

Stocky Galaxias – monitoring plan, Snowy 2.0

T.A. Raadik and M. Lintermans

December 2022



Arthur Rylah Institute for Environmental Research Published Client Report





Environment, Land, Water and Planning

Acknowledgment

We acknowledge and respect Victorian Traditional Owners as the original custodians of Victoria's land and waters, their unique ability to care for Country and deep spiritual connection to it. We honour Elders past and present whose knowledge and wisdom has ensured the continuation of culture and traditional practices.

We are committed to genuinely partner, and meaningfully engage, with Victoria's Traditional Owners and Aboriginal communities to support the protection of Country, the maintenance of spiritual and cultural practices and their broader aspirations in the 21st century and beyond.



Arthur Rylah Institute for Environmental Research Department of Environment, Land, Water and Planning PO Box 137 Heidelberg, Victoria 3084 Phone (03) 9450 8600 Website: www.ari.vic.gov.au

Citation: Raadik, T.A. and Lintermans, M. (2022). Stocky Galaxias - monitoring plan, Snowy 2.0. Published client report for Snowy Hydro Ltd, Cooma. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg, Victoria.

Front cover photo: (clockwise from top) Murrumbidgee River at junction with Tantangara Creek; Macquarie Perch; alpine plain in snow; Stocky Galaxias (Images: Tarmo A. Raadik).

© The State of Victoria Department of Environment, Land, Water and Planning 2022



This work is licensed under a Creative Commons Attribution 3.0 Australia licence. You are free to re-use the work under that licence, on the condition that you credit the State of Victoria as author. The licence does not apply to any images, photographs or branding, including the Victorian Coat of Arms, the Victorian Government logo, the Department of Environment, Land, Water and Planning logo and the Arthur Rylah Institute logo. To view a copy of this licence, visit http://creativecommons.org/licenses/by/3.0/au/deed.en

Edited by David Meagher, Zymurgy SPS.

ISBN 978-1-76136-265-1 (pdf/online/MS word)

Disclaimer

This publication may be of assistance to you but the State of Victoria and its employees do not guarantee that the publication is without flaw of any kind or is wholly appropriate for your particular purposes and therefore disclaims all liability for any error, loss or other consequence which may arise from you relying on any information in this publication.

Accessibility

If you would like to receive this publication in an alternative format, please telephone the DELWP Customer Service Centre on 136 186, email customer.service@delwp.vic.gov.au or contact us via the National Relay Service on 133 677 or www.relayservice.com.au. This document is also available on the internet at www.delwp.vic.gov.au

Arthur Rylah Institute for Environmental Research Department of Environment, Land, Water and Planning Heidelberg, Victoria

Stocky Galaxias – monitoring plan, Snowy 2.0

Tarmo A. Raadik¹ and Mark Lintermans²

¹Arthur Rylah Institute for Environmental Research 123 Brown Street, Heidelberg, Victoria 3084

² Threatened Fish Services: PO Box 111, Belconnen ACT 2616

Caveat: This report was completed in August 2021 and consequently does not contain more recent information which may have become available.

Arthur Rylah Institute for Environmental Research Published Client Report for Snowy Hydro Ltd, Cooma, Department of Environment, Land, Water and Planning

Arthur Rylah Institute for Environmental Research Department of Environment, Land, Water and Planning Heidelberg, Victoria

Acknowledgements

We wish to thank Hugh Allan (University of Canberra) for the provision of valuable information on recent activities related to Stocky Galaxias. Daniel Stoessel (DELWP-ARI), Lizzie Pope (Snowy Hydro) and Lachlan Barnes (SLR), are thanked for providing comments on earlier drafts of this document. Lindy Lumsden (ARI) provided invaluable additional comments on the final draft of this document.

Contents

Ackn	owledg	ements	ii			
1	Introdu		1			
1.1	Relevance to priority conservation actions					
2	Monito	ring plan aim and objectives	3			
2.1	Design	considerations	3			
3	Monito	ring activities	6			
3.1	Routine	e surveillance monitoring	6			
	3.1.1	Population monitoring	6			
	3.1.2	Population genetics	6			
	3.1.3	Predator surveillance	6			
	3.1.4 Monitoring methods and level of effort					
	3.1.5	Life-history parameters measured	11			
	3.1.6	Population genetics	12			
	3.1.7	Environmental variables	12			
	3.1.8	Threat monitoring	13			
	3.1.9	Monitoring sites	13			
4 moni	Trigge toring p	r Action Response Plan (TARP) criteria associated with potential results of the Ian	15			
4.1	Trigger	-based sampling	15			
	4.1.1	Spawning assessment	15			
	4.1.2	Egg hatching success	17			
	4.1.3	Predator surveillance (TARP Triggered)	17			
	4.1.4	Fish Condition Assessment	18			
5	Refere	nces	19			

Tables

Table 1. Population and species parameters important in monitoring Stocky Galaxias	4
Table 2. Summary of recommended monitoring activities as grouped into routine surveillance or trigger- based monitoring	8
Table 3. Proposed monitoring sites on Tantangara Creek, including access, land tenure, availability of previous comparable data., and stream dimensions	14

Table 4. Criteria, trigger levels, and suggested response activities, for each alert level for Stocky Galaxias 16

Figures

•

Figure 1 Map of proposed monitorin	g sites on Tantangara Creek1	4
I igule 1. Mup of proposed monitorin	g sites on runtungulu oreek	-

1 Introduction

Snowy Hydro Limited received approval in 2020 to construct a new large-scale pumped hydro-electric storage and generation scheme (Snowy 2.0), to increase hydro-electric capacity within the existing Snowy Mountains Hydro-electric Scheme. This will involve the connection of the existing Talbingo and Tantangara reservoirs via a series of underground pipes and an underground power generation station. Water will be transferred in both directions between the reservoirs, which are in separate river catchments.

The Arthur Rylah Institute for Environmental Research has been engaged by Snowy Hydro to provide specialist advice that can inform the selection of options and preparation of various aquatic Management Plans required as part of the NSW and Commonwealth approvals for the Snowy 2.0 project.

This report focusses on the development of a monitoring plan for the native Stocky Galaxias (*Galaxias tantangara*). It outlines a process of monitoring relevant to the priority conservation actions for the species and includes objectives and potential activities aimed at understanding population variability and trajectory to inform population management. As such, its value and relevance will extend beyond the Snowy 2.0 Management Plans.

Monitoring is an essential primary step in species conservation, which can also provide an early indication of rapid population decline, triggering crucial management interventions. However, there are very few threatened fish monitoring programs occurring in Australia, and many of these are compromised by the lack of longevity, design quality, lack of demographic parameters, and poor data availability and reporting (Lintermans and Robinson 2018). Program design is a challenge with threatened species, particularly with those that are poorly known or extremely restricted in range and abundance, which often prevents or limits rigorous controls and replication (Radford et al. 2018). For these species, an adaptive management framework can guide effective management interventions, which are evaluated from data from long-term, continuous monitoring programs (Radford et al. 2018), which provide data on the persistence, trajectory, variability and status of threatened species or populations, and informs conservation management directions (Legge et al. 2018). Consistent and regular monitoring is key to identifying when interventions may be needed and for providing feedback on the effectiveness of interventions, particularly for those species with short lifespans, such as Stocky Galaxias.

Monitoring of threatened galaxiid species and populations in Australia has been undertaken primarily in Victoria and Tasmania since the early 1990s and forms the basis for many management actions (e.g. Raadik 1993, 2002, DOC 2004, Hardie et al. 2006, Raadik et al. 2010, TSS 2006, Chilcott et al. 2013, Raadik and Nicol 2013, Lintermans et al. 2014, Raadik 2018). This collective knowledge and experience have been used to develop a suggested multi-year monitoring program specific for Stocky Galaxias, which is endemic to the headwater reaches of the upper Murrumbidgee River, where it is known from a short reach of one small stream (Raadik 2011, 2014, Raadik and Lintermans 2022).

Monitoring can be characterised as either *compliance* (have required actions been undertaken or statutory thresholds breached), *surveillance* (low intensity or generic monitoring (generally long term at set sites) to alert managers that additional management intervention is required) or *intervention* (monitoring of the efficacy of specific management interventions) (Lintermans 2013a). The monitoring approach used in the current plan is predominantly a surveillance approach, but there are elements of intervention monitoring triggered by a TARP (Trigger Action Response Plan).

The following monitoring plan relates to the Stocky Galaxias at the single population it is currently known from (Lintermans and Allan 2019) but the described methods may also be relevant to other populations which may be discovered or established by translocations. It will provide comparable data on Stocky Galaxias, relative to risks such as non-native species incursions (see Cardno 2019; Raadik and Lintermans 2022) and importantly, inform decisions on management interventions such as translocations, broodstock collection for artificial breeding, disease surveillance, and non-native species control.

Should other populations of Stocky galaxias be detected in future, the monitoring plan should be reviewed, and consideration given to including these locations.

1.1 Relevance to priority conservation actions

Priority actions identified by NSW DPI (2017) that are relevant to this plan include:

 Monitor the population at Tantangara Creek over time to assess trends in abundance and distribution and to identify emerging threatening processes (medium priority).

