Inland Waters & Catchment Ecology

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The Critical Fish Habitat Project

Assessment of the success of reintroductions of threatened fish species in the Coorong, Lower Lakes and Murray Mouth region 2011–2014

C. Bice, N. Whiterod and B. Zampatti

SARDI Publication No. F2012/000348-3
SARDI Research Report Series No. 792

SARDI Aquatics Sciences
PO Box 120 Henley Beach SA 5022

July 2014

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EXECUTIVE SUMMARY

The Critical Fish Habitat (CFH) project was developed in 2011 to provide a framework to guide and undertake reintroductions of threatened fish species to the Coorong, Lower Lakes and Murray Mouth (CLLMM) region, namely Yarra pygmy perch (*Nannoperca obscura*), southern pygmy perch (*Nannoperca australis*), Murray hardyhead (*Craterocephalus fluviatilis*) and southern purple-spotted gudgeon (*Mogurnda adspersa*), following extirpation and population declines during drought from 2007–2010. The primary objective of the CFH project was to re-establish self-sustaining wild populations of all four species. The CFH project involved the identification of potential receiving sites, assessment of site suitability (e.g. presence of favourable habitat, water quality and prey resources) and finally, development of methods for undertaking fish releases. From spring 2011 to autumn 2013, >15,800 fish, collectively across all four species, were released at 10 sites within the CLLMM region. In 2013/14, the current project aimed to (1) determine the status (e.g. distribution, abundance, recruitment) of Yarra pygmy perch, southern pygmy perch, Murray hardyhead and southern purple-spotted gudgeon in the CLLMM region, and (2) provide an initial evaluation of the success of reintroductions towards meeting the objective of re-establishing self-sustaining wild populations.

Since reintroductions commenced in spring 2011, a total of 403 threatened fish, across all four target species, have been sampled from 11 sites in the Lower Lakes. Murray hardyhead have been sampled in the greatest numbers (n = 333), followed by southern pygmy perch (n = 43), Yarra pygmy perch (n = 14) and southern purple-spotted gudgeon (n = 13). The abundance of Murray hardyhead has increased consistently since spring 2011 and the species is now broadly distributed in the Lower Lakes. Increased abundance likely reflects recruitment in the remnant wild fish population, but reintroductions have potentially supplemented and enhanced the wild population. Whilst they are present, and low levels of wild recruitment have been observed, populations of the remaining three species have not exhibited increases in abundance and distribution following initial reintroduction. As of autumn 2014 the presence of Yarra pygmy perch and southern pygmy perch are confirmed from just one site each, in low abundances, whilst southern purple-spotted gudgeon, was not detected during this round of sampling (this species was detected in spring 2013). Thus, as of autumn 2014, populations of these three species in CLLMM region are potentially not sustainable and remain at high risk of extirpation.

The reintroduction of fish into wild habitats to assist the restoration of viable, self-sustaining populations is a difficult task and an objective, with the exception of Murray hardyhead, that

remains an aspirational objective of the current project. The continued detection of all four species is encouraging, and in itself a partial success. Nevertheless, further reintroductions are likely required. Importantly, based on similar threatened fish reintroduction programs in Australia and internationally, prolonged (up to ten years) annual reintroduction efforts are most likely to deliver self-sustaining populations in the CLLMM region. Notably, habitat conditions within the CLLMM region are continuing to improve following the end of the drought (2010) and source populations (i.e. surrogate refuge dams) for three of the species (Yarra pygmy perch, Murray hardyhead and southern purple-spotted gudgeon) remain in good condition, meaning further reintroduction is a viable option for the conservation of these species in the CLLMM region in the future.

1. INTRODUCTION

1.1. Background

River regulation and a history of over-abstraction have greatly reduced freshwater flows throughout the Murray-Darling Basin (MDB) (Kingsford 2000). The situation was exacerbated over the period 1997–2010 when the most severe drought in recorded history was experienced in the MDB (Van Dijk *et al.* 2013), resulting in significantly diminished freshwater flows to the lower River Murray, South Australia. In the Coorong, Lower Lakes and Murray Mouth (CLLMM) region at the terminus of the MDB, water level in Lake Alexandrina fell below sea level for the first time in recorded history, accompanied by significant reductions in submerged aquatic vegetation cover, disconnection of fringing vegetation habitats and elevated salinity (Kingsford *et al.* 2011). This in turn resulted in substantial declines in threatened freshwater fish species in the CLLMM region (Wedderburn *et al.* 2012), several of which were exposed to extreme risk of local extinction. Subsequently, measures were taken to prevent the extirpation of select threatened species in the CLLMM region through the 'South Australian Drought Action Plan (DAP) for Murray-Darling Basin Threatened Freshwater Fish Populations' (Hall *et al.* 2009). In several instances this necessitated removal of individuals from the wild, captive maintenance and breeding, with the objective of reintroducing fish to wild habitats upon the return of favourable conditions (see Hammer *et al.* 2013).

Captive maintenance and breeding programs were established for populations of four species considered as threatened nationally and/or within South Australia; namely Yarra pygmy perch (*Nannoperca obscura*), southern pygmy perch (*Nannoperca australis*), Murray hardyhead (*Craterocephalus fluviatilis*) and southern purple-spotted gudgeon (*Mogurnda adspersa*) (Table 1). Concurrently, Flinders University, together with several other industry partners, initiated a project with the objective of enhancing the captive breeding programs by determining breeding pair choices for optimal offspring genetic fitness for each species (Carvalho *et al.* 2011, 2012a and 2012b). Captive maintenance and breeding programs involved collaboration between many different agencies including Aquasave – Nature Glenelg Trust, Native Fish Australia (SA), The Department of Environment, Water and Natural Resources (DEWNR), Flinders University, South Australian Research and Development Institute (SARDI), the Murray-Darling Freshwater Research Centre (MDFRC; Mildura), Alberton Primary School and Urrbrae Agricultural College.

Table 1. Summary of key threatened fish species in the CLLMM region and their conservation status. Conservation status is coded as Critically Endangered (CR); Endangered (E); Vulnerable (VU); Rare (R); and Protected (P) at international (International Union for Conservation of Nature) national (*Environment Protection and Biodiversity Conservation Act 1999*) and state (*Fisheries Management Act 2007*) levels, including interim state conservation listings (Hammer *et al.* 2009b).

Species	**International** (IUCN)	**National** (EPBC Act)	**State Fisheries**	**Action Plan 09**	**Local significance**
Yarra pygmy perch (*Nannoperca obscura*)	VU	VU	P	CR	A genetically distinct population of this species. Lake Alexandrina represents the only known MDB population.
Southern pygmy perch (*Nannoperca australis*)	-	-	P	E	SA MDB fish are genetically distinct and diverse (populations are found only in the Lower Lakes and their tributaries)
Murray hardyhead (*Craterocephalus fluviatilis*)	E	E	P	CR	Endemic species, few populations remain (two genetically different populations in SA, one of which is present in the lower lakes)
Southern purple-spotted gudgeon (*Mogurnda adspersa*)	-	-	P	CR	Only known southern MDB population (present in the Lower Murray region, below Lock 1, historical records in CLLMM region)

With the return of favourable hydrology to the CLLMM region in 2010−11 there was deemed to be potential for threatened species, maintained and bred as part of the various captive breeding programs, to be reintroduced to wild habitats. The Critical Fish Habitat (CFH) project was developed to provide a scientifically rigorous framework to guide and undertake reintroductions of threatened fish in the CLLMM region; the primary objective of the project was to facilitate the re-establishment of self-sustaining wild populations, which require minimal management attention (Hammer *et al.* 2009a; Watt *et al.* 2011). This framework considered many factors including knowledge and status of threatening processes, past and current environmental conditions, and species' former range and biology, and was largely adapted from the framework of Hammer *et al.* (2009a) and a review by George *et al.* (2009). The framework aimed to enhance the likelihood of success of the current reintroductions by (1) identifying potential receiving sites, (2) developing and undertaking a screening process to assess the suitability of receiving sites, (3) determining a method for fish release (i.e. numbers, spatial extent, transport, acclimatisation, etc.), (4) undertaking fish releases and (5) developing and conducting a monitoring and evaluation program to assess the success of the reintroductions (Hammer *et al.* 2009a, Watt *et al.* 2011).

Over 2011–2013, four rounds of site assessment and fish reintroductions occurred, with >15,800 fish, from the four threatened species, released at ten locations across the CLLMM region (Bice *et al.* 2012, 2013). Post-reintroduction monitoring, up until autumn 2013, detected evidence of 'wild survival' for all four species and some evidence of wild recruitment for Murray hardyhead, Yarra pygmy perch and southern pygmy perch (Bice *et al.* 2013). Nonetheless, whilst positive signs of population establishment were exhibited by all species, they remained present at a limited number of sites (individual sites in some cases) in limited abundance. Thus, populations of each species remained highly vulnerable to stochastic events and the objective of self-sustaining wild populations, that require minimal management attention, had likely not been met.

1.2. Objectives

Integral to any reintroduction program is the monitoring of reintroduced populations and subsequent assessment of the success of reintroduction activities (Fischer and Lindenmayer 2000). During 2013/14, further monitoring of threatened fish populations in the CLLMM region was undertaken. In this report, these data are integrated with post-reintroduction monitoring data from 2011–2013 to meet the following two objectives,

1. To assess site condition (physical habitat and physico-chemical parameters) and the current status (i.e. distribution, abundance and evidence of recruitment) of populations of Murray hardyhead, Yarra pygmy perch, southern pygmy perch and southern purple-spotted gudgeon in the CLLMM region; and
2. To provide an evaluation of the success of the CFH Project in facilitating the establishment of 'self-sustaining wild populations' of these species.

2. METHODS

2.1. Sites and reintroductions

From 2011 to 2013, a total of 22 sites across the CLLMM region were selected and considered as potential reintroduction sites under the CFH project (Figure 1; Table 2). These sites were selected based upon knowledge of the previous presence and abundance of these species, with particular consideration of these population metrics in, or prior to, 2005 (Bice *et al.* 2012 and references therein). Sites where water security in the immediate future was assured were evaluated for their suitability for reintroductions in regards to a range of species-specific biotic and abiotic parameters. Species-specific physico-chemical and habitat criteria were set to ensure fish were returned to habitats that were favourable in regards to water quality, provision of resources (e.g. prey abundance) and favourable habitat (e.g. shelter and spawning habitat), whilst not placing individuals at undesirable levels of risk regarding intense competition or predation; thus maximising the likelihood of success from reintroductions (see Bice *et al.* 2012, 2013).

