

# Stocky Galaxias – catchment survey, Snowy 2.0

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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.



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**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).

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# Stocky Galaxias – catchment survey, Snowy 2.0

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

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remnant populations

## 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 provides an outline for a catchment survey for Stocky Galaxias (*Galaxias tantangara*). It sets out objectives and potential activities to be undertaken to identify additional, remnant populations of Stocky Galaxias, and identify potential translocation sites, and their status/suitability, a key conservation action for the species. As such, its value and relevance will extend beyond the Snowy 2.0 Management Plans.

Stocky Galaxias are known from a single population (Raadik 2014; Raadik and Lintermans 2022a,b), and very little, intensive surveying for it has been undertaken across the broader landscape (Raadik and Lintermans 2022a). Consequently, a key early action, fundamental to mitigate the high risk of extinction of a single population species (Primak 2017; Brown et al. 2022), involves searching for additional, extant populations, and locating potential translocation sites.

Comprehensive distributional survey is of high importance in the conservation of galaxiids in Australia and is often undertaken once known threatened species populations are stabilised and secured from threats (Raadik et al. 2010; Raadik 2015). This is particularly so for species which exist as a single population, or a small number of populations in close geographic proximity to each other (e.g. Barred Galaxias: Raadik et al. 2010). Once the number and spread of extant populations and potential translocation sites are well known, recovery actions based on the available pool of populations, individuals, and genetic diversity, including options to spread extinction risk, can be developed.

A catchment survey for a threatened, very range-restricted, fish species needs to have high detection probability to ensure a high level of confidence in the survey results. It should therefore focus on areas with high probability of occupancy and include an appropriate density of survey sites and use of appropriate sampling techniques to detect small, fragmented, isolated remaining populations, if present.

Moreover, a catchment survey must be adaptive as it is difficult to predict the outcomes. As such a catchment survey must be designed to be adaptable to incorporate results as they are known while maintaining the ability to address the survey objectives.

Here we develop a catchment survey adapted to the headwaters of the Murrumbidgee River, based on relevant literature, including results and knowledge gained from undertaking catchment surveys for rare, threatened galaxiids in Victoria since the early 1990s as part of galaxiid recovery plans (Raadik 1995, 2002; Raadik et al. 2009, 2010; Ayres et al. 2012; Raadik and Nicol 2013; Raadik 2015, 2019a). Similar methodology is also currently being used as part of the conservation management of seven additional, narrow range, threatened galaxiid species in eastern Gippsland, Victoria (Raadik 2018a; 2019 a,b).

The success and value of the catchment survey is not the number of additional remnant populations of Stocky Galaxias, or potential translocation sites, found, but the increased confidence with which refinement of the conservation status of Stocky Galaxias can be made based on new, accurate data. This then allows informed decisions on additional management to be made. A catchment survey is a crucial, necessary step, but the outcomes are unknown: they will either reduce the potential complexity of recovery actions for Stocky Galaxias (more known populations or translocation sites) or maintain the status quo (no additional populations or translocation sites).

### 1.1 Relevance to priority conservation actions

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

- Survey similar habitat areas to Tantangara Creek to determine if other populations of Stocky Galaxias occur (High priority).
- Identify potential candidate sites for possible future translocation of Stocky Galaxias (Medium priority)
- Investigate distribution, habitat and movements (Medium priority).

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

- Further survey work to locate potential trout-free sites for future translocation.
- Broadscale fish survey work in upper Murrumbidgee catchment to locate additional populations.
- Identification of streams suitable for trout barrier installation (or augmentation)

## 2 Catchment survey aim and objectives

The overall aim of the catchment survey is:

• To improve knowledge on the distribution (presence/absence) and potentially suitable habitat of Stocky Galaxias, 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 do this first requires the status of these metrics for the species to be known, and this can only be undertaken by an appropriate level of survey within its predicted former range. Once this is undertaken, and there is a high level of confidence in the number of extant populations available for recovery activities, decisions can be made on whether to focus recovery effort on these populations, or a mix of extant populations and translocations to establish addition populations. Therefore, knowledge on available, potentially suitable, translocation sites is invaluable, and can be generated when searching for extant populations.

Consequently, the specific catchment survey objectives for Stocky Galaxias, to meet the overall aim, are to:

- 1. Locate any additional Stocky Galaxias populations.
- 2. Identify potential future Stocky Galaxias translocation sites.

### 2.1 Design considerations

The distribution of Stocky Galaxias has been fragmented and reduced by predatory trout (Raadik 2014; Raadik and Lintermans 2022a), which occurred before the species was discovered. Consequently, the former range of the species is poorly known, and potential extant populations are likely to be found in sections of catchments where trout have been unable to colonise, usually above a natural waterfall. These areas are likely to be in the upper reaches of smaller systems, in relatively short stream reaches and therefore small populations, and may be spread widely across the landscape depending on the abundance and location of effective instream barriers. This scenario has been found previously with other, similar, species of threatened upland galaxiids affected by predatory trout (Raadik et al. 2010, Raadik, 2014).

The factors which drive the number, or density, of potential survey sites is a compromise between maintaining a high probability of detection of valuable remnant populations or potential translocation sites, and cost. However, a key driver is to improve understanding of the status of Stocky Galaxias to prioritise conservation actions and effort, and this can only be undertaken by an accurate assessment of the number of populations that remain, and if potential translocations site for establishment of new populations are available. Therefore, to achieve a high level of detection probability in mountainous terrain requires undertaking survey at a high spatial density to minimise missing small, isolated populations, leading to a greater overall accuracy in presence/absence data.