- Investigate distribution, habitat and movements (medium priority).
- Identify and map important habitat (rivers/locations), particularly for recruitment and as potential drought refuge habitat (high priority).

As the monitoring plan could be applied to any, and all, known populations of Stocky Galaxias, it is also relevant to new populations which may be detected through a catchment survey or established from translocations. Therefore, the following, additional priority action (NSW DPI 2017), with those listed above, is also relevant:

• If other populations are discovered, undertake a genetic assessment of population structure throughout the species' range (Low priority).

The following priority action identified by the NSW FSC (2019) and included in the federal conservation advice for Stocky Galaxias (TSSC 2021) is also relevant to this plan:

• Population genetic analysis of current and new populations, to inform translocation plan and specific population management.

2 Monitoring plan aim and objectives

The overall aim of monitoring Stocky Galaxias is:

• To provide baseline, comparable data on the species, to inform decisions on management intervention for the long-term survival of the species.

Management of threatened species ultimately aims to recover populations and increase abundance and distribution so that the species may eventually be de-listed from threatened species legislation. To reach this endpoint, information is required on how the status of a species and trend of its population/s change through time, and so monitoring is an essential requirement of threatened species management. Monitoring should provide information on a focal population's 'status' (i.e. abundance; distribution and trend); demographics and recruitment; and if relevant, how these metrics respond to management actions. Other non-demographic metrics that may also be included in a monitoring program include identifiable threats; habitat availability and rate of loss; habitat condition and other critical resources needed. This information can then be used to frame, evaluate, and revise or refine management activities (Lintermans and Robinson 2018).

Consequently, the specific monitoring plan objectives for Stocky Galaxias, to meet the overall aim, is to provide baseline, comparable data on:

- 1. The persistence of Stocky Galaxias (presence and breeding).
- 2. The population trajectory (is the population increasing, stable or decreasing) and variability (significant change from normal).
- 3. The status of the Stocky Galaxias population (incorporating measures of abundance, distribution, reproduction, fish health and demographics).
- 4. The status of identifiable threats at Stocky Galaxias locations (e.g. riparian erosion, instream sedimentation, riparian vegetation condition with respect to ability to trap sediment).
- 5. The persistence and establishment of any new translocations of the species into the catchment.
- 6. Incursions of exotic fish species (Brown Trout (*Salmo trutta*), Rainbow Trout (*Oncorhynchus mykiss*), or invasive native species Climbing Galaxias (*Galaxias brevipinnis*) into known Stocky Galaxias populations.
- 7. Metrics informing triggers (as part of a TARP) for identified management interventions to mitigate potential sudden declines because of identified threats (e.g. fish incursion, fire, drought).

The effectiveness of the monitoring program will be dependent on obtaining estimates of critical population, habitat, and threat parameters, listed above. However, as no intensive, systematic monitoring has been undertaken to date, the effective analysis and interpretation of data will be constrained whilst data is accumulated over successive years.

2.1 Design considerations

Population and species criteria, including important attributes to be measured, are detailed in Table 1.

Specific monitoring questions are essential to establish the conceptual basis for monitoring and subsequent interpretation and to facilitate true adaptive management (Lindenmayer and Likens 2009, 2010, Lindenmayer et al. 2012). Importantly monitoring programs should lead to timely and informed management decisions and should not be an end in themselves (Lindenmayer et al. 2013).

All populations of Stocky Galaxias should be monitored to assess general population dynamics to understand population trajectories and persistence, but also include the use of a TARP to identify and where possible, mitigate, sudden declines that may be due to predator incursion or other stochastic events (e.g. drought, fire, etc.) (Raadik 2009 a,b; Raadik and Clunie 2007; Raadik 2019b). Monitoring will also be undertaken to inform outcomes from interventions such as stocking/translocations.

Table 1. Population and species parameters important in monitoring Stocky Galaxias

Criterion	Explanation	Life-history parameters				
Persistence	Continued presence over space and time	 presence of individuals across the sample range relative abundance of individuals 				
Trajectory	Direction of change over time	 individual condition (length / weight condition indice parasites, disease) 				
Variability	Fluctuation over time	 size structure of population (young of year, juveniles, sub-adults, adults)) 				
Status	Overall level of extinction risk	 – successful recruitment (abundance and proportion of population; abundance of individuals 				
		 level of genetic diversity and effective population size 				

All parameters are relevant to each criterion.

Specific monitoring questions can relate to threats, knowledge gaps, ecological attributes or life phases of the target species (e.g. Lintermans 2013b).

The proposed monitoring plan is framed by the following specific questions:

- 1. Will there be significant change over time in the abundance of Stocky Galaxias (Young-of-Year, juveniles and sub-adults/adults)?
- 2. Will there be a significant change over time in annual recruitment in Stocky Galaxias, or between populations if more are discovered or established?
- 3. Will translocated Stocky Galaxias survive following release, reproduce, and establish?

As there is currently only a single, known population of Stocky Galaxias, this monitoring plan's focus is on the whole species, considering the Tantangara Creek population represents the entire species' distribution. These questions and the methods set out here would also be appropriate for monitoring at the population level if additional populations are discovered (e.g. during a catchment survey). However, decisions on the duration of monitoring required for Stocky Galaxias conservation, the degree of monitoring undertaken for each population, and which populations need to be monitored cannot be determined in advance.

Stocky Galaxias are a critically endangered species, restricted to a single, small population and are at a high risk of extinction (Raadik and Lintermans 2022). Therefore, a major risk in undertaking monitoring of the species which involves capturing, handling, and removing tissue from, individuals, is an impact on the population/species through disturbance, injury or mortality, and habitat disturbance. This is a similar concern for other single-population, critically endangered, species of galaxiids (e.g. Raadik 1995; Raadik *et al.* 2010; Ayres et al. 2012a, b.; Raadik 2012; Stoessel et al. 2012, Chilcott et al. 2013; Raadik et al. 2019a,b; Stoessel et al. 2020), but is similarly justified, as monitoring is an essential part of broader conservation recovery management which aims to improve the resistance and resilience of the species, reduce extinction risk, and improve their conservation status. Without monitoring, no understanding of natural fluctuations in population/species is unknown, and no 'early-warning' of rapid decline is available. Some intervention is considered warranted to monitor and improve our understanding of this species which is of such a high risk of extinction.

Consequently, and like other single-population, critically endangered species of galaxiids (see above), monitoring and other management actions must be carefully undertaken to minimise the risk of impact as far as practicable, to achieve the goals of increasing knowledge and to progress species recovery. These should follow protocols which reduce impact to the population/species, such as biosecurity considerations to avoid introduction of novel disease/parasites, avoiding unnecessary disturbance of instream habitat, reducing impacts on spawning fish, eggs and larvae, and avoiding mortality during standard sampling events. Minimising disturbance during periods of stress, such as low-flow/cease-to-flow periods during drought, following fire, and during the spawning period and egg hatch/larval development stages is also critically important. Much of this can be achieved by utilising the expertise of field staff experienced in the conservation management of highly threatened species of fish, particularly galaxiids.

Crucially, minimising or ceasing harvest of adults and juveniles from the only known population of Stocky Galaxias, until new populations are discovered or captive management and breeding techniques are successfully developed, is critical for the persistence of Stocky Galaxias, as it removes an anthropogenic, and additional, stressor on the species.

3 Monitoring activities

The monitoring activities developed comprise three activities assigned as routine surveillance monitoring (long-term, ongoing) and four additional activities assigned as responses to trigger points under the TARP (See Section 4). Monitoring immediately following the establishment of a new translocated Stocky Galaxias population would occur as part of a Translocation Plan (Raadik and Stoessel 2022), but once established, this type of monitoring would revert to routine surveillance monitoring, as described here.

A summary of recommended activities is provided in Table 2. Further details and justification for the proposed methods are set out in section 3.1.

3.1 Routine surveillance monitoring

To avoid potential damage to the critically endangered population of Stocky Galaxias, the timing, frequency, and intensity of monitoring must be a compromise between reducing potential disturbance to the population and gathering adequate data to inform monitoring objectives.

3.1.1 Population monitoring

Purpose: Intensive population monitoring of most life-history attributes.

Value: Fish presence/absence, distribution, relative abundance, size and condition, recruitment success.

Timing: Autumn (March/April).

Frequency: Annually. Additional sampling may be required if triggered by the TARP (Table 4).

Method: Undertaken at multiple, pre-determined sampling sections at two monitoring sites on Tantangara Creek (Figure 1) using backpack electrofishing (see Section 4), maximising number of Stocky Galaxias collected, counted, measured for length (mm, length to caudal fork), weighed (grams), visually assessed for parasites or disease, returned alive to site of capture. Presence of other fish species or large invertebrates (crayfish) also to be recorded.

Water quality and stream characteristics should be measured at reach scale and a general threat assessment (instream and riparian zone) undertaken at reach scale.

Analysis and reporting: Annual.

3.1.2 Population genetics

Purpose: Monitoring level of population genetic variation and effective population size.

Value: Documentation of population genetic fitness and number of breeding adults (effective population size), which is not observable from life-history attributes; potential early warning of population genetic collapse and small parental stock. Can be undertaken during annual population monitoring.