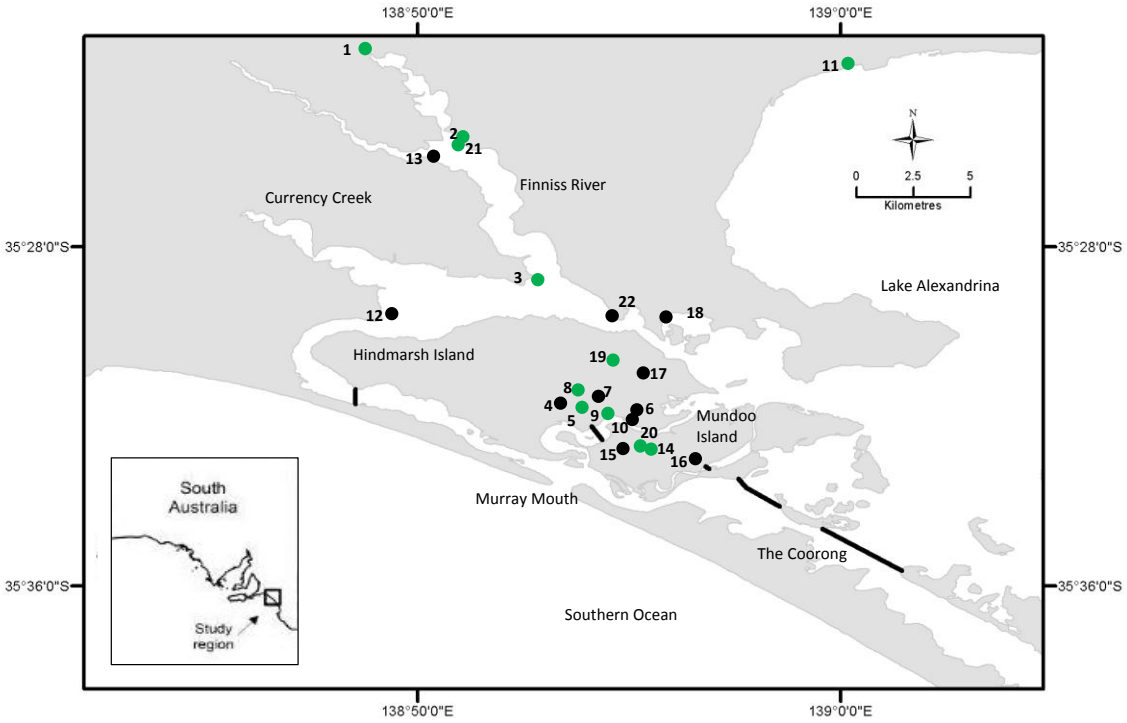

Figure 1. Summary of potential (black) and actual (green) reintroduction sites for southern purple-spotted gudgeon, Yarra pygmy perch and Murray hardyhead in the Coorong, Lower Lakes and Murray Mouth (CLLMM) region in 2012/13. Murray Barrages are indicated by black lines. Site numbers are cross referenced in Table 2.

Table 2. List of proposed receiving sites for reintroductions of southern purple-spotted gudgeon (SPSG), Yarra pygmy perch (YPP), Murray hardyhead (MHH) and southern pygmy perch (SPP). These sites (with the exception of Eastick Creek) were monitored in 2013/14.

Site #	Site name	Sub-region	Proposed species	Latitude	Longitude	Sampled		Reference
						Spring 2013	Autumn 2014	
1	Finniss at Winery Road	Lower Finniss	SPSG, YPP	35.396269 S	138.826406 E	Y	Y	(Hammer *et al.* 2009a)
2	Blue Lagoon (Pembroke)	Lower Finniss	YPP	35.429166 S	138.859059 E	Y	Y	(Wedderburn and Hammer 2003)
21	Blue Lagoon (outer site near river channel)	Lower Finniss	YPP	35.409380 S	138.839267 E	Y	Y	(Wedderburn and Hammer 2003)
3	Finniss River junction	Goolwa Channel	YPP, SPP	35.486760 S	138.893200 E	Y	Y	(Hammer 2008)
4	Hunters Creek (upstream of Denver Rd causeway)	Hindmarsh Island	YPP, SPP, MHH	35.527571 S	138.897927 E	Y	Y	(Wedderburn and Hammer 2003, Bice and Ye 2006, Bice and Ye 2007)
5	Hunters Creek (downstream of Denver Rd causeway)	Hindmarsh Island	YPP, SPP, MHH	35.527021 S	138.893191 E	Y	Y	(Wedderburn and Hammer 2003, Bice and Ye 2006, Bice and Ye 2007)
6	Eastick Creek	Hindmarsh Island	YPP, SPP, MHH	35.536366 S	138.921670 E	N	N	
7	Upper Hunters Creek (Drain behind Wyndgate)	Hindmarsh Island	YPP, SPP	35.527249 S	138.904974 E	Y	Y	(Bice and Ye 2006)
8	Natural channel connected to Hunters Creek	Hindmarsh Island	YPP, SPP, MHH	35.525690 S	138.898997 E	Y	Y	(Higham *et al.* 2005, Bice and Ye 2006, Bice and Ye 2007)
9	Steamer drain	Hindmarsh Island	YPP, SPP	35.53146 S	138.90810 E	Y	Y	(Bice *et al.* 2011)
10	Holmes Creek at Eastick Creek mouth	Hindmarsh Island	YPP	35.53778 S	138.92175 E	Y	Y	(Bice and Ye 2007, Hammer 2007a, 2008)
11	Turvey's Drain	Milang	SPP	35.39462 S	139.00787 E	Y	Y	(Bice *et al.* 2009, Hammer 2009, Bice *et al.* 2010)
12	Currency Creek Game Reserve	Goolwa Channel	YPP	35.49335 S	138.82333 E	Y	Y	(Hammer 2008)
13	Black Swamp	Lower Finniss	YPP	35.43119 S	138.84875 E	Y	Y	(Hammer 2009)
14	Mundoo Island Channel east*	Mundoo Island	MHH	35.54765 S	138.91821 E	Y	Y	(Wedderburn and Barnes 2009, Wedderburn and Hillyard 2010)
20	Mundoo Island Channel east 2	Mundoo Island	SPP, MHH	35.54877 S	138.92422 E	Y	Y	(Bice *et al.* 2012)
15	Mundoo Island Channel west*	Mundoo island	SPP, MHH	35.54848 S	138.91566 E	Y	Y	(Wedderburn and Barnes 2009, Wedderburn and Hillyard 2010)
22	Old Clayton*	Clayton	MHH	35.49398 S	138.91138 E	Y	Y	(Wedderburn and Barnes 2009, Wedderburn and Hillyard 2010)
17	Boggy Creek*	Hindmarsh Island	MHH	35.52107 S	138.92888 E	Y	Y	(Wedderburn and Barnes 2009, Wedderburn and Hillyard 2010)
18	Dunn's Lagoon*	Clayton	MHH	35.50246 S	138.93180 E	Y	Y	(Wedderburn and Hammer 2003, Bice and Ye 2007, Wedderburn and Barnes 2009, Wedderburn and Hillyard 2010)
19	Shadows Lagoon*	Hindmarsh Island	YPP	35.51738 S	138.91756 E	Y	Y	(Wedderburn and Barnes 2011)

**Denotes sites monitored by the University of Adelaide as part of the Murray-Darling Basin Authority's 'The Living Murray' Program.*

Following detailed fish, macroinvertebrate and habitat monitoring through 2011–2013, a total of 10 sites were deemed favourable for the release of threatened fishes. This involved the release of a total of 15,840 fish from the four threatened species (~7,520 Murray hardyhead at two sites, ~5,850 Yarra pygmy perch at five sites, ~ 1,350, southern pygmy perch at three sites and ~ 1,120 southern purple-spotted gudgeon at one site) (Table 3).

Table 3. Summary of sites (site number in brackets) and the numbers of Yarra pygmy perch (YPP), southern pygmy perch (SPP), southern purple-spotted gudgeon (SPSG) and Murray hardyhead (MHH) released over 2011–2013. The source of reintroduced fish is coded as either (1) surrogate dams (Crouch Dam (CD), Oster Dam (OD), Tupelo Grove Nursery (TGN) or Munday Dam (MD)), (2) Flinders University (FU, either equal contribution from broodstock (EC) or unequal contribution from broodstock (UC)), (3) the Aquasave Hatchery (AQ). See Bice *et al.* (2013) for hatchery/surrogate dam details.

Site name (number)	Species released	Numbers released (approx.)	Source
Reintroductions spring 2011			
Natural channel connected to Hunters Creek (8)	SPP	770	FU (EC)
Turvey's drain (11)	SPP	300	Wild, FU (UC)
Blue Lagoon (2 & 21)	YPP	400	CD
Finniss River junction (3)	YPP	800	CD
Finniss at Winery Road (1)	SPSG	200	AQ
Reintroductions autumn 2012			
Mundoo Island Channel east 2 (20)	SPP	280	FU (UC)
Streamer Drain (9)	YPP	2200	FU (EC)
Shadows Lagoon (19)	YPP	1500	CD, OD, FU (UC)
Finniss at Winery Road (1)	SPSG	400	AQ, AP
Mundoo Island Channel east (14)	MHH	3500	MD
Reintroductions spring 2012			
Mundoo Island Channel east (14)	MHH	3500	MD
Hunters Creek (d/s) (5)	YPP	400	TGD
	MHH	520	FU (EC)
Shadows Lagoon (19)	YPP	250	CD
Finniss at Winery Road (1)	SPSG	320	AQ
Reintroductions autumn 2013			
Hunters Creek (d/s) (5)	YPP	300	CD
Finniss at Winery Road (1)	SPSG	200	AQ

2.2. Fish monitoring and site condition assessment

All sites presented in Table 2 were sampled in spring 2013 and autumn 2014 (with the exception of Eastick Creek) to assess population status and evaluate the success of the CFH project. These included all sites where reintroductions had occurred and several of the originally selected potential reintroduction sites, as dispersal of reintroduced individuals and thus re-colonisation may have occurred at these sites.

Fish were monitored in spring 2013 (25/11/2014–29/11/2014) and autumn 2014 (31/03/2014–04/04/2014). All sites, except the Finniss River at Winery Road, were sampled with five single-winged fyke nets (four 6 m wing length, 0.6 m entry diameter and 0.003 m mesh; one 3 m wing length, 0.6 m entry diameter and 0.004 m mesh) set overnight. Fyke nets were set perpendicular to the bank, where possible, in habitat that was representative of the site being sampled. The Finniss River at Winery Road was sampled using a Smith-Root model LR-24 backpack electrofisher and a series of 20 box traps (0.4 m length x 0.24 m width x 0.24 m height, 0.03 m opening). This site was sampled with this technique due to its differing physical characteristics and also the potential ineffectiveness of fyke nets for sampling southern purple-spotted gudgeon in complex habitat. Several sites were monitored by the University of Adelaide (indicated throughout) as part of the Murray-Darling Basin Authority's (MDBA) *The Living Murray Program* and data are shared between these projects (Table 2).

All fish sampled were identified to species (Lintermans *et al.* 2007) and enumerated. All threatened and non-native species (i.e. redfin perch and common carp) sampled were measured for length (mm, total length (TL) or fork length (FL) depending on tail morphology) for up to 50 individuals per species per site. Non-native species were not returned to the water. Sampling was conducted under a *Section 115 permit* in accordance with the *Fisheries Management Act 2007* and Primary Industries and Regions South Australia (PIRSA) Animal Ethics Committee standards.