A further consideration, which concerns the conservation value of a site with respect to spreading extinction risk, is the geographical spread of sites from other valuable sites to avoid impacts from a large disturbance (see above). Therefore maximising spacing between potentially valuable locations, in the absence of knowing which sites will be important, will require sampling widely spaced sites.

### 3 Catchment survey tasks and activities

The catchment survey design comprises three main components (Figure 1), consisting of:

- 1. Desktop prioritisation of sites.
- 2. Initial 'rapid' survey.
- 3. Detailed survey of identified galaxiid or potential translocation sites.



#### Figure 1. Flow chart of Stocky Galaxias catchment survey tasks and summarised activities

The initial 'rapid' survey of prioritised sites (Task 2) would be expected to be completed in a single season. However, if no additional populations of Stocky Galaxias, or potentially suitable translocations sites, are found, consideration should be given to a second round of survey of additional lower priority sites that were not included in the original survey.

### 3.1 Task 1. Desktop prioritisation of sites

This task will provide a prioritised list of potential survey sites, selected to maximise the discovery of extant populations of Stocky Galaxias and sites potentially suitable for translocations. It will be undertaken using spatial analysis of the digital stream network, constrained by a suite of topographic criteria reflecting suitability for Stocky Galaxias, and further refined by additional biological and conservation recovery criteria, to develop a list of priority stream reaches. This will be accompanied by a manual feed-back process to ensure that analysis outputs are logical and do not exclude known important features (i.e. waterfalls). Consequently, multiple runs of analyses may need to be undertaken, with varying upper and lower bounds, for important criteria. A manual process will then be used to define sampling sites within priority reaches.

Whilst larger, and well-known waterfalls are spatially recorded, many smaller fish movement barriers, such as smaller waterfalls and steep cascade/waterfall sequences, particularly in remote locations, are poorly documented. The location of potential instream barriers can be identified remotely by fine-scale interrogation

of available LiDAR (Light Detecting and Ranging) data, or remotely from aerial and satellite imagery (e.g. Google Earth layers, aerial photography) if tree cover is sparse and the stream channel is relatively clearly visible, though this becomes more difficult for smaller streams due to increasing canopy cover, and if pixel resolution is low. Additional potential locations can be gathered by visually inspecting digital topographic maps at small scale, to locate steep gradients or elevation changes on the stream network (Enqvist 2019), often with abrupt changes in stream direction possibly due to differing geological substrates. This can also be informed by analysis of fine level (1–2 m interval) digital terrain models (Enqvist 2019), however, as some fish barriers can be quite small, they may not be detectable. Therefore, analysis of airborne LiDAR and high spatial resolution image data, if available, should also be undertaken to identify reaches potentially with waterfalls (areas of steep gradient or water surface discontinuity).

Given that little is known regarding the historical distribution of Stocky Galaxias before trout were introduced, and that only one population is known, the proposed suitability criteria are relatively simplistic and coarse. However, they are framed by the same assumptions as used for the detection of remnant populations, and un-occupied suitable habitat, of similar, upland, threatened galaxiids in the Mountain Galaxias complex in Victoria (Raadik 1995; Raadik et al. 2010; Ayres et al. 2012; Raadik 2019a).

### 3.1.1 Survey area

An initial important consideration is defining the broad survey area. This is the area with the highest probability of containing remnant, isolated populations of Stocky Galaxias, which would conform to its expected/predicted historical distribution, before trout colonisation pre 1900s. As with other similar species of upland galaxiids, Stocky Galaxias appear to be unable to persist in the presence of predators (trout species) (Tilzey 1976; Lintermans 2000, Raadik 2014; Lintermans and Allan 2019), and therefore, they will persist in areas where trout have been unable to invade, and where sufficient environmental conditions exists to sustain fish (i.e. upper Tantangara Creek). The former range of Stocky Galaxias is difficult to define due to lack of historical information, however, the species is confidentially presumed to have been more widespread, at least in the Murrumbidgee River headwaters (Raadik 2014; NSW FSC 2016), and highly likely extended into nearby catchments outside of this area (Raadik and Lintermans 2022a).

Further, an important factor in reducing overall extinction risk for a species is by ensuring populations are sufficiently spread across the landscape to reduce the probability of a stochastic event equally impacting each (i.e. separate 'locations' sensu IUCN Red List criteria: IUCN 2019). As the area of the Murrumbidgee River catchment, upstream of Tantangara Reservoir, is relatively small (about 30 km wide x 15 km long), even if additional remnant populations, or potentially suitable translocation sites, are found in this area, they may still lack the geographical spread to avoid impacts from large disturbances such as fire or drought.

Consequently, areas for consideration include the Murrumbidgee River headwaters, where Stocky Galaxias is still extant, headwater sections of nearby catchments (Yarrangobilly, Goobarragandra, Goodradigbee, Eucumbene and Tumut), and a section of the Murrumbidgee catchment downstream of Tantangara Reservoir (Figure 2). The headwaters of the Cotter River may also be of value to include, as this area also abuts the Murrumbidgee River catchment. This area can then be refined further to a 'Target Survey Area', based on a range of coarse analysis criteria.

### 3.1.2 Prioritisation criteria

Not all stream reaches within the broad catchment survey area, defined above (area outlined by grey dashed line, Figure 2) ,will be, or have been, suitable for Stocky Galaxias. This is primarily due to the species potentially not having been historically present in some areas as it is considered to have occupied a high elevation, relatively restricted range (Raadik 2014; Raadik and Lintermans 2022a). Consequently, an elevational boundary can be set below which the stream network is excluded from analysis. The lowest elevation Stocky Galaxias is currently found at is 1360 m (Raadik 2014; Raadik and Lintermans 2022a), and it is not inconceivable that it had a broader elevational range, extending to lower elevations. The Dargo Galaxias (*Galaxias mungadhan*) from Victoria, previously only found at a similar elevation to Stocky Galaxias (Raadik 2014), has now been found during a catchment survey for remnant populations to occur down to about 950 m elevation (T. Raadik, unpublished data). Consequently, we recommend a conservative elevation range for Stocky Galaxias of 1000 m and above.