Timing: Autumn.

Frequency: Every 3 years, or when triggered by TARP (Table 4).

Method: To occur during population monitoring. Collect a small sample of caudal fin tissue from all fish > 50 mm in length, clipping all fish up to a maximum of 45 individuals per reach (up to 90 individuals total) into 100% ethanol. Collected tissue sent for population genetic analysis using single nucleotide polymorphisms (SNPs) (effective population size, genetic diversity, parentage level analysis) at an appropriate facility. The number of tissue clips required each successive event may be able to be reduced following analysis of initial collection.

Analysis and reporting: every 3 years

3.1.3 Predator surveillance

Purpose: Detection of incursion of target predatory fish species into Stocky Galaxias populations.

Value: Early detection of predatory fish to avoid or minimise Stocky Galaxias decline.

Timing: Autumn (March/April) and early to mid-spring (Sept/Oct).

Frequency: Twice a year or when triggered by TARP (Table 4).

Method: Undertaken at the two population monitoring sites and just upstream of the waterfall on Tantangara Creek, 3 x eDNA water samples taken (max. 5 L each) per site. Samples analysed for presence of DNA of Climbing Galaxias, Brown Trout and Rainbow Trout, either onsite or at appropriate facility. Autumn sampling should occur in a manner that allows results to be available prior to the conclusion of the population monitoring campaign to enable rapid further investigation of any positive detections.

Analysis and reporting: Annual.

3.1.4 Monitoring methods and level of effort

Population monitoring

Several commonly used fish sampling techniques are available to monitor trends in relative abundance, population structure and distribution, ranging from active methods using electrofishing (boat or backpack) or dipnets to passive netting techniques (mesh nets, fyke nets, seine nets and bait/fish traps). Boat electrofishing and most netting techniques are effective in medium to large waterbodies with moderate depth, and often for larger species, however backpack electrofishing, dipnets and bait traps are commonly used in smaller, shallower waterbodies and small to medium-sized species. The most efficient method in smaller water bodies is backpack electrofishing, an active technique which can be used over large spatial areas and habitat or flow types, has relatively low species bias and immediate results. In comparison, dip netting is generally restricted to still environments, sampling small areas, rely on species entering the traps, and a long time-delay (from 4–12+ hours) for results.

Backpack electrofishing is the most common sampling method used in shallower, narrow upland, streams, which is like the habitat occupied by Stocky Galaxias. It is also an effective method for collecting sub-adult and adult galaxiids in small to medium-size streams (Lintermans 2000; Ayres et al. 2012a; Raadik 2014; Allan et al. 2018) and has previously been used to sample Stocky Galaxias (Raadik 2002, 2014, 2018; Allan et al. 2021) and to monitor population trends in other threatened upland galaxiids (e.g. Raadik 2002, 2019a,b; Raadik and Nicol 2013). Therefore backpack electrofishing is recommended as the most suitable sampling technique to use in the population monitoring of Stocky Galaxias for trends in relative abundance, population structure and distribution at monitoring sites. As it is also efficient in collecting trout in small to medium-sized streams, it is also suitable as a method for pest-fish surveillance.

However, the efficiency of backpack electrofishing operation for galaxiids is related to the power output and other settings of the unit (Reynolds 2016, Pottier et al. 2019), as well as the skill level of the operator in very low salinity (fresh) water, and their familiarity with galaxiids. These are discussed below. Further, electrofishing in low conductivity waters, such as at high elevation, will have difficulty stunning early-age larval fish due to their short length and therefore low susceptibility of stunning. Therefore, fine mesh dip netting is recommended for larval sampling as part of the trigger-based egg hatch success monitoring.

The timing of monitoring depends on the focus of monitoring activities and sampling efficiency, which can change with seasons and water levels. Monitoring of sub-adults and adults are best undertaken during autumn (March/April) when juveniles have reached a size at which they can be easily detected, adult gonad development is not overly influencing body weight, and stream conditions are conducive to efficient electrofishing (lower water levels and slower water velocity) (see Allan et al. 2021).

Annual population sampling of sites should be conducted in early to mid-autumn (March–April), as this is the time of lowest flows and Young-of-Year fish will be of an advanced size, both of which improve detection probability. Sampling earlier or later than this runs the risk of lower detection probability due to higher flows, with episodic high flow events during later sampling also introducing even greater variation in catch data.

The frequency of monitoring may depend to some degree on the focus of the monitoring activity, but as monitoring is focused on life-history aspects of the fish, will usually be undertaken once a year. For example, fish spawning and the presence of larvae occurs once annually. However, fish growth is continuous and size cohorts can be measured on multiple occasions throughout a year. The frequency of sampling in this case would therefore reflect the quality and quantity of data needed to monitor this life-history attribute; one monitoring event, repeated annually at the same time of year is considered adequate, based on other threatened species monitoring programs (Raadik et al. 2010, Lintermans 2013b, Tonkin et al. 2017).

Sampling Type	Frequency ⁺	Location/Area	Method	Duration/Level of Effort	Parameters measured			
Routine Surveillance Monitoring								
Population monitoring			Backpack electrofishing	0.75 day per site (up to 2 days Tantangara Creek). Continuous electrofishing per section to cover all habitat, single electrofishing run per section, fish processed before commencing next section.	No. of fish; length and weight; visual assessment for parasites; WQ* and stream characteristics; visual threat assessment.			
Population genetics	Collected and analysed every 3 years during population monitoring or as per TARP	As above	Caudal fin clipping	1.5–2.0 hrs per site. Fin clips to be collected from up to 90 individuals per site (Note: up to 45 individuals per reach at Tantangara Creek, 90 individuals total).	Using SNP data: Genetic diversity; effective population size; parentage analysis			
Predator surveillance	Twice a year, autumn (March/April) and early to mid- spring.	Immediately upstream of barrier protecting Stocky Galaxias population, and at each survey reach.	eDNA analysis of water samples.	~ 1.0 hrs per site. 3 replicate samples per reach taken across the stream (max. 5 L each) per site.	Detection of target species DNA (Climbing Galaxias, Brown Trout, Rainbow Trout) via developed and tested specific DNA probes.			
Trigger-based Monito	oring							
Spawning Monitoring	Trigger based in spring (Mid- October to early November).	Monitoring sites/reaches as specified in TARP	Backpack electrofishing	1 day duration populations x 2 sites x 4 events over 2 weeks (until spent fish detected). Up to 60 fish per site per sampling event.	As for population monitoring above with the addition of externally sexed and assessed for gonad stage (mature, running ripe, spent)			
Egg hatching success	Trigger based in summer (mid-summer)	Monitoring sites/reaches as specified in TARP	Fine-mesh dip nets x 2	~2–2.5 hrs per site. Total of 20 x 2 m long sweeps per reach.	Estimate of larval relative density, and total length (mm) range (subsample of 20 larvae per site); WQ* characteristics.			
Additional predator surveillance and incursion confirmation	Surveillance – Trigger based immediately following high rainfall events. Incursion confirmation – Trigger based immediately following positive eDNA detection	Monitoring sites/reaches as specified in TARP	eDNA analysis of water sampled. Backpack electrofishing.	Surveillance – see Predator surveillance monitoring above. Incursion confirmation - 2 days per population. Intensive continuous electrofishing (up to 3 runs) targeting all habitat sites, up to 500 m upstream from top of barrier.	Detection of target species DNA via developed and tested specific DNA probes. Target non-native species presence/absence, fish size, WQ* characteristics.			

Table 2. Summary of recommended monitoring activities as grouped into routine surveillance or trigger-based monitoring

Sampling Type	Frequency ⁺	Location/Area	Method	Duration/Level of Effort	Parameters measured
Fish condition assessment	Trigger based.	Monitoring sites/reaches as specified in TARP	Backpack electrofishing, fine mesh macroinvertebrate net and sample fixing equipment, WQ meter, fish stomach flushing equipment.	<i>Field</i> – collection of 20 adult fish total per site for parasites and stomach flushing; macroinvertebrate sampling and live pick, catchment and water condition assessment – 0.5 day per site (1 day total), <i>Laboratory</i> – Laboratory identification of stream macroinvertebrates and in gut samples – 1 to 1.5 day per site.	Fish length and weight; visual assessment for parasites; WQ* and stream characteristics; visual threat assessment, Stream macroinvertebrate collection for diversity and relative abundance, consumed macroinvertebrate mass, diversity and abundance.
					Parasite identification, location and load, and disease type – from histological examination.

* Water quality: includes water temperature, turbidity, dissolved oxygen, pH, conductivity. + Additional sampling may occur in response to the triggers identified in section 4.

Replication of sampling is important in monitoring projects to estimate sampling error (Dennis et al. 2010). We therefore recommend replicate sampling at both monitoring sites when undertaking population monitoring, by dividing each site into multiple sampling sections. We suggest separate 30 m long sampling sections, however, the number of sections will vary between 'Bottom Flat' and 'Top Flat' due to more overgrown stream reaches at the latter; five sections (150 m of stream length) can be established at 'Bottom Flat- Reach 2', though potentially only 3 sections (90 m stream length) at 'Top Flat – Reach 1' (Section 3.1.9). One section should be sampled at a time with all fish collected processed and released before moving to the next section upstream. A stop net should be positing at the upstream and downstream ends of each section before sampling, to maintain independence of sampling sections.