Site 'condition' was assessed to determine the continued suitability, in relation to threatened species requirements, of reintroduction sites and surrounding sites. To assess site condition, the composition of available physical habitat was evaluated and water physico-chemical parameters described. Physical habitat cover was described (by visual estimation) as the proportion of aquatic habitat area (i.e. below the water surface) comprised of submerged vegetation, emergent vegetation, other physical structure (e.g. woody debris, rock) and open water. A series of random depth measures were also taken to determine mean depth at the site and a maximum depth was also determined by attempting to locate the deepest point at the site. Various physico-chemical parameters were measured at each site. Turbidity was measured as secchi depth (m) using a secchi disk, whilst the following parameters were measured with a TPS 90-FLT water quality meter: conductivity ($\mu S.cm^{-1}$), pH, dissolved oxygen (ppm, readings at surface and at depth) and temperature (˚C). The 'condition' of all sites sampled in 2013/14 was assessed against species-specific criteria used in previous years to determine the suitability of sites for reintroductions of threatened fishes (Bice *et al.* 2012, 2013) (Table 4).

Table 4. Species-specific criteria for determining 'site suitability' for threatened fish species in the CLLMM region in 2013/14.

Target species	Physico-chemical parameters			Aquatic habitat			Food resources	Introduced predators/competitors abundance				References
	Salinity (μScm^{-1})	Dissolved oxygen (mgL^{-1})	pH	Species composition (submerged)	Species composition (emergent)	Percentage (%) physical habitat	Presence of known prey resources (Y/N)	Redfin perch (>120mm)	Juvenile common carp (<100mm)	Adult common carp (>250mm)	Eastern gambusia	
Yarra pygmy perch	<3000	>2.0	4-10	*Myriophyllum* spp, *Ceratophyllum demersum*, *Vallisneria australis*	*Schoenoplectus validus*	>50%	Y/N	<15 per 4 nets	<30 per net	<20 caught or observed	<100 per net	(Roberts *et al.* 1995, Mittlebach and Persson 1998, Wedderburn and Hammer 2003, Bice and Ye 2006, Hammer 2007b)
Southern pygmy perch	<3000	>2.0	4-10	*Myriophyllum* spp, *Ceratophyllum demersum*, *Vallisneria australis*	*Typha* spp and overhanging and fringing grasses	>50%	Y/N	<15 per 4 nets	<30 per net	<20 caught or observed	<100 per net	(Roberts *et al.* 1995, Mittlebach and Persson 1998, Hammer 2004, 2005, McNeil and Closs 2007)
Murray hardyhead	800-25,000	>2.0	4-10	*Myriophyllum* spp, *Potamogeton pecinatus, Ruppia* spp., *Vallisneria australis*	*Paspalum distichum,* cooch, other	>30%	Y/N	<15 per 4 nets	<30 per net	<20 caught or observed	<100 per net	(Mittlebach and Persson 1998, Wedderburn and Hammer 2003, Bice and Ye 2006, Bice and Ye 2007, Hammer and Wedderburn 2008, Wedderburn *et al.* 2008, Hammer *et al.* 2009b, Bice *et al.* 2011)
Southern purple-spotted gudgeon	800-5,000	>3.0	7-10	*Myriophyllum* spp, *Ceratophyllum demersum*, *Vallisneria australis*	*Schoenoplectus validus, Triglochin procerum*	>30% (includes other phys habitat e.g. woody debris)	Y/N	<15 per 4 nets	<30 per net	<20 caught or observed	<100 per net	(Nettlebeck 1926, Roberts *et al.* 1995, Mittlebach and Persson 1998, Llewellyn 2006, Hammer *et al.* 2009a)

2.3. Population status and assessment of reintroduction success

Detailed assessment of population status requires determination of (a) species presence, distribution and abundance, and (b) population demographics. Abundances of threatened species in 2013–14 were compared with abundance from sampling conducted in 2011–2013 to determine trends in abundance and length-frequency analyses was used to investigate post reintroduction recruitment dynamics. All four target species are relatively short-lived (from 1 year to several years) and investigation of length-frequency distributions through time is effective in determining recruitment/population dynamics.

Additionally, the ability to differentiate between wild produced (i.e. remnant wild fish or progeny of reintroduced fish) and recaptured reintroduced fish provides important insight on the success of reintroduction programs (e.g. are increases in abundance due to reintroductions or wild recovery?). Therefore all fish reintroduced under the current project were marked with calcein prior to reintroduction (see Bice *et al.* 2013). Calcein is a fluorescent chemical dye, which when applied through the process of osmotic induction, may produce an external and non-lethal detectable mark on fish (Mohler 2003, Crook *et al.* 2009, Smith *et al.* 2010). Issues with mark retention in both Murray hardyhead and southern purple-spotted gudgeon were noted in 2012/13 (Bice *et al.* 2013, Westergaard 2013) and subsequently, readings of fluorescence from wild caught fish in 2013/14 were limited to pygmy perch species. A total of three readings of fluorescence (sample fluorescence ratio) were taken from each pygmy perch sampled, using an Opti-sciences® GFP-meter. Readings were taken from the gills (alternating between sides) as this area exhibits the greatest retention of calcein (Westergaard 2013). Maximum fluorescence values were used for all analyses (Crook *et al.* 2009), with a reading of >300 units deemed to represent positive detection of a calcein 'mark' (Westergaard 2013; SARDI unpublished data).

Success of the reintroductions of each species to date was assessed by determining trends (i.e. positive, negative or static) in distribution (changes in the number of sites where detected) and abundance, evidence of wild recruitment and evidence of the contribution of reintroductions to these trends since 2011. Nevertheless, returning populations of these species to a status similar to pre-2007 (prior to dramatic declines) should be a long-term goal in the conservation of these species. Substantial data exists on the distribution and abundance of these species prior to significant water level recession in the Lower Lakes in 2007 (Hammer *et al.* 2002, Wedderburn and Hammer 2003, Bice and Ye 2006), and the development of appropriate population metrics (e.g. area of occupancy, number of sites, relative abundance, etc.) as 'recovery benchmarks' is a priority.

3. RESULTS

3.1. Catch summary

A total of 8,988 fish from 23 species were sampled in 2013/14 (Table 5). This included all four threatened species, which were sampled in low abundance, with the exception of Murray hardyhead, which was sampled in moderate abundance. In spring 2013, amoungst the other fish sampled, the most abundant species were flat-headed gudgeon (*Philypnodon grandiceps*; 30% of total catch), common galaxias (*Galaxias maculatus*; 29%), eastern gambusia (*Gambusia holbrooki*; 10%) and carp gudgeon (*Hypseleotris* spp.; 8%). In autumn 2014, the most abundant species were eastern gambusia (32%), flat-headed gudgeon (20%), unspecked hardyhead (*Craterocephalus stercusmuscarum fulvus*; 19%) and common galaxias (9%).

Table 5. Species, total number and number of sites from which fish were sampled in spring 2013 and autumn 2014.

		Spring 2013		Autumn 2014	
Common name	**Scientific name**	**Abundance**	**No. sites**	**Abundance**	**No. sites**
Murray Hardyhead[@]	*Craterocephalus fluviatilis*	47	1	198	4
Southern pygmy perch[@]	*Nannoperca australis*	1	1	14	1
Yarra pygmy perch[@]	*Nannoperca obscura*	1	1	1	1
Southern purple-spotted gudgeon[@]	*Mogurnda adspersa*	6	1	0	0
Golden perch	*Macquaria ambigua ambigua*	7	4	4	3
Unspecked hardyhead[@]	*Craterocephalus stercusmuscarum fulvus*	116	6	1097	8
Carp gudgeon[@]	*Hypseleotris* spp.	274	14	88	12
Flat-headed gudgeon[@]	*Philypnodon grandiceps*	973	16	1177	17
Dwarf flat-headed gudgeon[@]	*Philypnodon macrostomus*	43	11	40	8
Australian smelt[@]	*Retropinna semoni*	110	9	31	6
Bony herring[@]	*Nematalosa erebi*	44	8	434	9
Common galaxias[^]	*Galaxias maculatus*	959	13	533	15
Congolli[^]	*Pseudaphritus urvillii*	125	15	110	14
Tamar River goby[*]	*Afurcagobius tamarensis*	0	0	1	1
Western blue-spot goby[*]	*Pseudogobius olorum*	0	0	1	1
Lagoon goby[*]	*Tasmanogobius lasti*	9	4	0	0
Small-mouthed hardyhead[*]	*Atherinosoma microstoma*	1	1	6	3
Sandy sprat[*]	*Hyperlophus vitattus*	2	1	0	0
River garfish[*]	*Hyperhamphus regularis*	0	0	1	1
Eastern gambusia[e]	*Gambusia holbrooki*	326	13	1825	16
Redfin perch[e]	*Perca fluviatilis*	100	10	91	9
Common carp[e]	*Cyprinus carpio*	83	11	39	11
Goldfish[e]	*Carassius auratus*	50	8	20	8
	Totals	3277	20	5711	20

[@]*Freshwater fish,* [^]*Catadromous fish,* [*]*Estuarine fish,* [e]*Non-native fish*

3.2. Site condition

The ‘condition’ of all sites sampled in 2013/14 was assessed against species-specific criteria used in previous years to determine ongoing suitability of sites for threatened fishes (although reintroductions were not conducted) (Bice *et al.* 2012, 2013) (Table 4). Sites were deemed as ‘suitable’ if the target species was present or all species-specific criteria were met. In spring 2013, 11 sites were deemed suitable for habitation by threatened fish species (Yarra pygmy perch: 5 sites, southern pygmy perch: 1 site, Murray hardyhead: 7 sites, southern purple-spotted gudgeon: 1 site) (Table 6). In autumn 2014, the number of sites deemed suitable for habitation by threatened fish species increased to 16 (Yarra pygmy perch: 7 sites, southern pygmy perch: 6 sites, Murray hardyhead: 8 sites, southern purple-spotted gudgeon: 1 site) (Table 7). This represents an increase in the number of sites exhibiting conditions favourable to threatened species since 2012/13 (11 sites in spring 2012 and 8 sites in autumn 2013) (Bice *et al.* 2013), and reflects continued improvement of aquatic habitat following the return of favourable water levels to the Lower lakes in 2010/11.

Table 6. Summary of site condition assessments conducted in spring 2013. Cells coloured in green indicate criteria (Table 4) were met, whilst red cells indicate criteria were not met.