Figure 2. Headwater catchment of the Murrumbidgee River showing the current distribution of Stocky Galaxias (short yellow curved line), including headwaters of nearby river systems which may contain remnant populations.

Black dashed lines represent catchment boundaries, grey dashed line represents approximate broad area for Stocky Galaxias catchment survey.

An additional reason for unsuitable habitat for Stocky Galaxias is the contemporary presence of predatory trout which now occupy much of the aquatic habitat in the region, though their presence in many smaller tributaries, is poorly understood. As trout are known to be present in larger streams (e.g. > 6 m wide), the focal area for Stocky Galaxias persistence will be headwater areas where streams are narrower, most often above a natural instream barrier, such as a waterfall (Finlayson et al. 2005; Tweddle et al. 2009). These structures prevent upstream trout incursion and are essential to protect threatened fish species impacted by predators. Therefore, remnant Stocky Galaxias populations may persist above instream barriers, and in smaller headwaters streams, as instream barriers can be more common in these smaller, steeper catchments.

Based on the above, two criteria can be defined based on predators; 1) excluding stream reaches where trout have been detected, or larger streams due to the very high probability of trout presence (= no Stocky Galaxias), and 2) trout absence (= possibly Stocky Galaxias presence), either from survey data, or inferred from metrics which predict the possible presence of a waterfall. Where data exists on the presence of trout, the entire stream, including a buffered lower reach of all tributaries (e.g. 200–400 m upstream), downstream from the most upper detection point, can be excluded from the analysis.

Further, the very upper reaches of a stream network are usually only seasonally wet, unless permanent springs are present. These reaches usually conform to 1st order streams (at a scale of 1 : 25,000) depending on drainage density (Strahler 1957, Gordon et al. 2004), and in the upper Murrumbidgee River catchment may be approximately up to 2 km in length depending on catchment steepness (T. Raadik, pers. observation). However, if sites are sampled further downstream and permanent water is detected but no fish found, the longer 1st order tributaries at higher elevation locations may have their priority for sampling increased.

Stream order can also be used to further refine reaches of the stream network for exclusion which have a very high probability of containing trout (see above). We recommend excluding larger streams with an order of 4–6. We provide this range as stream width/depth can vary greatly within and between stream order sizes, and the influence of a particular stream order on the prioritisation should be tested during the analysis.

### 3.1.3 Prioritisation

The selection of stream survey sites is a three-step process, commencing with the reduction of the stream network based on various criteria (prioritisation step 1) to produce a target stream network in which all streams could potentially be sampled to meet project objectives. This constrained stream network is then split into segments found upstream or downstream of known or potentially present, instream barriers (waterfalls) (prioritisation step 2), and each segment divided into shorter reaches, and prioritised again based on criteria related to barriers and upstream habitat.

Following prioritisation, the location of sampling sites within reaches will be determined by a manual process (see further below)

#### Stream network

Based on the discussion above, the following criteria are proposed to be used to constrain the stream network in the broad catchment survey area (Figure 2) for reach prioritisation analysis:

- 1. Exclude areas below 1000 m elevational contour.
- 2. Exclude 1st and > 5th order streams (at 1 : 25,000 scale).
- 3. Exclude areas cleared for agriculture.
- 4. Exclude streams known to contain trout (Brown Trout Salmo trutta, Rainbow Trout Oncorhynchus mykiss, Brook Trout Salvelinus fontinalis).

Criteria 1 to 3 will reduce the available stream network substantially, excluding any disturbed areas (cleared land), low elevation areas where Stocky Galaxias is highly likely to not have been historically present, areas with high probability of trout presence, and intermittent (e.g. potentially drying) headwater reaches. Criterion 4 further constrains the network by excluding streams known to contain trout (all stream sections downstream of the detection point).

However, the 2019/20 bushfires have removed trout from many previously occupied headwater streams and natural barriers may prevent their recolonisation (M. Lintermans unpublished data), meaning they may now be potentially suitable as translocation sites. Therefore, 2nd–5th order streams containing historical records for trout which were burnt in the 2019–20 fires should be considered for inclusion in the target stream network.

At this point, the priority stream network has been reduced based on criteria with relatively high confidence and will be difficult to be reduced further based on available criteria or additional suitable data. This now represents the target stream network for the catchment survey, but as highlighted above, should be manually assessed to see if it is logical (i.e. that too much or too little network has been removed) and against the following two broad criteria, and consideration to given to re-running analyses with differing limiting values for important criteria if needed:

- Spatially spread across the target survey area.
- Multiple stream systems and adjacent streams are selected.

These criteria relate to ensuring an adequate survey density to minimise missing small, remnant populations or suitable habitat, and, from a conservation perspective, to maximising spread of populations or translocation sites to reduce extinction risk.

Once the target stream network is defined, further spatial analysis will prioritise the network based on the presence of known, or potential, instream barriers (waterfalls or large rapids). Sections upstream of instream barriers having highest priority, and those with no instream barrier downstream have the lowest priority.

Potential predictors of barrier presence are very narrow, incised valleys (e.g. < 20 m wide and > 5 m deep), and catchment steepness/stream gradient (e.g. drop of 10 m over a distance of 50 m, 5 m over 20 m distance, etc.). Spatial analysis should therefore include known barrier locations and undertake digital terrain model analysis to identify narrow valley constriction and areas of short and very steep stream gradients.