Physical sampling using backpack electrofishing equipment can be effective for surveillance of fish incursion upstream of the waterfall, however, detection probability can be very low if only a small number (e.g. <5) of fish are present, such as in the early phase of invasion. Therefore, as it is important to detect small numbers of invading fish, incorporating environmental DNA (eDNA) surveillance will improve detection probability (e.g. Bylemans et al. 2016, Hinlo 2018). The eDNA method must have high selectivity for the target fish (e.g. trout and Climbing Galaxias), but also very high sensitivity to a low abundance of DNA in the stream (Furlan et al. 2016, Hinlo et al. 2017).

Details of each gear and method is as follows:

Electrofishing. Backpack electrofishing for galaxiid monitoring should be undertaken as a single-pass, continuous sampling along a stream reach at a sampling site, which will facilitate comparisons with other long-term threatened galaxiid monitoring data. Therefore there is no restriction on the time taken to sample the reach, though power-on time and elapsed time of electrofishing should also be recorded for comparative purposes and for calculating catch metrics. Sampling is to be undertaken during daytime, beginning at a georeferenced starting point, with the operator and dip-netter wading upstream, stunning and retrieving fish, and sampling all available habitat. The finish location should also be georeferenced for future replication.

Due to the low conductance of freshwater in higher elevation and alpine regions because of very low dissolved salt and mineral levels, backpack electrofishing units should have a power output of > 900 volts to ensure they are not power limited in stunning or attracting fish. The frequency and '% of range' of the electrical wave output should also be able to be adjusted on the electrofishing unit to maximise its stunning and attracting capability for galaxiids, which are elongate, though relatively short, fish species: a 90-100 Hz frequency and 25-35% of range are usually effective (T. Raadik, unpublished data). These settings are also effective for the capture of trout, which are more sensitive to electrofishing.

Capture efficiency of galaxiids is further improved in low conductivity waters by altering the standard electrofishing approach of having the electrofisher anode and cathode fully immersed, to maximise the area of the cathode and minimise the area of the anode immersed. This effectively reduces the resistance of the anode (Reynolds 2016). Additionally, as water conductivity levels are low and fish may only be partially stunned by electrofishing, they can also be rapidly collected by the electrofisher operator using an anode ring covered in fine (~ 3 mm), non-conductive mesh. This reduces escape of recovering fish and the need to stun fish multiple times, allows continuous sampling without the need for the operator to wait until stunned fish are picked up by the dip netter, and avoids the need for the dip netter to come too close to the electrofishing operator.

Dip netting: To collect early-age larvae, a fine mesh (0.5–1 mm diam.) dip net (~ 300 m diam.) is to be used in flowing and still habitats. Sampling is to be undertaken by 20 x 30-second elapsed time sweeps of the net through the water at different habitats, with the content of the net placed into a bucket of water between sweeps.

eDNA water samples: For eDNA analysis of target pest species of fish, 3 x replicate water samples of up to 5 L each are to be collected in a transect across the sampling site, using an eDNA water filtering device. Collected samples are to be uniquely identified and either processed onsite using a portable and rapid analysis machine (e.g. Biomeme[®] for qPCR, Genie III[®] for LAMP or qPCR, etc.), or delivered to an eDNA analysis laboratory. Water samples are to be filtered using an agreed filter size, using a protocol to minimise cross-contamination between replicates and between sites, and stored appropriately, as advised in advance by the eDNA analysis laboratory. The water volume filtered and filter pore size per replicate are also to be recorded.

Onsite eDNA analysis is recommended from a time efficiency and lower cost perspective, though only if the analysis capability (sensitivity) of the portable unit is considered suitable (i.e. comparable to the sensitivity of lab-based analysis). Rapid, field-based, eDNA analysis methods and equipment are in continual development, and are rapidly improving for certain applications.

Fish processing and data management

When captured, fish will be placed into bucket of water carried by the dip netter. At the end of sampling, all fish captured will be placed into a larger, aerated, container whilst being processed. Fish are to be picked up using a soft, fine-mesh aquarium dip net for transfer to a wetted measuring board and then a wetted weighing dish, followed by release to the site of collection, or to another water-filled, aerated container before final release. Handling of fish must be done with wet hands. All fish species captured will be identified and measured for length (nearest mm; Caudal Fork Length or Total Length, as appropriate). Weight of each Stocky Galaxias captured will be recorded (0.1 g), and fish will be visually examined for deformities, injuries (e.g. cormorant strike) and external parasites (e.g. *Lernaea cyprinacea*).

Data collected in the field will be manually recorded then entered electronically and maintained (curated and backed up) during data analysis ensuring appropriate review and quality assurance procedures.

3.1.5 Life-history parameters measured

Population monitoring

Life history parameters to be recorded and reported as part of an annual population monitoring program for Stocky Galaxias are provided in Table 1. Many of these standard biological attributes are commonly recorded during monitoring programs for threatened fish, particularly galaxiids in Victoria. These can vary spatially and temporally within a population, and importantly, provide multiple but differing measures of population condition.

The continuing *Presence* of fish over time at a monitoring location provides evidence of the persistence of that species at that location or site. Moreover, when a suite of sites extends across the species distribution, or expected distribution, information is gathered on the populations range and any changes to such. The latter is particularly useful for rare species, where relative abundance measures are often variable and therefore uninformative. However, presence and range of fish alone does not provide data on population health or variability, or small changes to population trajectory.

Relative Abundance, usually from counting all individuals in a sample in a survey reach relative to sampling effort, or in a subsample of <u>age</u>/size classes, provides an indication of population structure and recruitment success and <u>expected</u> fish persistence over time.

Condition, usually measured as individual fish weight relative to length, and a visual inspection of external parasites or signs of disease, provides a qualitative measure of general fish health. Fish weight/condition can vary depending on the degree of gonad development, and consequently this should be measured when gonad development is minimal (e.g. during summer/autumn) or at a standard time of year (so between year changes are directly comparable). The number of fish visually identified with external parasites or disease indicates the degree of infection of the population and the level of individual infection can give important information on average disease/parasite load.

The *Size Structure*, using length data, of a population is based on length measurements of individuals, and indicates recruitment success, adult (spawning stock) abundance, fish persistence and growth (age/length cohorts) over time.

Spawning and *Recruitment* success is an important indicator of fish persistence and abundance over time, and of successful reproduction. As part of the population monitoring program, the success of spawning and recruitment will be evidenced through presence of an adequate proportion of Young-of-Year individuals.

Statistical analysis

No previous standardised monitoring data exists, and therefore, initial analysis of data will be relatively simplistic until at least 2 and possibly 3 years of monitoring data is accumulated. Further, as only one population is known, to improve inference, replication is provided at the population level (two monitoring sites in Tantangara Creek), at each site consists of multiple, independent, sampling sections (Section 3.1.9).

Distribution. As only one population is currently known, annual fluctuation in species distribution is not expected. Therefore, presence data at monitoring sites will provide a simple descriptive approach of temporal trends in distribution in Tantangara Creek and will consider any heterogeneity in the distribution of juvenile and adult life stages. If additional Stocky Galaxias populations are found or established, monitoring of fish presence can then inform temporal trends in distribution for the species, presented as reporting rates across sites (frequency of detection across a range).

Relative abundance, condition and parasite load (will also inform triggers in Section 6). Generalised Additive Models (GAMs) are often employed to assess trends in abundance through time and will likely suit these datasets once more than two or more years of data are collected. The response variables (e.g. relative abundance, condition, parasite load) will be modelled against year (to determine inter-annual variability), and environmental variables like annual rainfall to help explain any observed variability in the data, using GAMs

or generalised additive mixed models (GAMMs). Such models would detect changes both within populations and for the species as a whole (if further populations are found and included in the monitoring). This approach compares sampling occasions to an arbitrarily chosen baseline year (e.g. the first sampling year) out of necessity as there is no pre-existing data. If the first year is uncharacteristic (e.g. due to the recent fires), leading to wide confidence intervals in the time-series, a different base year may be chosen. As Stocky Galaxias are unable to effectively disperse, as they are located above a waterfall with predators below, emigration or immigration are not factors to consider in population fluctuations, and this is expected to be similar if additional populations are found or established.

Recruitment and adult survivorship (will also inform triggers in Section 6). Analysis of change in lengthfrequency will provide insight into temporal trends in adult stock, survival, and recruitment that aren't captured by simple relative abundance estimates. For example, a large increase in population size accompanied by a reduction in mean fish length within the lower size range reflects recruitment of Young-of-Year fish into the population (fish > 1 year of age). Conversely, an increase in population size but with no reduction in the lower size range is more likely to reflect no change in recruitment rates but rather an increase in survival of fish already accounted for in the population. Length-frequency histograms will accurately determine the age cohorts (e.g. Young-of-Year and age 1+), thereby informing recruitment dynamics. Changes in the relative frequency of these cohorts (i.e. abundance of fish of those size classes) between years will then be assessed using GAMs, like those described above.