Site name (number)	Target spp	Native species		Water quality			Aquatic habitat			Introduced predators/competitors				Assessment/ comments
		Target spp Detect	Native spp	EC	DO	pH	Species composition (submerged)	Species composition (emergent)	% physical habitat	Redfin perch (>120mm)	Juvenile common carp (<100mm)	Adult common carp (>250mm)	Eastern gambusia	
Finniss at Winery Road (1)	SPSG	YES	2	2016	3.5	7.5	n/a	*Typha, Phragmites, Triglochin, Berula, Paspalum*	50	0	0	0	0	**Site appears favourable for target species**
Blue Lagoon (Pembroke) (2)	YPP	NO	8	1740	11.1	8.3	n/a	*Typha, Phragmites, Schoenoplectus,* grass	40	3	0	4	0	**Potentially unsuitable. No submerged vegetation and total physical habitat below criteria**
Blue Lagoon (outer) (21)	YPP	NO	10	1780	12.6	8.2	*Myriophyllum*	*Typha, Phragmites, Schoenoplectus*	45	3	0	1	27	**Potentially unsuitable. Total physical habitat below criteria**
Finniss River junction (3)	MHH, YPP	YES (MHH) NO (YPP)	11	826	15.8	8.8	*Myriophyllum*	*Schoenoplectus, Typha, Phragmites*	50	0	9	2	13	**Site appears favourable for both target species**
Hunters Creek upstream rd (4)	YPP, SPP, MHH	NO	3	1230	8.6	8.0	*Myriophyllum, Potamogeton crispus*	*Typha,* grass	35	1	0	0	0	**Potentially unsuitable for YPP and SPP Total physical habitat below criteria.** **Site appears favourable for MHH**
Hunters Creek downstream road (5)	YPP, SPP, MHH	NO	7	1310	10.0	8.2	*Myriophyllum, Potamogeton crispus, Vallisneria*	*Schoenoplectus, Typha, grasses*	15	0	0	0	0	**Potentially unsuitable. Total physical habitat below criteria**
Steamer Drain (9)	YPP, SPP	NO	3	899	1.3	7.4	*Myriophyllum, Ceratophyllum* , algae	*Typha, Bolboschoenus,* grasses	80	0	17	1	119	**Potentially unsuitable. Dissolved oxygen below criteria**
Natural connected to Hunters Creek (8)	YPP SPP	YES (SPP)	5	3140	11.0	8.1	*Myriophyllum, Ceratophyllum*	*Typha, Bolboschoenus, Juncus,* grasses	50	0	2	0	14	**Salinity slightly elevated, however, site still appears favourable for target species**

Table 6 continued.

Site name (number)	Target spp	Native species		Water quality			Aquatic habitat			Introduced predators/competitors				Assessment/ comments
		Target spp detect	Native spp	EC	DO	pH	Species composition (submerged)	Species composition (emergent)	Percent (%) physical habitat	Redfin perch (>120mm)	Juvenile common carp (<100mm)	Adult common carp (>250mm)	Eastern gambusia	
Holmes at Eastick Creek mouth (10)	YPP	NO	9	833	12.1	8.6	*Myriophyllum, Vallisneria*	*Typha, Phragmites, Schoenoplectus Juncus,* grasses	50	2	0	1	0	**Site appears favourable for target species**
Turvey's Drain (11)	SPP	NO	2	4850	0.5	7.3	*Ceratophyllum Myriophyllum,* algae	*Typha, Phragmites,* grasses	90	0	0	0	2	**Potentially unsuitable. Salinity elevated and dissolved oxygen below criteria**
Currency Creek Game Reserve (12)	YPP, MHH	NO	8	868	8.7	8.6	*Myriophyllum*	*Typha, Phragmites*	80	4	0	3	32	**Site appears favourable for target species**
Black Swamp (13)	YPP, SPP	NO	4	1230	12.0	8.0	n/a	*Typha*	50	3	0	0	0	**Potentially unsuitable. No submerged vegetation**
Mundoo Island Channel east (14)	MHH	NO	2	2816	-	7.9	*Ceratophyllum Myriophyllum,* algae	*Typha*	69	0	0	0	25	**Site appears favourable for target species**
Mundoo Island Channel east 2 (20)	SPP	NO	0	1094	1.1	7.1	*Ceratophyllum Myriophyllum*	*Typha,* grass	95	0	0	0	11	**Potentially unsuitable. Dissolved oxygen below criteria**
Mundoo Island Channel west (15)	SPP	NO	1	968	-	7.1	n/a	*Typha*	86	0	0	0	51	**Potentially unsuitable. No submerged vegetation**
Dunn's Lagoon (18)	MHH	NO	8	821	-	7.6	*Vallisneria, Myriophyllum*	*Schoenoplectus*	78	9 (total)	0		7	**Site potentially favourable for target species**
Old Clayton (22)	MHH	NO	8	780	-	8.1	n/a	Grass, *Triglochin*	46	22 (total)	1 (total)		11	**Site potentially favourable for target species**
Boggy Creek (17)	MHH	NO	5	1463	-	7.5	*Myriophyllum, Vallisneria*	*Phragmites, Typha, Ludwigia,* grass	80	1 (total)	31 (total)		3	**Site potentially favourable for target species**
Shadows Lagoon (19)	YPP	YES	8	1653	-	7.8	*Vallisneria*	*Typha, Phragmites*	38	6 (total)	16 (total)		4	**Total physical habitat below criteria, however, presence of target species suggests site remains favourable**

Table 7. Summary of site condition assessments conducted in autumn 2014. Cells coloured in green indicate criteria (Table 4) were met, whilst red cells indicate criteria were not met.

Site name (number)	Target spp	Native species		Water quality			Aquatic habitat			Introduced predators/competitors				Assessment/ comments
		Target spp detect	Native spp	EC	DO	pH	Species composition (submerged)	Species composition (emergent)	Percent (%) physical habitat	Redfin perch (>120mm)	Juvenile common carp (<100mm)	Adult common carp (>250mm)	Eastern gambusia	
Finniss at Winery Road (1)	SPSG	NO	2	2780	6.4	7.3	none	*Typha, Phragmites, Triglochin, Berula, Paspalum*	65	0	0	0	6	**Site appears favourable for target species**
Blue Lagoon (Pembroke) (2)	YPP	NO	8	2200	8.1	8.0	*Myriophyllum*	*Typha, Phragmites, Schoenoplectus*	40	12	0	1	0	**Potentially unsuitable. Total physical habitat below criteria**
Blue Lagoon (outer) (21)	YPP	NO	8	2200	5.9	7.6	*Myriophyllum*	*Typha, Phragmites, Schoenoplectus*	50	2	0	1	0	**Site appears favourable for target species**
Finniss River junction (3)	MHH, YPP	YES (MHH) NO (YPP)	8	921	7.7	8.2	*Myriophyllum, Vallisneria*	*Schoenoplectus, Typha, Phragmites*	60	2	0	2	1	**Site appears favourable for both target species**
Hunters Creek upstream rd (4)	YPP, SPP, MHH	NO	4	1180	3.4	7.1	*Myriophyllum, Ruppia*	*Typha, Bolboschoenus, Paspalum*	50	0	2	1	2	**Site appears favourable for both target species**
Hunters Creek downstream rd (5)	YPP, SPP, MHH	NO	8	1200	7.3	7.5	*Myriophyllum, Vallisneria, Ruppia*	*Typha, Bolboschoenus, Paspalum*	40	0	0	0	44	**Potentially unsuitable for YPP and SPP. Total physical habitat below criteria** **Site appears favourable for MHH**
Steamer Drain (9)	YPP, SPP	NO	3	879	7.4	7.4	*Myriophyllum, Ceratophyllum*, algae	*Typha, Bolboschoenus,* grasses	80	0	0	0	401	**Potentially suitable. Nonetheless, high abundance of gambusia**
Natural connected to Hunters Creek (8)	YPP, SPP	NO	3	2020	7.7	8.0	*Myriophyllum, Ceratophyllum*	*Typha, Bolboschoenus, Juncus,* grasses	85	0	3	0	437	**Potentially suitable. Nonetheless, high abundance of gambusia**

Table 7 continued.

Site name (number)	Target spp	Native species		Water quality			Aquatic habitat			Introduced predators/competitors				Assessment/ comments
		Target spp detect	Native spp	EC	DO	pH	Species composition (submerged)	Species composition (emergent)	Percent (%) physical habitat	Redfin perch (>120mm)	Juvenile common carp (<100mm)	Adult common carp (>250mm)	Eastern gambusia	
Holmes at Eastick Creek mouth (10)	YPP	NO	5	802	9.7	8.7	*Myriophyllum, Vallisneria*	*Typha, Phragmites, Schoenoplectus Paspalum*	40	6	0	3	4	**Potentially unsuitable. Total physical habitat below criteria**
Turvey's Drain (11)	SPP	NO	4	1060	4.6	7.3	*Ceratophyllum ,Myriophyllum,* algae	*Typha, Phragmites,* grasses	80	0	0	0	16	**Site appears favourable for target species**
Currency Creek Game Reserve (12)	MHH, YPP	YES (MHH) NO (YPP)	10	989	7.7	7.9	*Myriophyllum*	*Typha, Phragmites, Schoenoplectus*	60	1	0	3	86	**Site appears favourable for target species**
Black Swamp (13)	YPP, SPP	NO	3	1650	6.0	7.5	*Myriophyllum*	*Typha*	50	1	0	1	0	**Site appears favourable for target species. Nonetheless, submerged vegetation cover is minimal**
Mundoo Island Channel east (14)	MHH	YES	6	1677	-	7.0	*Ceratophyllum ,Myriophyllum,* algae	*Typha*	100	0	0	0	178	**Site appears favourable for target species**
Mundoo Island Channel east 2 (20)	SPP	NO	2	1080	8.5	7.1	*Ceratophyllum Myriophyllum*	*Typha,* grass	90	0	0	0	54	**Site appears favourable for target species**
Mundoo Island Channel west (15)	SPP	YES	2	1145	-	6.8	none	*Typha*	77	0	0	0	177	**Submerged vegetation lacking, but presence of target species suggests site is favourable**
Dunn's Lagoon (18)	MHH	NO	8	936	-	7.3	*Myriophyllum*	*Schoenoplectus*	92	3 (total)	0		16	**Site potentially favourable for target species**
Old Clayton (22)	MHH	YES	7	954	-	7.5	none	*Typha, Phragmites*	40	4 (total)	3 (total)		295	**Site potentially favourable for target species**
Boggy Creek (17)	MHH	NO	3	1314	-	7.0	*Ceratophyllum*	*Typha,*	100	0	0		3	**Site potentially favourable for target species**
Shadows Lagoon (19)	YPP	YES	4	1767	-	7.1	*Vallisneria*	*Typha, Phragmites*	58	1 (total)	9 (total)		105	**Site potentially favourable for target species**

3.3. Threatened species population status and reintroduction success

In 2013/14, all reintroduction sites were monitored (either directly through the current project or through *The Living Murray* condition monitoring by The University of Adelaide). For completeness, monitoring data from autumn 2012 to autumn 2013 (Bice *et al.* 2013) are also presented. Since reintroductions commenced in spring 2011, a total of 403 threatened fish, across all four target species, have been sampled from 11 sites in the Lower Lakes under the current project. Murray hardyhead have been sampled in the greatest numbers (*n* = 333) followed by southern pygmy perch (*n* = 43), Yarra pygmy perch (*n* = 14) and southern purple-spotted gudgeon (*n* = 13) (Table 8). Monitoring data is summarised for each species below.