#### Stream reach

The stream network can now be divided into shorter reaches, corresponding to a stream section between tributaries of > 1st stream order. Prioritisation of reaches can then be undertaken based on analysis of the degree of potential protection from predatory trout (presence and number of instream barriers) and the amount of potentially suitable habitat (upstream wetted stream length). A proposed ranking system is presented in Table 1. For example, reaches protected by a downstream barrier are valued more than reaches with a barrier, and reaches protected by more than one downstream barrier are valued more than a reach protected by a single barrier, due to the extra protection from predatory fish incursion more barriers provide. Similarly, a reach with a large total upstream length of potential wetted stream habitat is valued more than reaches with less upstream total stream habitat, due to the potential for larger areas of upstream total habitat to support larger fish populations.

# Table 1. Proposed priority ranking for potential sampling reaches based on the number of instream barriers downstream and upstream wetted total stream length.

Criteria: scores in parentheses; above barrier – number of instream barriers the reach is above; stream length upstream – estimated length potentially habitable by fish to top of catchment; scores are additive. Priority: 1 = 6-7, 2 = 4-5, 3 = < 4.

Above barrier	Stream length upstream (km)	Total score	Priority
>1 barrier	> 4 (3)	7	1
(4)	2–4 (2)	6	1
	> 2 (1)	5	2
1 barrier	> 4 (3)	6	1
(3)	2–4 (2)	5	2
	> 2 (1)	4	2
No barrier	> 4 (3)	4	2
(1)	2–4 (2)	3	3
	> 2 (1)	2	3

#### Sites

The priority survey sites are locations within the prioritised reaches scale (see above). The general location of a site, and number of sites, to be sampled within a priority reach needs to be determined by a manual desktop process which includes of assessment of finer-scale landscape features such as reach slope, valley

constrictions, sharp changes in stream direction, distance upstream of potential barriers, presence of uplands plains, etc., and access issues. It should also include consideration of the length of sample reach and appropriate spacing for sampling sites to meet project objectives. Finally, the position of specific survey sites may need to be adjusted further, if necessary due to local conditions or features, during the field survey.

### 3.2 Task 2. Initial 'rapid' survey

The primary aim of this stage of investigations is to undertake a field investigation of the prioritised reaches from Task 1 to identify locations which contain galaxiids or could be of value as future translocation sites. To enable survey of many sites, an initial, rapid survey protocol will be followed, with galaxiid, or valuable, sites revisited in Task 3 for more detailed assessment. At each site, the initial survey will undertake aquatic fauna sampling, and general, qualitative assessment of instream habitat availability, water permanency, and possible presence of predator barriers.

The summarised survey activities are provided in Figure 1 (above). As this survey is a relatively rapid initial assessment, it is assumed that if there are more than one priority site on a stream or tributary, that they will be sampled in an upstream order. This potentially negates the need for rapid assessment of additional sites in reaches further upstream of Stocky Galaxias detection sites; these will be revisited during Task 3.

**Purpose:** Initial identification of prioritised reaches to detect the presence of galaxiids or potential translocation sites.

**Value:** A relatively rapid assessment of a large number of priority sites to identify those of value (contain galaxiids or potentially suitable as translocation sites) which can then be assessed in greater detail in follow-up sampling.

Timing: Summer to autumn (February to May)

**Method:** Undertaken at multiple, prioritised reaches and sites across the target survey area, using a combination of eDNA sampling and physical sampling using backpack electrofishing. Sampling for fish along a reach at each site, fish collected are identified, counted, measured for length (mm, length to caudal fork), weighed (grams), visually assessed for parasites or disease, returned alive to site of capture, water quality parameters measure, stream habitat characteristics visually assessed and a broader reach visually assessed for presence of instream barriers or a potential location to construct a barrier. Positive results will lead to Task 3 (below).

Analysis and reporting: Following survey.

### 3.2.1 Methods

The details of activities undertaken at each site are listed below. Note that physical sampling for target fish, along with remote sensing using DNA analysis are considered complementary activities as the target species DNA sensing in water requires further refinement (see below).

Considering these two survey methods, and that most sites may need to be accessed by hiking, the survey team (2 people) must be able to safely carry the sampling, preserving, water quality and communication equipment required, including water, food, and first aid equipment (including defibrillator). An additional requirement is to carry enough batteries for the planned sampling, including a spare. Consequently, this will affect the choice of equipment specified below.

#### Implementation timing

The optimal time for the field component of the catchment survey is when detection probability for target fauna is maximised (Raadik et al. 2010). For trout and galaxiids, this coincides with the time of lowest stream flows, as sampling efficiency is increased due to decreased water depth and width, reduced flow velocities and turbulence, and larval galaxiids and juvenile trout have increased to a sufficient size to be more easily detectable. These conditions can be met in the project area from about February to May. Ability to access montane streams is hampered by snow cover, or considerations about damage to access trails, from late May to early October.

Low water levels also enable qualitative assessment of the presumed minimum habitat quality and quantity available for Stocky Galaxias, and water permanency, during the warmer months of the year.

### Fish sampling

Physical fish sampling is to be undertaken using back-pack electrofishing by a team of two staff qualified in, and experienced with, electrofishing in smaller freshwater streams. Remote sensing of fish is to be

undertaken using collection of environmental DNA samples at the same time from each site for later analysis in a laboratory (see below).

**Permits.** Relevant permits to undertake the catchment survey will need to be organised well in advance. These are, but may not be restricted to, the following.