As annual Stocky Galaxias population monitoring data accumulates, consideration should be given to using state-space populations models, which provide a more mechanistic process for analysis (Lintermans et al. 2022).

3.1.6 Population genetics

Population/species genetic diversity is an important component of population persistence, health, and evolutionary potential. Monitoring a population's genetic structure is now an established technique that provides additional information on population demography and dynamics. Measures such as genetic diversity, level of inbreeding, effective population size, and kinship as determined by genetic analysis of a subset of individuals, are important components to enable an assessment of population persistence, health, and evolutionary potential.

Besides taxonomic-level genetic information on Stocky Galaxias (Adams et al. 2014), no population level information is published on genetic diversity of Stocky Galaxias. As such, non-destructive sampling of genetic tissue collected from individuals captured (small fin tissue sample stored in 100% ethanol), is proposed as part of the broader monitoring program. Once genetic diversity is assessed for Stocky Galaxias (year 1), we suggest re-assessing this once a generation, which is estimated to be every 3 years (Lintermans and Allan 2019), unless the TARP indicates more frequent assessment.

The analysis of genetic data to assess changes in genetic diversity, survival of translocated offspring, and to identify whether individuals captured are of translocated or natural spawned origin, should follow the methods described in Lutz et al. (2021). In summary this will involve analysis of identity, sibship and parentage using SNP data, including analysis to assign individuals to a translocated or source population, and to generate genetic indices such as proportion of heterozygous loci (PHt), effective population size (*Ne*), levels of heterozygosity (expected and observed), allelic richness and private alleles, etc.).

3.1.7 Environmental variables

Environmental variables are also important to monitor, as they can provide qualitative understanding to the life-history parameters above, and to sampling efficiency. Data on environmental variables will be collected as part of the routine surveillance monitoring.

Standard water quality parameters are water temperature (°C), electrical conductivity (μ S/cm), dissolved oxygen levels (mg/L and % saturation), pH, turbidity (NTU). Water hardness (mg/L CaCO₃) can also be important if fish are to be kept in aquaria at some stage. Data is to be collected on-site with a portable, calibrated multimeter. As water quality may not vary greatly along a short stream reach, it is to be taken once at each sampling site on each sampling event.

Variables related to the sampled aquatic habitat are also important, particularly to standardise survey data (e.g. abundance, etc.) and to define physical and aquatic changes since the last site visit. These are average stream wetted width and depth (m) calculated from measurements from several georeferenced transects at ~ 10 m intervals (width by measuring tape and depth by steep ruler), and water level height (m) at the time of sampling, as a measure of relative change (e.g. measured from a fixed height marker). These should be recorded on each monitoring occasion, at each monitoring site, and at all sampling sections within reaches.

3.1.8 Threat monitoring

Visual assessment and characterisation of threats at each sampling site will give early warning of potential management issues. For example, is there excessive sediment accumulating in the stream (a threat to refuge pool habitats and spawning habitats); is general vegetative cover adequate (to minimise sediment input), etc. A rapid, visual assessment of potential threats will be undertaken at each sampling site, on each sampling occasion, and habitat conditions scored. This may provide additional qualitative understanding to measured fish life-history parameters and should be supported by images taken from two georeferenced photo points (one from upstream and one from downstream). For example, increased levels of instream sedimentation may explain a lack of recruitment success if it reduces or eliminates egg deposition sites.

As aquatic predators and competitors (Brown Trout (*Salmo trutta*), Rainbow Trout (*Oncorhynchus mykiss*), and Climbing Galaxias (*Galaxias brevipinnis*)), are a major threat to Stocky Galaxias (Raadik 2014, Lintermans and Allan 2019, Lintermans et al. 2020), their continued absence within the habitat of the Stocky galaxias should be monitored more frequently and rigorously. This is essential, as trout can eliminate a population of galaxiids from a short, narrow section of stream in 6–12 months (Raadik et al. 2010). Therefore, frequent surveillance for non-native fish incursion is critical, as early detection is essential to allow for rapid control of invading fish before the Stocky Galaxias population is severely impacted or eliminated.

Given the current high extinction risk to Stocky Galaxias, surveillance will therefore be undertaken twice a year, once during autumn and once in early to mid-spring following higher winter stream flows. Additional sampling will be informed by the TARP. For example, a positive eDNA detection will initiate physical sampling while very high rainfall events may trigger additional eDNA sampling as high flows may allow the colonisation of non-native fish upstream into the Stocky Galaxias population above the waterfall on Tantangara Creek.

3.1.9 Monitoring sites

Currently there is only one known population of Stocky Galaxias, confined to a short, 3 km headwater section of Tantangara Creek. If additional populations are discovered (e.g. via the catchment survey), they must also be monitored, though this can be prioritised based on their geographic spread from other known populations, and value to Stocky Galaxias conservation, such as range size, population abundance, genetic health (genetic diversity and uniqueness), etc.

With respect to Tantangara Creek and based on previous unpublished work by Hugh Allan (*pers. comm.*), it is suggested that two monitoring sites be established, one in the mid reaches of the catchment ('Top Flat' – Reach 1), and one in the downstream section but upstream of the waterfall ('Bottom Flat' – Reach 2) (Table 3, Figure 1). Stream characteristics of these two sites varies, with the upper being smaller and shallower, and consequently supporting a lower abundance of fish. Recent impacts from the 2019/20 fires differed between the two sites (M. Lintermans, unpublished data), so monitoring both will give some indication of fire recovery, given the very short home-range (30 m) of Stocky Galaxias (Allan and Lintermans 2019). Consequently, monitoring two sites differing in fish community structure will provide a more accurate assessment of population fluctuations over time, particularly as fish in the upper site may be more prone to impacts from reduced flows during drier periods. Access to these two locations is by hiking a short distance from the nearby Alpine Creek Firetrail.

Trout/Climbing Galaxias invasion surveillance should be undertaken at the two population monitoring sites, and further downstream, just above the waterfall and just above the planned galaxiid barrier (Site 3) (Figure 1). As a requirement of surveillance is early detection, before species can spread or multiply, the additional location (early occupation zone) allows for potential detection of invasion at the most likely place, over the waterfall into the most downstream reach of the Stocky Galaxias population. Subsequent spread upstream can be detected at 'Bottom Flat' and 'Top Flat', though these can also allow for detection if target fish are deliberately placed into Tantangara Creek further upstream.

Table 3. Proposed monitoring sites on Tantangara Creek, including access, land tenure, availability of previous comparable data., and stream dimensions

Coordinates are given for approximate downstream extent of sites. Approximate average stream width and depths from Allan et al. (2021), M. Lintermans (unpublished data) and T. Raadik (unpublished data).

Site/ name	Coordinates: Decimal Latitude, Longitude	Public or private access	Land Tenure	Previous, comparable monitoring data available	Average Width (m)	Average depth (m)
Tantangara Creek Site 1 – Top Flat	-35.849597 148.569540	Public	National Park	No	0.7	0.06
Tantangara Creek Site 2 – Bottom Flat	-35.840144 148.568365	Public	National Park	No	0.9	0.15
Tantangara Creek Site 3 – Falls	-35.838890 148.569120	Public	National Park	No	1.1	0.25
(Pest fish eDNA monitoring only)						

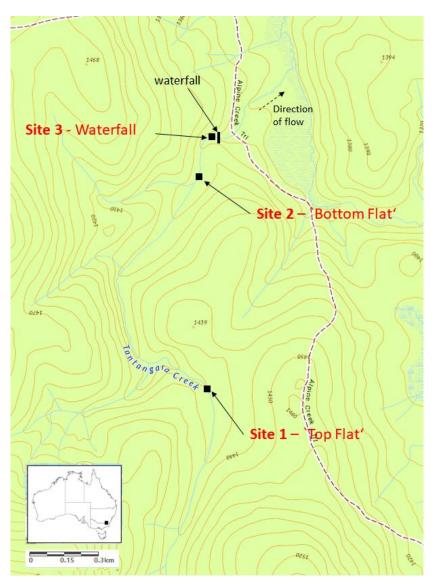


Figure 1. Map of proposed monitoring sites on Tantangara Creek Filled squares indicate approximate position of the downstream extent of each site.

4 Trigger Action Response Plan (TARP) criteria associated with potential results of the monitoring plan

A TARP provides a framework for decision making in response to a deviation from a 'normal' condition in key criteria. For Stocky Galaxias, the objective of such a plan is to identify threats that may cause a sudden decline in Stocky Galaxias and enable an assessment of cause with trigger values being informed by monitoring activities. A TARP for environmental monitoring also has several levels of activity or response, from 1 (normal), 2 (alert/investigation), to 3 (high alert/rectify).

Criteria, trigger levels and suggested response activities for Stocky Galaxias are provided in Table 4. These have been developed from an understanding of the biology/ecology of the species and key threats (Raadik and Lintermans 2022), and key monitoring activities proposed above, including potential monitoring outcomes. Note that specific trigger levels for genetic diversity cannot be defined as the genetic diversity of Stocky Galaxias is currently unknown.