Table 8. Summary of the number of individual threatened fish species sampled during post-reintroduction monitoring between 2012 and 2014. Data are presented for all reintroduction sites (including when threatened species were not detected) and sites where threatened species were sampled but reintroductions did not occur for that particular species (sites represented by *).

Species	Site name (number)	No. individuals sampled				
		Autumn 2012	**Spring 2012**	**Autumn 2013**	**Spring 2013**	**Autumn 2014**
Yarra pygmy perch	Blue Lagoon (2 & 21 combined)	0	0	0	0	0
	Finniss River Junction (3)	0	0	0	0	0
	Steamer Drain (9)	0	2	0	0	0
	Shadows lagoon (19)	0	8	2	1	1
	Hunters Creek d/s road (5)	0	0	0	0	0
	Total	**0**	**10**	**2**	**1**	**1**
Southern pygmy perch	Natural channel connected to Hunters Creek (8)	11	4	4	5	0
	Turvey's Drain (11)	1	0	0	0	0
	Mundoo Island Channel east 2 (20)	0	1	0	0	0
	Mundoo Island Channel east* (14)	0	2	0	0	0
	Mundoo Island Channel west* (15)	0	0	1	0	14
	Total	**12**	**7**	**5**	**5**	**14**
Murray hardyhead	Mundoo Island Channel east (14)	0	4	9	0	1
	Finniss Junction* (3)	12	7	42	47	173
	Hunters Creek d/s road (5)	0	0	0	0	0
	Dunn's Lagoon* (18)	0	0	7	0	0
	Old Clayton* (22)	0	0	7	0	22
	Currency Creek Game Reserve* (12)	0	0	0	0	2
	Total	**12**	**11**	**65**	**47**	**198**
Southern purple-spotted gudgeon	Finniss at Winery Rd (1)	3	1	3	6	0
	Total	**3**	**1**	**3**	**6**	**0**

Yarra pygmy perch

Yarra pygmy perch (~5,850) were released across five sites over the four rounds of reintroductions (Table 3) and have been detected in subsequent monitoring at two of these sites (Table 8). Initially in spring 2011, 800 and 400 individuals were released at the Finniss River Junction and Blue Lagoon sites (includes Blue Lagoon 1 and 2), but post reintroduction monitoring has failed to detect the species at these sites. Both sites have recovered considerably since the return of favourable water levels, with the Finniss River Junction site in particular characterised by extensive beds of submerged vegetation (*Myriophyllum* spp.), and in 2013/14 both sites were considered favourable for Yarra pygmy perch based on the reintroduction site condition assessment criteria (Tables 6 and 7). Nevertheless, the large area of both sites may result in low sampling efficiency and subsequent non-detection of the species. Approximately 700 fish were released at Hunters Creek downstream Denver Road across two reintroduction events (400 fish in spring 2013; 300 fish in autumn 2013), but subsequent monitoring has also failed to detect Yarra pygmy perch at this site.

In 2012, approximately 2,200 and 1,750 fish were released at Steamer Drain and Shadows Lagoon, respectively. Subsequent monitoring of Streamer Drain yielded two individuals in spring 2012 (the first Yarra pygmy perch sampled in the MDB since 2007; Figure 2); however, no fish have been detected since (i.e. summer/autumn 2013, spring 2013 and autumn 2014). In contrast, low numbers (i.e 1–8) of fish have been consistently sampled at Shadows Lagoon since the first release at this site in autumn 2012 (Table 8).

Figure 2. Yarra pygmy perch sampled from the Streamer Drain during spring 2012 – the first individual sampled from the CLLMM region (and MDB) since 2007.

The Yarra pygmy perch sampled from Steamer Drain and Shadows Lagoon in spring 2012, ranged 42–55 mm TL and all but one individual exhibited fluorescence readings consistent with a calcein mark (Figure 3a and b), suggesting they were recaptures from the previous releases. The two individuals sampled from Shadows Lagoon in autumn 2013 were <40 mm TL and exhibited fluorescence readings below that consistent of a calcein mark (Figure 3c and d). Based on length and fluorescence, these individuals were likely young-of-year (YOY) recruited in the wild following the previous spawning season. The one individual sampled in spring 2013 was 63 mm TL (Figure 3e) and exhibited fluorescence consistent with a calcein mark (Figure 3f), indicating this individual was a recaptured fish from the release in either autumn or spring 2012, and wild survival of >12 months. Another individual was sampled in autumn 2014, measuring 50 mm TL and exhibiting fluorescence inconsistent with a calcein mark suggesting this fish was recruited in the wild.

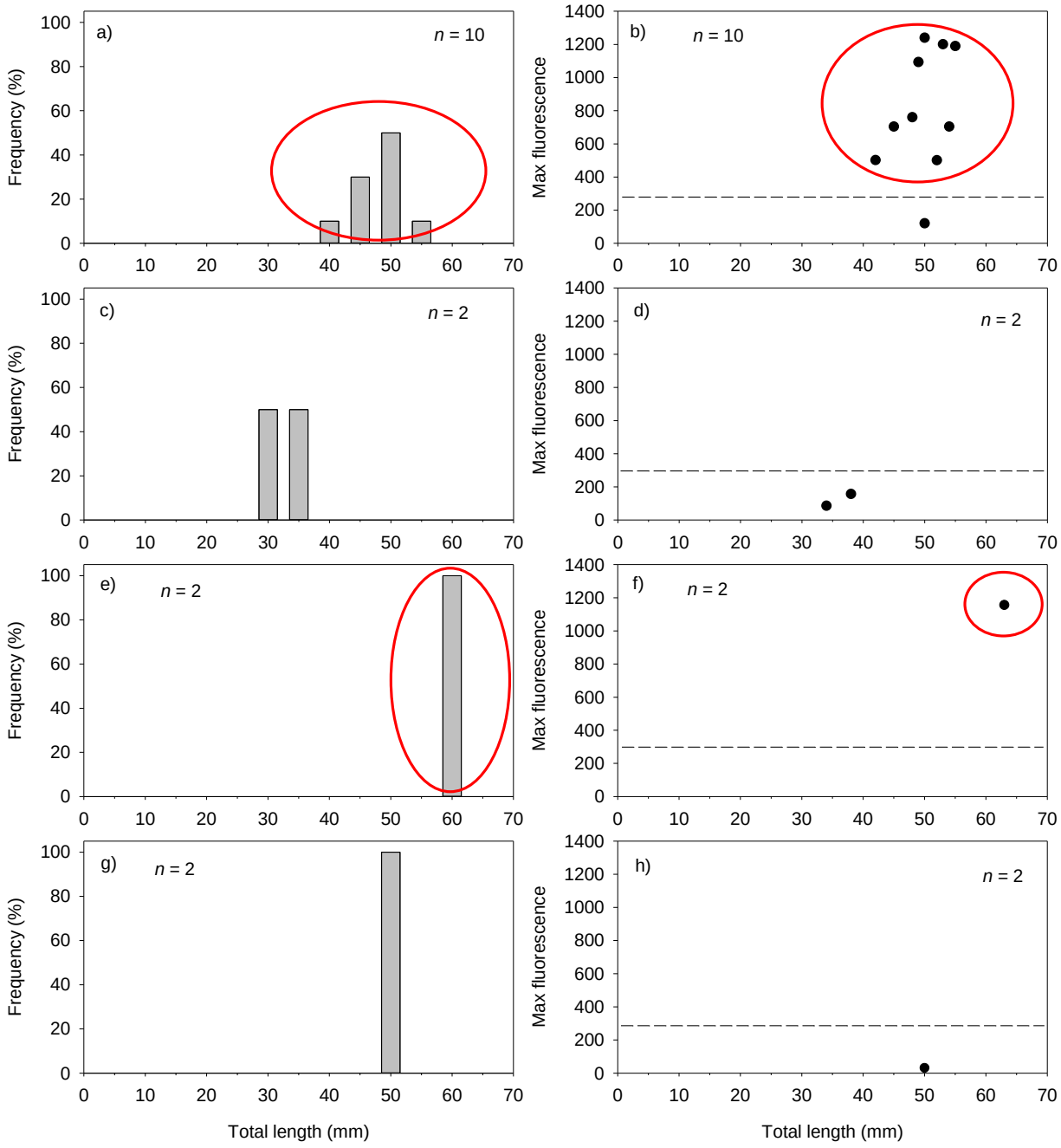

Figure 3. Length-frequency distributions and corresponding max fluorescence (sample fluorescence ratio) against length plots for all Yarra pygmy perch sampled in spring 2012 (a & b), autumn 2013 (c & d), spring 2013 (e & f) and autumn 2014 (g & h). Dashed line represents the fluorescence reading deemed to indicate a calcein mark and red ellipses represent likely recaptured released fish.

Southern pygmy perch

A total of ~1,350 southern pygmy perch were released across three sites in 2011/12 and subsequent monitoring has detected fish at all three sites, as well as at two adjacent sites (Table 8). In spring 2011, ~300 and ~770 southern pygmy perch were released into Turvey's Drain and the 'natural channel connected to Hunters Creek' sites, respectively. A single southern pygmy perch was sampled at Turvey's Drain in autumn 2012, but the following four monitoring rounds have failed to detect any individuals. Southern pygmy perch were detected at the 'natural channel connected to Hunters Creek' site in autumn 2012 (n = 11), spring 2012 (n = 4), autumn 2013 (n = 4) and spring 2013 (n = 5; Figure 4), but were not detected in autumn 2014.

In autumn 2012, ~280 southern pygmy perch were released at the Mundoo Channel east 2 site, with a single fish sampled in spring 2012. Another individual was sampled during the same monitoring round at an adjacent, hydrologically connected site at Mundoo Island Channel east. No southern pygmy perch have since been detected at these two sites, but one fish was sampled in autumn 2013 and 14 sampled in autumn 2014 from another adjacent and hydrologically connected site (Mundoo Island Channel west), suggesting small-scale dispersal from the original release site.

In autumn 2012, southern pygmy perch ranged 32–53 mm TL, with all but one fish ranging 44–53 mm TL (Figure 5a). The larger individuals all exhibited fluorescence consistent with a calcein mark, suggesting they were recaptures from the previous reintroduction; the remaining individual was 32 mm TL and exhibited low fluorescence suggesting it was a likely YOY recruited in the wild following the previous spawning season (2011) (Figure 5a and b). In spring 2012 a cohort of large fish was present (46–56 mm TL), with fluorescence readings consistent with a calcein mark (Figure 5c and d). All of these fish were sampled from the Mundoo Island Channel group of sites and were likely recaptures from the autumn 2012 release at Mundoo Island Channel east 2. A cohort of smaller fish (18–19 mm TL) was sampled at the 'natural channel connected to Hunters Creek' site and exhibited low levels of fluorescence indicating that they were recently 'wild recruited' YOY (Figure 5c and d). Of the five fish sampled in autumn 2013, one was from the Mundoo Island Channel west site, and based upon length (40 mm TL) and low fluorescence, was likely a wild recruited fish from the previous spawning season (2012) (Figure 5e and f). The remaining fish (n = 4) sampled in autumn 2013, were captured from the 'natural channel connected to Hunters Creek' site during ad-hoc sampling.