- <u>Scientific Collection Permit</u> authorises the taking and possession of fish for the purpose of research, under section 37 of the NSW *Fisheries Management Act 1994*, and threatened species under the *Biodiversity Conservation Act 2016*. Available from the NSW Department of Primary Industries.
- <u>Scientific Licence</u> authorises research in the National Parks and Wildlife Service reserve system, authorised under section 132C of the NSW National Parks and Wildlife Act 1974. Available from the NSW Office of Environment & Heritage.
- <u>Animal ethics approval</u> for sampling of fish, and collection of voucher material (either NSW or institutional).

**Electrofishing.** Backpack electrofishing is the recommended method for fish sampling as it is an efficient, non-destructive method for use in upland, smaller streams, and is efficient for galaxiids and trout species. 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 frequency between 90–100 Hz 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, maximising the area of the cathode and minimising 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 (about 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.

Electrofishing should be undertaken as single-pass, sampling along a stream reach (see below) at a sampling site. 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. The specifics of the sampling reach and metrics to record follow.

Sampling reach: maximum 200 m reach in streams < 2 m wide; maximum 300 m reach in stream > 2 m wide. Sampling within a reach should commence as continuous, covering all areas of the stream length upstream, or shift to spot fishing (sampling optimal habitats/locations), or a mix of both further upstream.

Sampling metrics to record: GPS position of start and end locations, start and end time, electrofisher poweron time (seconds), electrofisher settings (voltage, frequency, % of range), reach length (m).

From an efficiency perspective, fish sampling will need to be adaptive and may therefore differ between sites, based on the number of individuals encountered as sampling progresses along a reach, to avoid unnecessary sampling, and to save time. This will not affect data analysis, as sites will only be compared qualitatively. Consequently, sampling may vary in reach length sampled and/or time spent sampling if the sampling objectives are met. The decision for, and details of, the variation in sampling, along with the associated length or stream area details, must be recorded on the site datasheet.

Some examples follow of situations where fish sampling may need adjustment.

- A moderate to high abundance (30–50 individuals) of galaxiids is encountered in the first 100 m of the sampling reach (juveniles < 60 mm in length may also be present), indicating a large galaxiid population and lack of predators. Sampling can cease once sufficient voucher specimens and genetic samples have been collected (see details in 3.3.4), as the site can be further investigated in Task 3.
- A high abundance of trout (~>20 individuals in < 2 m wide stream; >30 in a >2 m wide stream) is encountered in the first 100 m or 150 m respectively of sampling and no galaxiids are detected, and the remaining stream reach appears similar in morphology, flow characteristics, and instream habitat structure, the probability of galaxiid presence is potentially very low. Therefore, sampling of the

remaining reach may be stopped as there is high probability that galaxiids do not exist, and due to the high abundance of trout, this location may be too costly to rehabilitate for use in translocations.

- Very few fish (<10 total), all salmonids, have been detected during continuous sampling along the first 50 m a reach, indicating low fish abundance. Sampling may need to shift to spot sampling in suitable fish habitat patches (e.g. deeper pools, undercut banks, areas of structural habitat, riffles, stream edges, etc.) to maximise the probability of detecting additional individuals or species, and as above, the entire remaining reach may not need to be sampled. If there are no trout and the fish are galaxiids, proceed with normal site sampling protocol.</li>
- All target species (e.g. trout and galaxiids) are detected in the first 100 m of the sampling reach, and the remaining stream reach appears similar in morphology, flow characteristics, and instream habitat structure. Therefore, if enough individuals are available for measuring and vouchering, and the relative abundance of species is considered unlikely to alter over the remaining reach, sampling may be stopped.

Beside the examples above, there are many other nuanced decisions which can be made regarding altering sampling length/effort, based on the site and fauna characteristics. These, along with the knowledge of suitable stream habitat to specifically target for galaxiids, can be made with confidence by electrofishing operators experienced in sampling for galaxiids in upland streams.

#### 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. Handling of fish must be done with wet hands.

All fish species captured will be identified and the number of individuals counted, including any seen but not captured. Specific details for particular species follow:

- Galaxiids:
  - i. Inspect galaxiids carefully and separate specimens which differ in colour or morphology (see voucher material below).
  - ii. Measure length (mm) and weight (0.1 g) of a subset of individuals per species or colour/morphology (about 20), including observation of deformities, disease, damage (e.g. bird strike) or external parasites (e.g. *Lernaea cyprinacea*).
- Trout: measure length (mm) of smallest to largest individuals per species and estimate approximate of average length.
- Crayfish: whilst not a target species, record abundance of each species collected.
- Other fish species: if additional species (other than those above) are collected, identity each species and count individuals collected. If non-native to the catchment, preserve 1 or 2 individuals in 70% ethanol and retain as vouchers. Any species considered to be pests are to be euthanised, rather than returned to the water in accordance with relevant permits.

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

#### Voucher specimen collection and identification.

Due to the morphological similarity between Stocky Galaxias and other species in the Mountain Galaxias complex (Raadik 2014), any individuals of galaxiids located during the initial survey work must be carefully identified to avoid misidentification leading to false positive or negative detections. Expertise in the identification of galaxiids in the Mountain Galaxias species complex (which includes Stocky Galaxias) is very limited, and usually requires detailed examination of voucher material (Raadik 2014). Therefore, as undertaken previously (Raadik 2018b), this will require the collection of appropriately formalin-fixed material for detailed morphological analysis, and retention of genetic tissue (as fin clips) for genetic analysis to support morphological outcomes.

Stocky Galaxias potentially co-occur with Mountain Galaxias (Raadik 2018b; Lintermans, Raadik and Unmack unpublished confidential data), and there is the possibility of additional, undiscovered, species in the Mountain Galaxias complex also present in this area of the Snowy Mountains (Raadik 2014). Therefore, if

galaxiids are encountered when sampling a site, they may comprise a single taxon or a mix of taxa, including or excluding Stocky Galaxias.