As monitoring continues and knowledge of Stocky Galaxias increases, the trigger levels should be periodically reviewed (annually for three years and then every third year following) to ensure they are fit for purpose.

If monitoring indicates an Alert Level 3 for one or more of the criteria in Table 4 below, significant intervention may be required such as the emergency collection of individuals into captivity or restocking of a population from captive breeding. Such an activity will require significant input from NSW DPI. Given that such interventions are often very time sensitive, it is recommended that an emergency intervention procedure is developed in conjunction with NSW DPI if one does not already exist.

4.1 Trigger-based sampling

The TARP details a decision-making framework for additional sampling in response to deviations from a 'normal' condition in Stocky Galaxias population metrics (see section 5) or other threats identified in the routine surveillance monitoring (see section 3.1). This trigger-based sampling aims to identify specific processes (e.g. spawning failure) and threats (e.g. trout/Climbing Galaxias incursion) that may cause a sudden decline in population metrics such as recruitment and survival.

Assessment of successful spawning and/or egg hatch

If failure to detect recruitment of Young-of-Year occurs, it is necessary to investigate at which life phase the failure has occurred (spawning, hatching, larval growth and survival) and why. Determination of whether 1) spawning has occurred can be simply quantified by monitoring for adult fish (mainly females) with 'spent' gonads (mid-spring), 2) successful hatch by presence of newly hatched larvae (mid-summer), and 3) larval growth and survival; by presence of juvenile fish at the time of normal population monitoring (autumn). Only 1 and 2 are trigger-based. By undertaking this assessment in the spring following an observed > 30% decline of age 0+ fish in 1 year, a repeated failure can be detected as early as possible as well as an investigation into the cause of the decline.

4.1.1 Spawning assessment

Purpose: Monitoring of spawning activity.

Value: Evidence of gonad maturation in adult fish and indirect evidence of spawning (spent adult fish). Contributes to identifying unsuccessful life-history stage(s) if recruitment failure is detected (e.g. gonad maturation, spawning, hatching).

Timing: Mid-October to early November.

Frequency: Trigger based (see Table 4).

Assessing **spawning success** must commence close to the spawning period (mid-October to early November; Allan et al. 2021). Once a week, until spent (mainly female) fish are detected, 60 mature fish are to be randomly collected by backpack electrofishing over a selected reach at each monitoring site (sites 1 and 2), and visually, externally sexed and assessed for spawning condition based on gonad developmental stage (mature, ripe, running ripe, spent) following Stoessel et al. (2012, 2015), adapted from Pollard (1972).

Table 4. Criteria, trigger levels, and suggested response activities, for each alert level for Stocky Galaxias

Criteria Alert level	Predatory fish invasion	Decline or loss of recruitment	Decline in relative population abundance	Decline in fish condition	Genetic decline	Fire or drought (risk of sedimentation, water loss)	Water quality decline (pH, oxygen, temperature)
1 (Normal)	Continue predator surveillance.	Continue population monitoring.	Continue population monitoring.	Continue population monitoring.	Continue monitoring.	Continue monitoring	Continue monitoring.
2 (Alert / investigate)	<i>Trigger:</i> rainfall event > 90 mm/24 hrs or predatory fish detection by eDNA. <i>Response:</i> - undertake predator confirmation sampling (See 4.1.3) Note: rainfall trigger level to be reviewed following construction of Climbing Galaxias barrier.	<i>Trigger:</i> Complete loss, or > 30% decline of 0+ fish in 1 year. <i>Response:</i> - undertake spawning and egg hatching monitoring in subsequent season to establish the life stage where the failure may be occurring and seek to identify the cause (See 4.1.1 and 4.1.2).	<i>Trigger:</i> > 15% decline in non- larval/early juvenile phases in 1 year. <i>Response:</i> - undertake egg hatching monitoring in subsequent season to determine level of recruitment (4.1.2). - increased disease/parasite monitoring (samples to Veterinary clinic) (See part of 4.1.4).	<i>Trigger:</i> > 30% of individuals declined in condition by > 25%; > 10% of individuals with externally visible parasites/disease. <i>Response:</i> - investigate diet and catchment conditions (See 4.1.4). - increased disease/parasite monitoring (samples to Veterinary clinic) (See 4.1.4).	<i>Trigger:</i> Loss of genetic diversity, increasing level of inbreeding. <i>Note</i> – specific trigger levels to be defined following first population genetic analysis. <i>Response:</i> - undertake additional round of genetic sampling in subsequent year (See 3.1.2).	Trigger: > 10% of catchment area burnt or streamflow ceases at one or both monitoring sites (visual inspection). Response: Following fire, a visual assessment is required as soon as practical regarding the potential for a Level 3 trigger to occur. If a cease to flow is observed during population monitoring, weekly visual assessments should commence to determine the need for additional population monitoring (See 3.1.1).	Trigger: 5-day average air temperature at the nearest meteorological station exceeds 15.0°C (winter) or 29.0°C (summer), dissolved oxygen levels < 5 mg/l, pH change by 1.0. <i>Response</i> : - undertake additional WQ monitoring to determine the need for additional population monitoring (See 3.1.1) and/or identify if a Level 3 trigger is met.
3 (High alert / rectify)	<i>Trigger:</i> Predatory fish detection confirmed by electrofishing. <i>Response:</i> Immediately notify NSW DPI and enact emergency intervention procedure.	<i>Trigger:</i> Loss or > 30% decline of 0+ fish in 2 consecutive years. <i>Response:</i> Notify NSW DPI and enact emergency intervention procedure.	<i>Trigger.</i> > 50% decline in non- larval/early juvenile phases in 1 year or > 15% decline in consecutive years. <i>Response:</i> Notify NSW DPI and enact emergency intervention procedure.	<i>Trigger:</i> > 50% of individuals declined in condition by > 25%; > 25% of individuals with externally visible parasites/disease. <i>Response:</i> Notify NSW DPI and enact emergency intervention procedure.	<i>Trigger:</i> Note – specific trigger levels to be defined following first population genetic analysis. <i>Response:</i> Notify NSW DPI and enact emergency intervention procedure.	<i>Trigger</i> : High risk of post- fire debris flow/instream sedimentation event, or pools drying. <i>Response</i> : Notify NSW DPI and enact emergency intervention procedure	<i>Trigger</i> : Water temperature > 10°C (winter) > 25°C (summer), dissolved oxygen levels < 3 mg/l, pH change by 1.5. <i>Response</i> : Notify NSW DPI and enact emergency intervention procedure.

All percentage changes are relative to mean baseline data following the first year of monitoring.

The length (LCF to nearest mm) and weight (to nearest 0.1 g) of each fish, including sex and gonad stage are to be recorded, including the previously specified WQ parameters. Assessment ceases when more than two spent females are found during one assessment event. Fish should also be visually assessed for parasites or disease and returned alive to site of capture, and a general threat assessment (instream and riparian zone) undertaken at the site scale. The reach at each monitoring site is to be selected and established on the first trigger event.

4.1.2 Egg hatching success

Purpose: Monitoring success of spawning and hatching.

Value: Evidence of success (larvae present) or failure (no larvae detected) of egg development and hatching. Contributes to identifying unsuccessful life-history stage(s) if recruitment failure is detected (e.g. gonad maturation, spawning, larval hatching).

Issue: Recruitment success or failure is also determined from the intense population monitoring activity.

Timing: Mid-January to mid-February.

Frequency: Trigger based (see Table 4).

Assessment of **larval presence** is to be undertaken within a 60 m long georeferenced reach in each of monitoring sites 1 and 2 on Tantangara Creek using dipnets and visual observation. It is to occur after egg hatch by visual observation using polaroid glasses, into pools and slower flowing areas, and presence and relative abundance of fish by 10 x 2 m-long sweeps of a fine-mesh dip net (see Section 3.1.4 above), randomly chosen across the reach, and estimating water volume sampled. Total number of larvae to be estimated, and total length of a subsample of 20 larvae to be measured for length (to nearest mm).

4.1.3 Predator surveillance (TARP Triggered)

Purpose: Detection of incursion of non-native fish (Climbing Galaxias, Brown Trout, Rainbow Trout) into Stocky Galaxias population.

Value: Early detection of predatory fish to avoid, or minimise, Stocky Galaxias decline.

Timing: Trigger based (see Table 4).

Frequency: Trigger based (see Table 4).

Method: Undertaken at the two population monitoring reaches (sites 1 and 2) and just upstream of the waterfall on Tantangara Creek (site 3) when 1) triggered by intense rainfall events (undertake eDNA), or a positive eDNA detection from routine, or trigger-based, predator surveillance. Positive eDNA detections are to be verified by intensive backpack electrofishing for predatory species, and standard water quality parameters, including effort, recorded.

Detection of predatory fish

Non-native fish (Brown Trout, Rainbow Trout and Climbing Galaxias) are a significant threat to Stocky Galaxias, mainly through predation of small to large size classes. Early detection of an incursion of these species into the Stocky Galaxias population is critically important for the ongoing survival of the species. The routine population, and predator surveillance monitoring has the capacity to detect the presence of these species. However, positive detections via eDNA analysis require rapid confirmation with physical sampling. Randomly occurring, intense rainfall events, outside of routine monitoring periods, may also provide opportunities for predatory fish incursion and therefore require additional eDNA sampling and analysis, with follow-up of positive detections with physical sampling.

