shrimp (Atyidae)	4.55	Rotifers	7.58
Water mite (Arachnida)	1.52	Protozoa	12.12
Seed shrimp (Ostracoda)	1.52	Diatoms	15.15
		Copepods	3.03

7.5 Water chemistry

The water chemistry of Ten Mile Brook dam is consistent throughout the water body, but with very slight variation in alkalinity, ammonia, bicarbonate, calcium, conductivity, nitrate, pH, sulphate, iron and zinc (Table 2).

Table 2. Water Chemistry from Ten Mile brook Dam.

	Units	Site 1	Site 2	Site 3	Site 4	Mean
Inorganic						
Alkalinity as CaCO ₃	mg/L	12	11	10	11	11
Ammonia as NH ₃ -N	mg/L	0.012	0.011	0.010	<0.010	0.011
Bicarbonate as CaCO ₃	mg/L	12	11	10	11	11
Calcium- Filterable	mg/L	4	4	3	3	3.5
Carbonate as CaCO ₃	mg/L	<1	<1	<1	<1	<1
Chloride	mg/L	90	90	90	90	90
Conductivity at 25C	mS/m	38	35	35	34	36
FRP as P	mg/L	<0.005	<0.005	<0.005	<0.005	<0.005
Magnesium- Filterable	mg/L	6	7	7	6	6.5
Nitrate as NO ₃ -N(Calc)	mg/L	0.074	0.078	0.081	0.083	0.079
Nitrite as NO ₂ -N	mg/L	<0.010	<0.010	<0.010	<0.010	<0.010
pH		7.2	6.5	6.5	6.5	6.7
Potassium- Filterable	mg/L	2	2	2	2	2
Sodium- Filterable	mg/L	50	50	50	50	50
Sulfate	mg/L	15	14	15	16	15
Trace Elements						
Copper- Total	mg/L	<0.005	<0.005	<0.005	<0.005	<0.005
Iron- Filterable	mg/L	0.009	0.008	0.008	0.009	0.0085
Iron- Total	mg/L	0.091	0.10	0.091	0.094	0.094
Zinc- Total	mg/L	<0.005	0.013	<0.005	<0.005	0.007

In comparison with water chemistry from both the Margaret River catchment and other water bodies in the southwest bioregion that contain both marron and native fish (Table 3), Ten Mile Dam has slightly lower levels of iron and zinc. However, all water parameters are within the range suitable for freshwater fish and crayfish.

Table 3. Water chemistry from 6 river systems containing marron and native fish populations in Western Australia.

	Units	Margaret	Warren	Donnelly	Harvey	Shannon	Kent	Min	Max	Mean
Inorganic										
Alkalinity	mg/L	10	70	23	8	6	50	6	70	28
Ammonia	mg/L	0.054	0.052	0.027	0.031	0.027	0.03	0.027	0.054	0.037
Bicarbonate	mg/L	10	70	23	8	6	50	6	70	28
Calcium	mg/L	1	27	6	2	2	38	1	38	13
Carbonate	mg/L	<1	<1	<1	<1	<1	<1	<1	<1	<1
Chloride	mg/L	70	570	120	50	70	1300	50	1300	363
Conductivity (25C)	mS/m	25	205	48	19	28	440	19	440	128
ortho-Phosphate	mg/L	<0.005	<0.005	<0.005	<0.005	<0.005	<0.005	<0.005	<0.005	<0.005
Magnesium	mg/L	5	58	11	4	5.7	127	4	127	35

Total Oxidised Nitrogen	mg/L	<0.01	0.11	0.13	<0.01	0.013	0.016	<0.01	0.016	0.048
pH		6.5	7.7	7.1	6.7	6.3	7.1	6.3	7.7	6.9
Potassium	mg/L	1	3	2	<1	1.6	4.9	1	4.9	2.3
Sodium	mg/L	26	250	55	26	40	1500	26	1500	316
Sulphate	mg/L	<5	40	13	6	6	92	6	92	27
Trace Elements										
Copper - Total	mg/L	<0.001	<0.001	<0.001	<0.001	<0.001	0.004	<0.001	0.004	<0.001
Iron - Total	mg/L	2.5	0.6	0.3	0.1			0.1	2.5	0.9
Zinc - Total	mg/L	0.028	0.018	0.016	0.016	0.018	0.028	0.016	0.028	0.021

8.0 Conclusions

Ten Mile Brook Dam contains both native fish (Western minnows, Nightfish), and crayfish (Marron and Gilgies). No feral fish or crayfish species were found in Ten Mile Brook Dam.

The marron population of Ten Mile Brook Dam is heterogenous and does not consist of any one dominant species; rather it is a pool of hybrid animals. These hybrids represent 5 different genotypes, none of which are pure critically endangered “hairy” marron *C. tenuimanus*.

The abundance of fish and crayfish in Ten Mile Brook Dam is likely to be limited by the low levels of nutrients in the water column and consequently low abundance of macroinvertebrate prey.

9.0 Recommendations

- Twelve months after pumping commences a biological survey should be conducted and data compared with that of this baseline survey. This will enable Water Corporation to quantify the impact and changes (negative, nil or positive) of the pumping of bore water on aquatic fauna in Ten Mile Brook Dam.
- The existing hybrid marron in Ten Mile Brook Dam should be removed and replaced with pure stocks of the endemic critically endangered “hairy” marron *Cherax tenuimanus*. This would achieve two outcomes, firstly it would prevent genetic pollution caused by the feral marron species currently residing in Ten Mile Brook Dam entering the Margaret River catchment, and secondly it could prevent the extinction of the critically endangered “hairy” marron *Cherax tenuimanus* by establishing this waterbody as a key repository or ark for this species.

10.0 Acknowledgements

Rebecca Richardson (Water Corporation) for the efficient co-ordination of this study.

Tony Booth (Water Corporation) and Water Corporation Rangers for access to the dam and safety induction.