Consequently, the following steps and actions are proposed if galaxiids are recorded at a survey site:

- 1. Carefully visually observe all galaxiids collected, looking for differences in body pattern, colour, head or mouth shape, position of fins, etc., which may signify different groups).
- 2. If visual examination indicates more than one group or species, separate the individuals into groups.
- 3. For each different group of galaxiids (except for Climbing Galaxias, *Galaxias brevipinnis*) undertake the following steps for vouchering:
  - a. Depending on fish numbers, select 5–10 representative individuals (e.g. 2–4 large, 2–6 average length). If < 20 fish are collected from the sampling reach, select only 5 individuals and undertake steps b–f (below); if <10 fish are collected only undertake b–cii (below) and release all fish alive. If genetic analysis confirms Stocky Galaxias or Mountain Galaxias, that site will be revisited during Task 3 and preserved vouchers may be able to be retained if the population is found to be larger.</p>
  - b. Take clear digital images of the most representative fish (on their side) to illustrate colour pattern and morphology.
  - c. For each individual selected fish undertake the following and record all details onto the site datasheet:
    - i. measure for length (mm length to caudal fork) and total weight (0.1 g).
    - ii. take a caudal fin clip sample (~ 2 mm square piece from lower or upper caudal lobe), place this into a 2.0 ml polypropylene screw cap (with O-ring) tube (or similar) filled with 100% ethanol and a short, unique sample identification number on waterproof paper)(e.g. TR-383, etc.).
    - iii. Place the specimen into a shallow container of water and add 40 mg/L of Clove oil to euthanise the fish (10 minutes) (repeat with the other individuals in the group).
  - d. Repeat steps a-ciii for all groups of fish, keeping the euthanased fish from each group separate. All fish which require handling to remove genetic tissue will therefore have been processed before the next step, to avoid formalin potentially contaminating tissue for genetic analysis.
  - e. When all groups of galaxiids have been processed, each group can then be formalin-fixed to prevent specimen shrinking or twisting, so that it is suitable for morphological analysis (Raadik 2014). Fish from a group are placed into a 250–300 ml polypropylene, plastic jar with a well-sealing wadded lid and fill with a 10 % solution of formaldehyde in water. A waterproof label containing the date, site code, waterbody name, collector initials and a reference to the group of fish in the jar, should also be place inside the jar.
  - f. After 5 days of immersion, all remaining free formaldehyde can be removed by decanting, soaking, and flushing in freshwater in (about 3–5 events at 0.5-hour intervals), and then specimens can be placed into 70% ethanol for longer term storage.

Genetic samples should be sent to a laboratory for analysis (DArT analysis of SNPs) and comparison against previous genetic data, and the fixed specimens to a taxonomist appropriately experienced in this group of fishes. The process of morphological examination is provided in Raadik (2014, 2018b).

#### Targeted eDNA collection

The initial rapid survey could commence with the collection of site water samples and their analysis for DNA of target species, which is called environmental DNA (eDNA) assessment (Taberlet et al. 2018). This process is a relatively new and evolving technique and relies on the ability to detect short strands of DNA, unique to each target species, from a water sample (Wood et al. 2013; Wilcox et al. 2016; Taberlet et al. 2018). However, many factors can influence the presence and amount of DNA in the water column, and the ability to isolate and detect DNA in the sample, leading to potential false negatives (non-detection when present) or false positives (detection when absent) (Furlan and Gleeson 2016; Hinlo et al. 2017; Taberlet et al. 2018; Stewart 2019).

This can be overcome by developing effective genetic probes which have high sensitivity to the DNA of target species, particularly at very low amounts, as well as specificity, to avoid positive detection of evolutionarily closely related species (i.e. similar DNA) (Furlan et al. 2016; Bylemans et al. 2019; Hinlo et al. 2018). Once a probe is developed, it should be field tested alongside physical sampling to provide a level of

confidence in its detection ability and modified if required to improve. Only once a probe has been optimised can a high level of confidence in results be reached from the sole use of this technique.

Probes have yet to be developed for Stocky Galaxias, or for Mountain Galaxias populations specific to the study area. Multispecies metabarcoding, which can detect Brown Trout, Rainbow trout and Climbing Galaxias, has been undertaken (Cardno 2019; Weeks et al. 2019), however species-specific probes for these species, which would have higher detection and specificity, have not yet been developed.

**eDNA water samples:** Collection of samples should follow the procedures set out by the eDNA service provider considering the desired objective and access considerations. Indicatively this would involve collecting 3 x replicate water samples along a transect across the waterbody at a sampling site. Hiking will constrain the type of water sampling unit used and the amount of water which can be filtered. If sites are easily accessible from a vehicle, filter up to 5 L of water per replicate using an eDNA water filtering machine with self-preserving filters. If hiking, filter as much water as possible using a 60 ml disposable syringe with a canister filter, per replicate. Collected samples are to be uniquely numbered 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 (back at the field vehicle or accommodation) is preferred 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.

#### Site assessment

This component relates to the suitability of the site as fish habitat, and level of security from pest fish (predatory trout) incursion. Once fish sampling is completed, the following should be recorded:

**Sample reach metrics.** Record whether the stream at the site was flowing or still, a continuous wetted reach or isolated pools, and estimate average width, average depth, and maximum depth of the sampled reach.

**General water quality.** Using an appropriate and calibrated multimeter, measure the following water quality parameters at 0.1 m below the surface at one site (flowing stream or continuous wetted reach) or a total of two sites with one in each of two isolated pools: water temperature, electrical conductivity (expressed as at 20°C), dissolved oxygen (mg/L and % saturation), pH (compensating for low salinity water), turbidity (NTU).