If a report of a pest fish sighting or positive eDNA detection is received conventional sampling using backpack electrofishing is to be undertaken at the three monitoring sites to validate the detection result. Intensive, continuous electrofishing, targeting all habitats over a 500 m reach at each monitoring site (upstream from most downstream point), using 1–2 teams and repeated sampling runs (max. of three) at each site if required. Water quality parameters, and standard electrofishing effort data to be recorded, and if detected, live images to be taken and preserved vouchers (1–2 individuals) of each species of target non-native fish to be retained, including georeference of detection points. Additional non-native fish to be euthanased and appropriately disposed of.

Additional surveillance outside of routine surveillance periods, is to be undertaken immediately following one, or successive, high intensity rainfall events which may affect the waterfall effectiveness in preventing fish movement by cause flooding, erosion, or temporary barrier bypass options. A rainfall intensity trigger (e.g. 24-hour rainfall total), based on the Tantangara catchment characteristics and current waterfall/future predator barrier condition, should initiate surveillance, if exceeded (see Section 5). If triggered, eDNA

sampling, as per routine predator surveillance (section 3.1.3) to be undertaken, and positive eDNA detections to be verified as per preceding paragraph.

4.1.4 Fish Condition Assessment

Purpose: Identification of cause of decline in fish condition.

Value: To enable early intervention to reduce or reverse decline in condition before impact to Stocky Galaxias population occurs.

Timing: Trigger based (see Table 4).

Frequency: Trigger based (see Table 4).

Method: Undertaken at the two population monitoring reaches (sites 1 and 2), triggered by observed decline in fish condition metric (fish length/weight relationship) generated from annual monitoring and/or increased parasite load. Depending on the observed decline and catchment conditions consider the following activities:

- Collect individuals by electrofishing for intensive investigation of internal and external disease/parasite by appropriate Veterinary Clinic. Total of 5–10 adults, preserved appropriately.
- Undertake detailed investigation of diet (stomach flushes of adults to determine incidence of feeding (e.g. gut fullness, etc.)). 10 adults per site, undertake flushing at site and analysis of contents at laboratory, fish released alive to reach of capture.
- Assess instream aquatic macroinvertebrate abundance and diversity. At both sites, kick sample and edge sweep, followed by live sort, and laboratory processing as per EPA (2003).
- assess catchment conditions for threats likely impacting aquatic macroinvertebrates (e.g. sediment) or water quality.

Consider the need to continue monitoring fish condition and parasite/disease, including standard water quality parameters whilst undertaking investigations.

5 References

- Adams, M., Raadik, T.A., Burridge, C. and Georges, A. (2014). Global biodiversity assessment and hypercryptic species complexes: more than one species of elephant in the room? *Systematic Biology* 63(4), 518–533.
- Allan, H., Duncan, R.P., Unmack, P., White, D., and Lintermans, M. (2021). Reproductive ecology of a critically endangered alpine galaxiid. *Journal of Fish Biology* **98**(3), 622–633.
- Allan, H. and Lintermans, B. (2019). *Current knowledge of the critically endangered Stocky Galaxias* Galaxias tantangara. Report to EMM Consulting Pty Ltd.
- Allan, H., Unmack, P., Duncan, R.P. and Lintermans, M. (2018). Potential impacts of PIT tagging on a critically endangered small-bodied fish: A trial on the surrogate mountain galaxias. *Transactions of* the American Fisheries Society **147**, 1078–1084.
- Ayres, R.M., Nicol, M.D. and Raadik, T.A. (2012a). Establishing new populations for fire-affected barred galaxias (Galaxias fuscus): Site selection, trial translocation and population genetics. Black Saturday Victoria 2009 – Natural values fire recovery program. Heidelberg, Australia: Department of Sustainability and Environment.
- Ayres, R.M., Nicol, M.D. and Raadik, T.A. (2012b). Guidelines for the translocation of Barred Galaxias (Galaxias fuscus) for conservation purposed. Black Saturday Victoria 2009 – Natural values fire recovery program. Department of Sustainability and Environment, Heidelberg, Victoria.
- Bylemans, J., Furlan, E. M., Pearce, L., Daly, T. and Gleeson, D.M. (2016). Improving the containment of a freshwater invader using environmental DNA (eDNA) based monitoring. *Biological Invasions* 18, 3081–3089.
- Cardno (2019). Appendix M.2 Aquatic Ecology Assessment, Snowy 2.0 Main Works. Prepared for EMM Consulting Pty Ltd. Cardno, St Leonards, NSW.
- Chilcott, S., Freeman, R. Davies, P.E., Crook, D.A., Fulton, W., Hamr, P., Jarvis, D. and Sanger, A.C. (2013). Extinct habitat, extant species: lessons learned from conservation recovery actions for the Pedder galaxias (*Galaxias pedderensis*) in south-west Tasmania, Australia. *Marine and Freshwater Research* 64, 864–873.
- Dennis, B., Ponciano, J.M. and Taper, M.L. (2010). Replicated sampling increases efficiency in monitoring biological populations. *Ecology* **91**(2), 610–620.
- DOC (Department of Conservation). (2004). *New Zealand non-migratory galaxiid fishes recovery plan 2003–* 13. Threatened Species Recovery Plan 53. Wellington.
- EPA (2003). Rapid Bioassessment methodology for rivers and streams. Guideline for Environmental management. EPA Victoria, Southbank, Melbourne.
- Furlan, E.M., Gleeson, D., Hardy, C.H. and Duncan, R.P. (2016). A framework for estimating the sensitivity of eDNA surveys. *Molecular Ecology Resources* **16**, 641–654.
- Hardie, S., Jackson, J.E., Barmuta, L.A. and White, R.W.G. (2006). Status of galaxiid fishes in Tasmania, Australia: Conservation listings, threats and management issues. *Aquatic Conservation: Marine and Freshwater Systems* **16**(3), 235–250.
- Hinlo, R., Gleeson, D., Lintermans, M. and Furlan, E. (2017). Methods to maximise recovery of environmental DNA from water samples. PLoS ONE **12**(6): e0179251.
- Hinlo, R., Lintermans, M., Gleeson, D., Broadhurst, B. and Furlan, E. (2018). Performance of eDNA assays to detect and quantify an elusive benthic fish in upland streams. *Biological Invasions* **20**,3079–3093.
- Legge, S., Lindenmayer, D., Robinson, N.M., Scheele, B.C., Southwell, D.M. and Wintle, B. (2018). Introduction: making it count. In: Legge, S., Lindenmayer, D., Robinson, N.M., Scheele, B.C., Southwell, D.M. and Wintle, B. (Eds.) *Monitoring Threatened Species and Ecological Communities*, pp. 1–10. CSIRO Publishing, Clayton South, Victoria.
- Lintermans, M. (2000). Recolonization by the mountain galaxias *Galaxias olidus* of a montane stream after the eradication of rainbow trout *Oncorhynchus mykiss*. *Marine and Freshwater Research* **51**, 799–804.