These fish ranged 46–52 mm TL (Figure 4e), but were not assessed for fluorescence. A single fish was sampled from the 'natural channel connected to Hunters Creek' in spring 2013, measuring 23 mm TL (Figure 5g), whilst >90% of fish sampled from Mundoo Channel west in autumn 2014 were <40 mm TL (Figure 5i). The length of these fish and low fluorescence values (Figure 5g-j), indicate these fish were new recruits from spawning in 2013 and likely represent progeny of fish released at Mundoo East 2.

a) b)

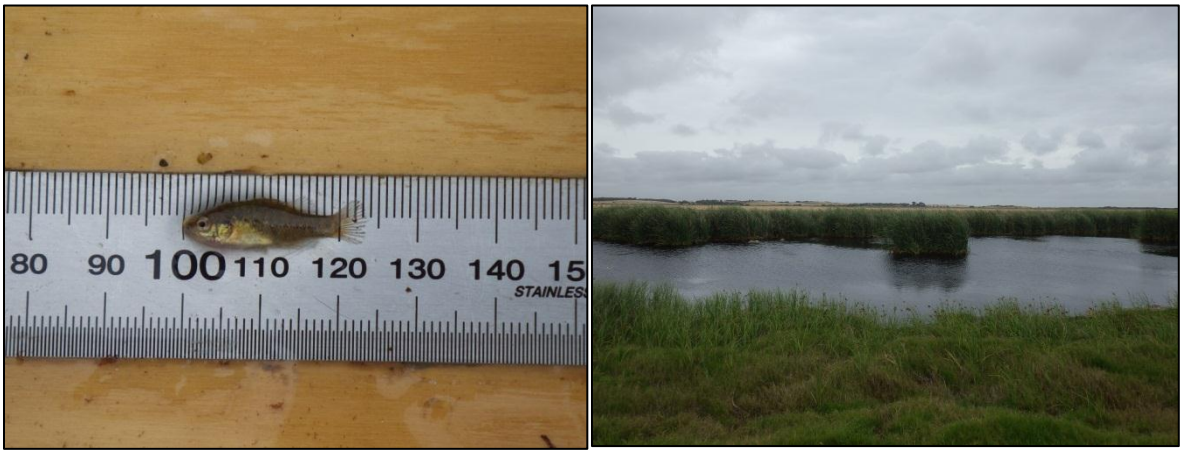

Figure 4. a) Young-of-year southern pygmy perch sampled from b) the 'natural channel connected to Hunters Creek' site in spring 2013.

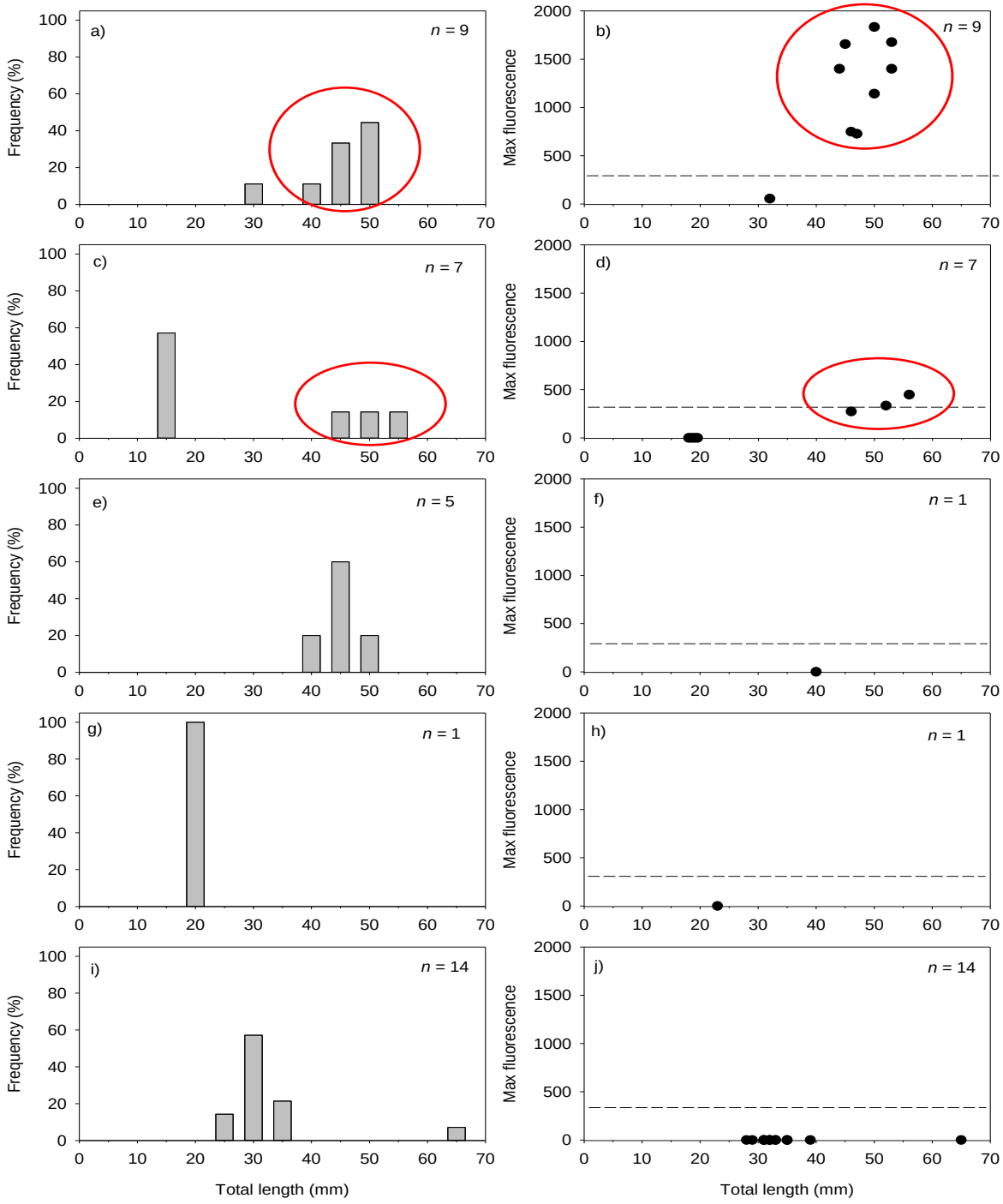

Figure 5. Length-frequency distributions and corresponding maximum fluorescence (sample fluorescence ratio) against length plots for all southern pygmy perch sampled in autumn 2012 (a & b), spring 2012 (c & d), autumn 2013 (e & f), spring 2013 (g & h) and autumn 2014 (I & j). Dashed line represents the fluorescence reading deemed to indicate a calcein mark and red ellipses represent likely recaptured released fish.

Murray hardyhead

During autumn and spring 2012, >7,500 Murray hardyhead were released at two relatively contained sites on Hindmarsh (i.e. Hunters Creek downstream Denver Road) and Mundoo Islands (Mundoo Island Channel east) (Table 3). No individuals have subsequently been sampled at Hunters Creek downstream of Denver Road, but low numbers have been sampled at Mundoo Island Channel east in spring 2012 (*n* = 4), autumn 2013 (*n* = 9) and autumn 2014 (*n* = 1). Additionally low-high numbers of Murray hardyhead have been sampled at four non-reintroduction sites since autumn 2012 (Table 8). Murray hardyhead have been consistently sampled at the Finniss River Junction since autumn 2012 and have exhibited a gradual increase in abundance with 173 individuals sampled in autumn 2014 (Figure 6a & b). Low numbers (*n* < 10) were sampled at Dunn's Lagoon and Old Clayton in autumn 2013, with a further 22 fish sampled at Old Clayton in autumn 2013. Additionally, two Murray hardyhead were sampled at the Currency Creek game reserve in autumn 2014, the first record of the species at the site since spring 2010.

a) b)

Figure 6. a) Murray hardyhead sampled from the b) Finniss River Junction site in autumn 2014.

In autumn 2012, Murray hardyhead ranged 22–40 mm FL (Figure 7a) and represent remnant wild fish, all being sampled from the Finniss Junction site prior to any reintroductions as part of this project. In spring 2012, sampled Murray hardyhead were larger, ranging 38–58 mm FL (Figure 7b) and represent a mixture of fish from both the reintroduction site at Mundoo Channel east and fish sampled from the Finniss River Junction. In autumn 2013, Murray hardyhead

ranged 20–63 mm FL, but approximately 65% were <35 mm FL, likely representing wild recruited YOY (Figure 7c). Progression of this cohort was evident in spring 2013, with all fish sampled >35 mm FL (Figure 7d). Murray hardyhead sampled in autumn 2014 ranged 20–57 mm FL, but >80% were <40 mm FL (Figure 7e) and likely represent newly wild recruited individuals from spawning in 2013/14.