If hiking, smaller meters can be used for site measurement of water temperature, electrical conductivity and pH, with a water sampled taken (sample bottle filled to the brim to exclude an airspace), stored in the hiking pack, and analysed back at the field vehicle for the other parameters.

**Water permanency.** Undertake a rapid, objective, and qualitative appraisal of the potential permanency of adequate water at the location. This can be based on the observation of water depth, flow, and connectivity across the sampled reach, depth and substrate of pools, the presence and population structure of fish, potential presence of instream aquatic vegetation which may indicate permanency if known (e.g. some species of bryophytes: Meagher and Fuhrer 2003), or active spring outflow (McCartney et al. 2013).

**Predator barrier.** Undertake a rapid, qualitative assessment at each site of whether an effective, or partially effective, instream barrier is present to prevent upstream movement of predatory fish (e.g. trout species) or a location exists which is conducive to installing a barrier. The assessment may be based on:

- The presence of galaxiids (these may be difficult to accurately identify in the field without taking voucher material (see above).
- The presence of one of more instream barriers such as waterfalls( > 60° from horizontal and greater than 1.5 m high) or cascade/waterfall series (stream gradient > 45°) nearby (up to 100 m downstream), stretching across the width of the valley (e.g. no pathway for upstream fish movement during over-bank flows). Upstream barriers will be identified within the fish sampling reach.
- An absence of trout. This would need to be confirmed that this is from a physical barrier and not from adverse environmental conditions at the site at the time of observation, such as elevated water temperature, low water levels, etc.), which would be undertaken in Task 3.
- Presence of potentially ineffective barriers which may be able to be modified to improve effectiveness for predator exclusion (to be assessed in detail in Task 3).

- Site conditions that may be conducive to the installation of an artificial barrier e.g. a valley constriction and/or steep section of channel, if barriers are not already present.
- The area of suitable habitat above the barrier or potential barrier location.

The presence of trout, however, does not necessarily indicate that a barrier does not exist further downstream. A partially effective barrier to upstream trout invasion may be present but cannot prevent trout movement over all seasons and flow regimes, or, an effective barrier is present, but trout have been deliberately introduced further upstream.

### 3.2.2 Value as translocation sites

Once prioritised reaches and sites are sampled, sites without galaxiids need to be evaluated for their value as potential translocation sites, which are then assessed in greater detail under Task 3.

Primary criteria to evaluate suitability of sites include:

- Degree of water permanency (ability to support fish).
- Area of potentially permanent wetted habitat upstream (e.g. > 1 km of stream length may be suitable to sustain a small population, but areas with > 2 km stream length may sustain larger populations).
- Presence of, or ability to modify existing, or build new, instream barriers to the upstream movement of
  predatory fish (trout).
- Absence of salmonids, as this may indicate the presence of an effective downstream barrier (but see below).

Trout presence at a site, however, does not automatically imply little value as a translocation site, as sites with trout may also have instream barriers (see above), which are either effective or require modification to improve effectiveness. In addition, sites with trout, but no barrier, may contain locations suitable for the construction of artificial barriers. The evaluation of both scenarios needs to include the feasibility of modifying an existing barrier or building a new barrier (appropriate site conditions, distance from access, etc.), including the feasibility to remove all trout from upstream. Whilst the presence of trout would not immediately exclude a site from consideration for translocation, it would mean the site would be given a lower priority due to the additional effort required to eradicate them. The number of potential instream barriers is also important, as multiple barriers decreases the risk of future natural upstream movement of predators if the lower barrier were to fail.

The presence of a healthy population of fish (trout or Mountain Galaxias) also adds to the value of a site as it implies that conditions for fish survival are present. However, a lack of any fish does not necessarily indicate that conditions for survival are lacking. A proposed scenario for galaxiid/trout interactions (Raadik 2017), includes a situation where trout invade and eliminate galaxiids from the catchment, followed by trout being eliminated by low water levels and high-water temperatures which may be lower than the tolerance limit for galaxiids, leaving a fishless stream. Alternatively, trout may have been eliminated from a stream reach due to recent fire impacts, and instream barriers are potentially preventing their recolonisation.

### 3.2.3 Second prioritisation

Once a site has undergone initial sampling (Task 2), it can now be re-prioritised for more detailed assessment based on the presence of Stocky Galaxias, or its value as a potential translocation site for Stocky Galaxias (e.g. upstream stream length and barrier criteria from Table 1). Sites containing Stocky Galaxias will have the highest priority, followed by sites potentially important as translocation sites, with those sites assessed as containing poor habitat or no potential for barrier installation, having the lowest priority (Figure1; Table 2).

Therefore, it is imperative that results of genetic and morphological analysis of any galaxiids collected in Task 2 are known before the second prioritisation can commence.

The highest priority sites are those which contain Stocky Galaxias (priority A). These are followed by sites which contain either Mountain Galaxias, trout or no fish and are further prioritised based on a combination of amount of upstream habitat and presence/absence of instream barriers, with sites with no fish rated lower than those with fish, unless there is acceptable upstream habitat and instream barriers (priority B). Sites with Mountain Galaxias, trout or no fish, with without barriers, or where barriers are unknown, are rated lower (priority C) sites with no barriers and small upstream catchments (Table 2).

# Table 2. Proposed priority ranking for initially sampled sites based on survey results, upstream wetted stream length, and barrier presence/absence (modified from Table 1)

Criteria: scores in parentheses; stream length upstream – estimated length potentially habitable by fish, M - > 2 km, S - < 2 km; Barrier(s) present (Y), partial or unknown (?), absent (N); scores are additive. Priority: A = >14, B = 10–14, C = 7–9, D = <7.