- Lintermans, M. (2013a). Conservation and management. Pp 283–316 In, Humphries, P and Walker, K (eds) The Ecology of Australian Freshwater Fishes. CSIRO Publishing, Collingwood, Victoria.
- Lintermans, M. (2013b). The rise and fall of a translocated population of the endangered Macquarie perch *Macquaria australasica* in southeastern Australia. *Marine and Freshwater Research* **64**, 838–850.
- Lintermans, M. and Allan, H. (2019). *Galaxias tantangara*. The IUCN Red List of Threatened Species 2019: e.T122903246A123382161. http://dx.doi.org/10.2305/IUCN.UK.2019-3.RLTS.T122903246A123382161.en
- Lintermans, M., Geyle, H.M., Beatty, S., Brown, C., Ebner, B., Freeman, R., Hammer, M., Humphreys, B., Kennard, M.J., Kern, P., Martin, K., Morgan, D., Raadik, T.A., Unmack, P.J., Wager, R., Woinarski, J.C.Z., and Garnett, S.T. (2020). Big trouble for little fish: Australian freshwater fishes in imminent risk of extinction. *Pacific Conservation Biology* **26**(4), 365–377.
- Lintermans, M., Lyon, J.P., Hames, F., Hammer, M.P., Kearns, J., Raadik, T.A. and Hall, A. (2014). Managing fish species under threat: case studies from the Native Fish Strategy for the Murray– Darling Basin, Australia. *Ecological Management and Restoration* **15**(S1), 57–61.
- Lintermans, M. and Robinson, W. (2018). The extent and adequacy of monitoring for Australian threatened freshwater fish species. In: Legge, S., Lindenmayer, D., Robinson, N.M., Scheele, B.C., Southwell, D.M. and Wintle, B. (Eds.) *Monitoring Threatened Species and Ecological Communities*, pp. 85–99. CSIRO Publishing, Clayton South, Victoria.
- Lintermans, M., Tonkin, Z., Lyon, J. and Gilligan, D. (2022). *Macquarie Perch monitoring plan, Snowy 2.0.* Published client report for Snowy Hydro Ltd, Cooma. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg, Victoria.
- Lindenmayer, D.B. and Likens, G.E. (2009). Adaptive monitoring: a new paradigm for long-term research and monitoring. *Trends in Ecology & Evolution* **24**(9), 482–486.
- Lindenmayer, D.B. and Likens, G.E. (2010). The science and application of ecological monitoring. *Biological Conservation* **143**(6), 1317–1328.
- Lindenmayer, D.B., Likens, G.E., Andersen, A., Bowman, D., Bull, C.M., Burns, E., Dickman, C.R., Hoffman, A.A., Keith, D.A., Liddell, M.J., Lowe, A.J., Metcalfe, D.J., Phinn, S.R., Russell-Smith, J., Thurgate, N. and Wardle, G.M. (2012). Value of long-term ecological studies. *Austral Ecology* **37**(7), 745–757.
- Lindenmayer, D.B., Piggott, M.P. and Wintle, B.A. (2013). Counting the books while the library burns: why conservation monitoring programs need a plan for action. *Frontiers in Ecology and the Environment* **11**(10), 549–555.
- NSW DPI. (2017). Priorities Action Statement Draft Actions for Stocky Galaxias. NSW Department of Primary Industries, Crows Nest. Available at: <u>https://www.dpi.nsw.gov.au/fishing/threatened-</u> <u>species/what-current/critically-endangered-species/stocky-galaxias/priorities-action-statement-draft-</u> <u>actions-for-stocky-galaxias</u> (accessed 14 May 2021).
- NSW FSC (Fisheries Scientific Committee). (2019). CAM Assessment Galaxias tantangara. NSW Department of Primary Industries, Crows Nest.
- Pollard, D.A. (1972). The biology of a landlocked form of the normally catadromous salmoniform fish Galaxias maculatus (Jenyns). III. Structure of the gonads. *Australian Journal of Marine and Freshwater Research* **23**(1), 17–38.
- Pottier, G., Beaumont, W.R., Marchand, F., Le Bail, P-Y., Azam, D., Rives, J., Vigouroux, R. and Roussel, J-M. (2019). Electrofishing in streams of low water conductivity but high biodiversity value: Challenges, limits and perspectives. *Fisheries Management and Ecology* 27(1), 52–63.
- Raadik, T.A. (1993). A research recovery plan for the Barred Galaxias, Galaxias fuscus Mack, 1936, in south-eastern Australia. Report to the Australian National Parks and Wildlife Service, Canberra. Department of Conservation and Natural Resources, Victoria.
- Raadik, T.A. (1995). A research recovery plan for the Barred Galaxias, Galaxias fuscus Mack, 1936, in south-eastern Australia. Flora and Fauna Technical Report No. 141, Department of Conservation and Natural Resources, Victoria.
- Raadik, T.A. (2002). Barred Galaxias recovery project 13695 final report, Endangered Species Program. Report to Environment Australia, Canberra. Freshwater Ecology, Arthur Rylah Institute, Victoria
- Raadik, T.A. (2009a). *Barred Galaxias*, Galaxias fuscus, *recovery actions: predator detection and control summary report 2008–2009.* Client report to Goulburn–Broken Catchment management Authority. Arthur Rylah Institute for Environmental Research, Heidelberg, Victoria.

- Raadik, T.A. (2009b). *Barred Galaxias,* Galaxias fuscus, *recovery actions: fire and drought impacts summary report 2007–2008.* Client report to Goulburn–Broken Catchment management Authority. Arthur Rylah Institute for Environmental Research, Heidelberg, Victoria.
- Raadik, T.A. (2011). Systematic revision of the Mountain *Galaxias olidus* Günther, 1866 species complex (Teleostei: Galaxiidae) in eastern Australia. PhD Thesis, University of Canberra, Canberra.
- Raadik, T.A. (2012). Fact File: Conserving the threatened Shaw Galaxias. Critical Conservation Actions in the Alpine National Park. Arthur Rylah Institute for Environmental Research, and Parks Victoria, Heidelberg, Victoria.
- Raadik, T.A. (2014). Fifteen from one: a revision of the *Galaxias olidus* Günther, 1866 complex (Teleostei, Galaxiidae) in south-eastern Australia recognises three previously described taxa and describes 12 new species. *Zootaxa* **3898**, 1–198.
- Raadik, T.A. (2018). *Identification of galaxiid species (Telostei, Galaxiidae) from the area of the proposed Snowy 2.0 Project*. Unpublished client report to EMM Consulting Pty Ltd. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg, Victoria.
- Raadik, T.A. (2019a). Recovery actions for seven endemic and threatened Victorian galaxiid species. Biodiversity On-ground Actions Regional Partnerships and Targeted Actions Project 2017–18. Published Fact Sheet. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg.
- Raadik, T.A. (2019b). Conservation of endemic and threatened Victorian galaxiid species. Biodiversity Response Planning Projects 118, 119, 120 & 121 2018–2020. Published Fact Sheet. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg.
- Raadik, T. and Clunie, P. (2007). Emergency responses in Victoria. Case studies of fish emergency drought/fire responses in Victoria. p. 45. In, Guzman, I., Higham, J. and Hall, A. 2007. Native Fish Emergency Response Workshop. Summary, abstract and recommendations. Proceedings of a workshop held in Adelaide SA 21st–22nd November 2007. Department of Environment and Natural Resources, Adelaide.
- Raadik, T.A., Fairbrother, P.S. and Smith, S.J. (2010). *National recovery plan for the Barred Galaxias* (Galaxias fuscus). Department of Sustainability and Environment, Heidelberg, Victoria. Available from: <u>http://www.environment.gov.au/cgi-</u> bin/sprat/public/publicspecies.pl?taxon_id=26168#recovery_plan_loop
- Raadik, T.A. and Lintermans, M. (2022). *Stocky Galaxias review of existing information, Snowy 2.0.* Published client report for Snowy Hydro Ltd, Cooma. Arthur Rylah Institute for Environmental Research, Department of Environment, Land, Water and Planning, Heidelberg, Victoria.
- Raadik, T.A. and Nicol, M.D. (2013). Searching for threatened upland galaxiids (Teleostei, Galaxiidae) in the Thomson and La Trobe river catchments, West Gippsland, Victoria. Arthur Rylah Institute for Environmental Research Technical Report 248. Department of Environment and Natural Resources, Heidelberg, Victoria.
- Raadik, T.A. and Stoessel, D.J. (2022). *Stocky Galaxias translocation strategy, Snowy 2.0*. Published client report to Snowy Hydro Ltd, Cooma. Arthur Rylah Institute for Environmental Research, Department of Environemt, Land, Water and Planning, Heidelberg, Victoria.
- Radford, J., Wager, R. and Kerezsy, A. (2018).Recovery of the red-finned blue-eye: informing action in the absence of controls and replication. In: Legge, S., Lindenmayer, D., Robinson, N.M., Scheele, B.C., Southwell, D.M. and Wintle, B. (Eds.) *Monitoring Threatened Species and Ecological Communities*, pp. 375–385. CSIRO Publishing, Clayton South, Victoria.
- Reynolds, J.B. (2016). Spheres, rings, and rods as electrodes in electrofishing: Their effects on system resistance and electrical fields. *Transactions of the American Fisheries Society* **145**, 239–248.
- Stoessel, D.J., Ayres, R.M. and Raadik, T.A. (2012). Improving spawning success for Barred Galaxias (Galaxias fuscus) in streams affected by bushfire an aid to recovery. Black Saturday Victoria 2009 Natural values fire recovery program. Arthur Rylah Institute for Environmental Research, Department of Sustainability and Environment, Heidelberg, Victoria.
- Stoessel, D.J., Raadik, T.A. and Ayres, R.M. (2015). Spawning of threatened barred galaxias, *Galaxias fuscus* (Teleostei: Galaxiidae). *Proceedings of the Linnean Society of New South Wales* **137**, 1–6.

- Stoessel, D.J., Raadik, T.A., Nicol, M.D., Fairbrother, P.S. and Campbell-Beschorner, R. (2020). Captive breeding of two rare non-migratory galaxiids (Teleostei, Galaxiidae) for species conservation. *Proceedings of the Royal Society of Victoria* **131**, 42–48.
- Tonkin, Z., Kearns, J., Lyon, J., Balcombe, S.R., King, A.J., and Bond, N.R. (2017). Regional-scale extremes in river discharge and localised spawning stock abundance influence recruitment dynamics of a threatened freshwater fish. *Ecohydrology* **10**(6), e1842.
- TSS (Threatened Species Section) (2006). *Recovery Plan: Tasmanian Galaxiidae 2006–2010*. Department of Primary Industries, Water, Hobart.
- TSSC (Threatened Species Scientific Committee) (2021). *Conservation Advice* Galaxias tantangara *Stocky Galaxias*. Department of Agriculture, Water and the Environment, Canberra. Available from: <u>http://www.environment.gov.au/biodiversity/threatened/species/pubs/87879-conservation-advice-03032021</u>

www.ari.vic.gov.au

www.delwp.vic.gov.au