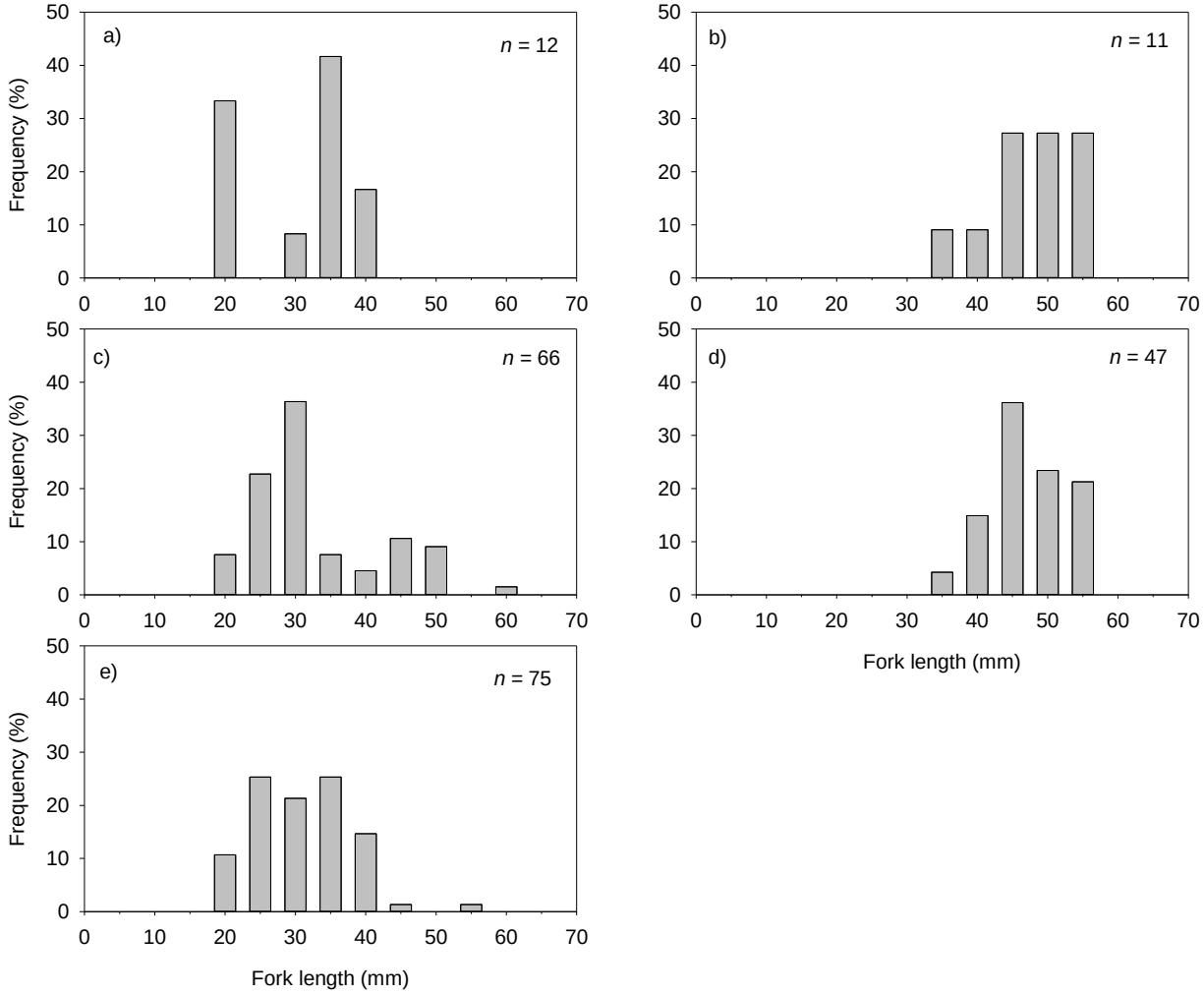

Figure 7. Length-frequency distributions for Murray hardyhead sampled in a) autumn 2012, b) spring 2012, c) autumn 2013, d) spring 2013 and e) autumn 2014.

Southern purple-spotted gudgeon

A total of 1,120 southern purple-spotted gudgeon have been reintroduced into the lower Finniss River (Winery Road) site across four events (Table 3) and low numbers ($n \leq 6$) have been

sampled during all subsequent monitoring events (Figure 8) with the exception of autumn 2014 (Table 8). In autumn 2012, three individuals (64–72 mm TL; Figure 9a) were collected and based on length were likely all recaptured fish from the previous release. The one individual sampled in spring 2012 measured 70 mm TL and also likely represented a recaptured reintroduced fish (Figure 9b). Three individuals were sampled in autumn 2013, ranging 43–70 mm TL (Figure 9c) and six individuals were sampled in spring 2013 ranging 56–99 mm TL (Figure 9d). Smaller individuals (i.e. <60 mm TL) sampled in both autumn 2013 and spring 2013, may represent wild recruited individuals, but differentiating 'released' and wild recruited fish, at this point in time, based upon length alone is not possible due to the multiple releases that occurred at this site. Short-term survival (6–18 months) of southern purple-spotted gudgeon was clearly evident at the lower Finniss River site, but the current status of the species at the site is uncertain given no fish were detected in autumn 2014.

Figure 8. Large southern purple-spotted gudgeon (99 mm TL) sampled from the lower Finniss River (Winery Rd) in spring 2013.

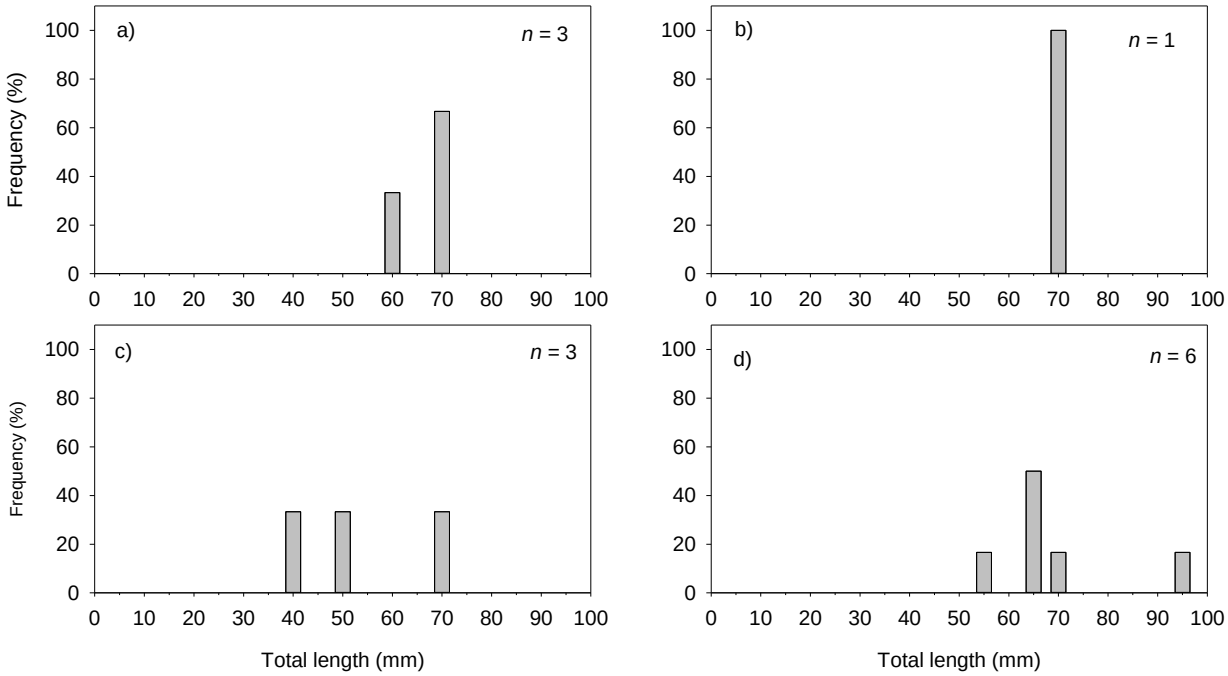

Figure 9. Length-frequency distributions for southern purple-spotted gudgeon sampled in a) autumn 2012, b) spring 2012, c) autumn 2013 and d) spring 2013.

4. DISCUSSION

Unprecedented drought and continued over-abstraction of water from the MDB over the period 2007–2010 placed the CLLMM region on the point of ecological collapse (Kingsford *et al.* 2011, Wedderburn *et al.* 2012). Over this period, water level recession in the Lower Lakes led to habitat fragmentation, broad-scale loss of submerged vegetation and elevated salinities, in turn resulting in significant declines in threatened fish populations (Wedderburn *et al.* 2012). Numerous urgent conservation actions, including fish rescue and captive maintenance/breeding, were required to avert the local extinction of Yarra pygmy perch, southern pygmy perch, Murray hardyhead and southern purple-spotted gudgeon (Hammer *et al.* 2013), and ensured a supply of individuals for reintroduction upon the return of favourable conditions.

Following widespread, above average rainfall in the MDB in 2010/11 and significant inflows, the Lower Lakes returned to typical water levels (~0.75 m AHD) and there has been a gradual recovery of submerged vegetation (Gehrig *et al.* 2012), preferred habitat of the aforementioned species. Over four reintroduction events between 2011 and 2013, ~15,840 fish, across the four species, were released at ten suitable locations in the region. Integral to any reintroduction program is the monitoring of reintroduced populations and subsequent assessment of the success of reintroduction activities (Fischer and Lindenmayer 2000). Monitoring from 2012–2014, has detected low numbers of all four species and evidence of wild recruitment in Yarra pygmy perch, southern pygmy perch and Murray hardyhead. In light of the results of this monitoring, the current status of each threatened fish species, evaluation of success of the CFH project and a way forward for threatened fish management in the region are discussed.

4.1. Current species and site condition status

As of autumn 2014, two and a half years after the first reintroductions of threatened species in the CLLMM region, the current status of Yarra pygmy perch, southern pygmy perch, Murray hardyhead and southern purple-spotted gudgeon differ markedly. Each species was detected in in the CLLMM region in 2013−14 and without conservation actions undertaken as part of the DAP, during the period of critical water shortage (see Hammer *et al.* 2013), and the reintroductions as part of the current project, this outcome may not have been achieved. Nonetheless, whilst Murray hardyhead were sampled in low-moderate abundance from a

number of sites, the current status of Yarra pygmy perch, southern pygmy perch and southern purple-spotted gudgeon remain cause for concern.

Substantial sampling effort over the period 2008–2011 (Bice *et al.* 2009, Wedderburn and Barnes 2009, Bice *et al.* 2010, Wedderburn and Hillyard 2010, Bice *et al.* 2011, Bice and Zampatti 2011, Wedderburn and Barnes 2011, 2012, Whiterod and Hammer 2014) failed to detect any Yarra pygmy perch in the CLLMM region, suggesting the species was extirpated from the wild, and that its short- and long-term future in the region was entirely dependent upon the current reintroduction program. Reintroductions occurred at five sites over 2011–2013, but the species has not yet been detected at three (Blue Lagoon, Finniss River Junction and Hunters Creek downstream Denver Rd) of these sites during post reintroduction monitoring, suggesting reintroductions at these sites may have failed. Additionally, fish (n = 2) were recaptured at the Steamer Drain site six months post reintroduction, in spring 2012, but have not been detected since. In contrast, Yarra pygmy perch have been consistently sampled in low numbers ($n \leq 8$) from Shadows Lagoon since reintroductions, and there has been evidence of 'wild recruitment'. Nonetheless, only one individual has been sampled in each of the last two monitoring rounds. Thus, as of autumn 2014, this species is confirmed from the region at just one site, in very low abundance, and therefore remains at high risk of extirpation. Ironically, site condition assessments suggest that Shadows Lagoon and another six sites are currently suitable for habitation (facilitated by dispersal and recolonisation from other sites or reintroductions) by Yarra pygmy perch. Whilst the criteria developed to determine site suitability (Bice *et al.* 2012) are not infallible, they likely provide a reasonably accurate guide. For a population to establish, a critical number of progeny must be produced by reintroduced adults such that, when taking into account mortality (e.g. predation), enough progeny recruit to the reproductively mature population which in turn, perpetuate the population. Given the likely high mortality rate of reintroduced fish (e.g. post-release predation, starvation), it is possible that the number of Yarra pygmy perch released at reintroduction sites has been too low to allow population establishment, although conditions appeared favourable at the time of release. Encouragingly, favorable conditions at a number of sites remain promising should the prospect of further reintroductions be entertained, and greater post-release monitoring (e.g. prey abundance, gut content of resident predatory species) could help to resolve fate of reintroduced individuals. Importantly, sources of Yarra pygmy perch remain abundant in surrogate refuges and present an opportunity to continue reintroductions.

Southern pygmy perch underwent similar declines to Yarra pygmy perch over the period 2007–2011 (Bice *et al.* 2011, Wedderburn *et al.* 2012) and the species presence in the region is likely dependent upon the reintroductions undertaken by the current project, or the unlikely

downstream dispersal from eastern Mount Lofty Ranges populations (e.g. Tookayerta Creek) (see Whiterod and Hammer 2014). Southern pygmy perch have been released at three sites and subsequently been detected at least once at each of these sites as well as two adjacent sites. Nonetheless, no fish have been detected at Turvey's Drain since autumn 2012 and thus, reintroduction at this site appears to have been unsuccessful. Initial reintroductions at the 'natural channel connected to Hunters Creek' site met with success, with individuals sampled, and recruitment evident, in the four subsequent monitoring rounds (autumn 2012, spring 2012, autumn 2013 and spring 2013). Nonetheless, abundance appeared to be declining at this site and no fish were detected in autumn 2014. This site remains in favourable condition and thus, the species may remain in very low abundance. The remaining detections of southern pygmy perch, post-reintroduction, have come from a series of three adjacent and hydrologically connected sites on Mundoo Island. Numbers of fish have typically been low ($n \leq 2$), but 14 individuals were sampled at Mundoo Island Channel west in autumn 2014, the majority of which were newly recruited individuals (<40 mm TL), likely spawned the previous (i.e. 2013) spawning season. Whilst this result is promising for persistence of the species at the site, this site represents the only confirmed location of the species in the region as of autumn 2014. As per Yarra pygmy perch, a number of sites were deemed favourable for habitation by this species; nevertheless, no surrogate refuge or captive population exists for this species and thus, an expansion of current distribution is reliant on natural dispersal from the Mundoo Island sites.

A total of four releases of southern purple-spotted gudgeon have occurred at the Finniss River at Winery Road since spring 2011 and low numbers (n = 1–6) have typically been detected during post reintroduction monitoring, indicating wild survival of 6–24 months. Several individuals have been sampled that, based on length, could be wild recruited individuals; however, given multiple releases were undertaken and the inconsistency in calcein mark retention in this species (Bice *et al.* 2013), differentiation of recaptured and wild recruited fish was not possible. The capture of small fish (<60 mm TL) in any future monitoring, should no further reintroductions occur, would likely represent evidence of wild recruitment. Sampling in spring 2013 yielded the greatest number of southern purple-spotted gudgeon sampled at this site to date, but was followed by a failure to detect any fish in autumn 2014, despite the site meeting suitability criteria. At this site, the species was released across a reach of stream ~300 m long and recaptures have consistently occurred from a particular ~50 m reach (additional monitoring has not detected the species at nearby sites upstream or downstream; Whiterod and Hammer 2014). In autumn 2014, this reach had become significantly degraded due to stock access, which may have contributed to the failure to detect any fish (Figure 10). This represents an issue for future management of the site. Whilst not detected in autumn 2014, this species is

difficult to sample due to its sedentary nature and use of highly complex habitats and thus, may remain at the site in low numbers.

a) b)

Figure 10. a) Stock access and b) associated damage in the lower Finniss River (Winery Rd) in autumn 2014.

In autumn 2014, Murray hardyhead were sampled in the greatest numbers in the Lower Lakes since 2009 (Wedderburn and Hillyard 2010, Bice *et al.* 2011). The species was detected at four sites during sampling conducted for the current project, whilst it was detected in low numbers at a further nine sites during a broader fish survey conducted by The University of Adelaide, particularly concentrated in the Currency Creek-Goolwa Channel area (Wedderburn 2014). Furthermore, length-frequency analysis suggests wild recruitment has occurred regularly over the past two years. Whilst Murray hardyhead exhibited declines, sporadic captures of individuals were recorded over the period 2007–2010 (Bice *et al.* 2011, Wedderburn and Barnes 2011) suggesting a low number of individuals remained in wild habitats. Given the species high mobility and tolerance to elevated salinity (Wedderburn *et al.* 2008), Murray hardyhead were potentially more resilient to the prevailing drought conditions than both pygmy perch species' through 2007–2010. Murray hardyhead were detected at the Finniss River Junction site prior to any reintroductions (autumn 2012) and have been detected, in increasing abundance, at this site in all subsequent monitoring rounds. Additionally low-moderate (*n* = 2–22) numbers of Murray hardyhead have been sampled at several nearby sites (i.e. Dunn's Lagoon, Old Clayton, Currency Creek Game Reserve), and it is hypothesised that these fish are progeny of remnant wild fish. Under the current project, Murray hardyhead were released at Hunters Creek (downstream Denver Road) and at Mundoo Island Channel east. No fish have since been detected at Hunters Creek, but low numbers (*n* = 1–9) have been consistently sampled at

Mundoo Island Channel east suggesting likely survival and recruitment of reintroduced fish at this site. In general, the current status of this species is encouraging, with substantially increased abundance and distribution. A number of further sites appear to be suitable for habitation and given the high mobility of this species, natural expansions in distribution and abundance may occur. The species remains absent from the Lake Albert region, however, despite targeted sampling (Wedderburn 2014), so reintroductions to this area may be necessary given the distance from identified populations. The species remains in high numbers in one surrogate location, which could be used for any reintroductions into habitats within Lake Albert.

4.2. Evaluating success of the CFH Project and future conservation of threatened species in the CLLMM

Yarra pygmy perch, southern pygmy perch and southern purple-spotted gudgeon were likely all absent from the CLLMM region prior to the commencement of the current project and from this viewpoint, consistent detection of low numbers of these species in the region since spring 2011 is a successful outcome of the CFH project. Nonetheless, against an objective of 'establishing self-sustaining wild populations', the project to-date has likely been unsuccessful. As of autumn 2014, the abundance of Yarra pygmy perch is in decline (since spring 2012), and no increase in distribution (i.e. number of sites where detected) has been observed. Similarly, whilst southern pygmy perch were sampled in their greatest numbers since release in autumn 2014, abundance was not substantially greater than abundance in autumn 2012, and there has been no evidence of increasing distribution. Whilst some level of recruitment has been observed for both species, the persistence of both species remains precarious. Failure to detect any southern purple-spotted gudgeon in autumn 2014 also casts doubt over the success of reintroductions for this species.

Increased abundance and distribution of Murray hardyhead suggests this species is recovering following population declines, but recovery appears to have largely been driven by remnant wild fish rather than reintroduced individuals. Nonetheless, given the high mobility of this species and issues in relation to the use of calcein to differentiate 'stocked' and 'wild' individuals, it is possible that reintroduced fish contributed to broader increases in the abundance of this species. At the least, reintroductions likely expanded the distribution of this species and thus, could be viewed as successful at this stage.

When viewed in light of other reintroduction programs for threatened fishes both in Australia (Lyon *et al.* 2012) and internationally (Shute *et al.* 2005, Rakes and Shute 2006, Bezold 2007,

George *et al.* 2009), reintroduction effort expended in the current project has been comparatively minimal. Reintroductions of European sturgeon (*Acipenser sturio*) and lake sturgeon (*Acipeser fulvescens*) in Germany and North America, respectively, have involved the reintroduction of tens of thousands of individuals over multiple years (Bezold 2007). Reintroductions of small-bodied threatened species, such as darters (Percidae) and madtoms (Ictaluridae), in the south-eastern United States have released similar numbers of fish to the current project, but reintroduction programs have occurred over periods of up to 20 years (George *et al.* 2009). Re-establishment of yellowfin madtom (*Noturus flavipinnis*), smokey madtom (*Noturus baileyi*) and Citico darter (*Etheostoma sitikuense*) into Abrams Creek, Tennessee (Shute *et al.* 2005) is often viewed as a success story for threatened fish reintroductions; nevertheless, it took five years before there were any recaptures of reintroduced fish and ten years before wild recruitment was observed (George *et al.* 2009). Lyon *et al.* (2012) suggest the long-term (10 years) stocking program for trout cod (*Maccullochella macquariensis*) in the Ovens River, in the MDB, provided the opportunity for some cohorts to encounter favourable conditions for survival, whilst fish stocked in other years did not survive and contribute to future populations. Such projects suggest further reintroductions of the target species in the current project are likely required to meet the objective of re-establishing self-sustaining wild populations. Indeed, common to all the aforementioned programs is the annual release of small-medium numbers of individuals over multiple years, which may have resulted in the success of these programs.

The sheer size of Lake Alexandrina (>600 km^2) also warrants consideration in regards to the length of this reintroduction program and the number of fish released. Other reintroduction programs for imperiled small-bodied freshwater fish have typically released fish into relatively small (<20 km^2) streams (Shute *et al.* 2005, George *et al.* 2009) or lakes (Ozer and Ashley 2013). Such environments may be more conducive to establishing self-sustaining populations due to a lower risk of ‘diluting’ individuals and provide a greater capacity to determine success through monitoring due to greater sampling efficiency. As such, reintroductions in large lakes may require greater numbers of fish to be released over a longer period to facilitate population establishment and enhancement to levels which are better detectable through monitoring. Furthermore, greater sampling effort is arguably required to detect individuals in such environments.

Site condition assessment in 2013/14 suggests several sites in the CLLMM region are now suitable for habitation (either facilitated through dispersal from existing populations or by reintroduction) by each of the target species. The source of fish for such programs is often most

problematic (i.e. not available, considered too expensive to produce), yet low-maintenance surrogate populations exist for three of the four species. In the case of Murray hardyhead, further reintroductions are likely not necessary to Lake Alexandrina (but possibly Lake Albert); rather natural colonisation of these sites via dispersal from existing locations is likely. Sources of Yarra pygmy perch remain abundant in two surrogate refuges, whilst surrogate refuges have recently been established for southern purple-spotted gudgeon. These refuges present an opportunity to continue ongoing reintroductions of small numbers of individuals on an annual basis. No surrogate refuges were established for southern pygmy perch.

5. CONCLUSION

Under the CFH project, >15,000 fish, across four threatened species, namely Yarra pygmy perch, southern pygmy perch, Murray hardyhead and southern purple-spotted gudgeon, have been reintroduced to the Lower Lakes since spring 2011. Each species has been detected in the CLLMM region multiple times since reintroduction, with evidence of wild recruitment for some species. With the exception of Murray hardyhead, these outcomes are likely a direct result of the current project. Nonetheless as of autumn 2014, with the exception of Murray hardyhead, the abundance of these species remains very low and their distributions are highly restricted. As such, evidence suggests that reintroductions of Yarra pygmy perch, southern pygmy perch and southern purple-spotted gudgeon have likely been unsuccessful in facilitating the re-establishment of self-sustaining wild populations. Future reintroductions, where possible, are likely required to ensure the long-term future of these species. Ongoing monitoring of threatened fish populations (both reintroduced and remnant populations) throughout the CLLMM region remains integral, to monitor threatened species population status and provide further specific assessment of the success of reintroductions.

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