Survey result	Stream length upstream	Barrier(s)	Total score	Priority
Stocky	M (3) (10) S (2)	Y (5)	18	А
Galaxias present (10)		? (3)	16	А
		Y (5)	17	А
		? (3)	15	А
Trout or other	M (3)	Y (5)	11	В
galaxiids <sup>1</sup> ,		? (3)	9	С
present (3)		N (1)	7	С
		Y (5)	9	С
		? (3)	7	С
		N (1)	5	D
No fish (2)	M (3)	Y (5)	10	В
		? (3)	8	С
		N (1)	6	D
	S (1)	Y (5)	8	С
		? (3)	6	D
		N (1)	4	D

Sites with higher priority can proceed to detailed evaluation (Task 3).

Given the expected lack of Climbing Galaxias in nearly all the priority catchments except for some streams in the Eucumbene system (where it is native), the presence or absence of Climbing Galaxias is not included in the prioritisation. However, detection of climbing galaxiids at a site is an important variable to record which would render the site as Priority D, i.e. unsuitable for translocation.

If the rapid survey in Task 2 does not identify remnant Stocky Galaxias populations or sufficient suitable potential translocation sites (e.g. <5), consideration should be given to visiting additional lower priority sites from Task 1 and/or extending the area of survey to additional sections of nearby catchments.

### 3.3 Task 3. Detailed survey of identified sites

The aim of a more detailed survey of re-prioritised sites is to investigate, in more detail, those specific aspects required to confirm their value and priority to Stocky Galaxias, and therefore, further investment (e.g. valuable Stocky Galaxias populations or translocation sites). If multiple populations of Stocky Galaxias are identified (e.g. > 6), the focus should be on assessing these further and protecting them. If no, or limited Stocky Galaxias populations are found, those with Stocky Galaxias should be assessed and protected, but the focus should also include further examination of good habitat sites containing potential barriers, followed by those with good habitat and potential artificial barrier locations with a view to these being used for translocations.

Depending on the focus, the assessments would entail detailed, longer-duration, investigation of a range of specific physical or biological factors at sites, and further downstream or upstream from the reach assessed during Task 2. This would include one or more of the following:

<sup>1</sup> Excludes Climbing Galaxias.

- 1. Status of Stocky Galaxias population (relative abundance/density, evidence of recent recruitment, distribution extent, and potential threats, see Raadik and Lintermans 2021b).
- 2. Extent of habitat, galaxiid abundance and potential threats if only Mountain Galaxias found.
- 3. Abundance and distribution of trout if present.
- 4. Probable cause of lack of fish if none found.
- 5. Degree of security from non-native fish upstream incursion (barrier assessment).
- 6. Potential to modify existing, or construct new instream barriers, if required.

#### 3.3.1 Methods

Methods to achieve the above are as follows.

#### Fish assessment

Addresses point 1, 2 and 3, above. Sampling for galaxiids or trout is to be undertaken by backpack electrofishing (see Fish sampling in 3.2.1 above). Sampling may be required to improve knowledge on *abundance* or *size classes* and can be achieved by sampling additional reaches, as undertaken during Task 2.

To understand the likely *extent* of a population, spot-sampling of short-reaches (e.g. each 10 m in length) for fish detection should be undertaken over a reach of 1 km maximum distance upstream, and downstream, at an appropriate interval, e.g. every 200 m. <u>Downstream</u> sampling can cease a) when the distance is achieved and detections are still occurring, or b) if three consecutive zero detections are achieved in < 1 km distance, or c) if an instream barrier is reached in < 1km distance, downstream and only trout are found on the downstream side (i.e. after sampling up to 20 m on the downstream side). <u>Upstream</u> sampling can cease following criteria a) and b) above.

This assessment can provide information on fish extent up to 2.0 km along the target stream, estimate of fish abundance or data on fish length, and extent of predators or galaxiids, and potential downstream predator barrier locations. The overall nature of the assessment of new Stocky Galaxias populations is the same as that proposed to be undertaken for the species in Tantangara Creek (Raadik and Lintermans 2021b).

#### Coarse habitat and threat assessment

(Addresses 1, 2, 4, above). A visual assessment of habitat (instream habitat quantity and quality) and water permanency are to be undertaken (see Site assessment in 3.2.1 above) to subjectively categorise the location for its quality of instream habitat and water permanency to sustain fish. This may also be evident to some degree by the extent of fish distribution; however, unoccupied upstream habitat may also be available above instream barriers. The assessment should be undertaken during fish sampling, and over the same distance.

It is acknowledged that understanding water permanency may be difficult from a relatively simplistic assessment, due to the complex interaction between groundwater outflow and surface water. Therefore, if water permanency is an outstanding question for potentially valuable sites (i.e. all other criteria are rated highly), consideration should be given to undertaking more detailed investigation (e.g. Cartwright and Morgenstern 2016; Cartwright et al. 2020; Hayashi 2020).

A stream with poor habitat (e.g. shallow, little shading, high abundance of instream sediment, no flow, etc.) and poor potential water permanency, rates lower than one with pools, shading, little sediment, flowing water and evidence of water permanency. Whilst these are coarse habitat features, they reflect general habitat features found at sites occupied by galaxiids (e.g. Tantangara Creek) and are considered to suitable for Stocky Galaxias.

#### Fish absence

Addresses point 4, above. An absence of fish identified in Task 2 must be reconfirmed by additional sampling, following the procedure outlined above under 'Fish assessment'. It is acknowledged that unless a large instream barrier has prevented fish colonisation to the site, or there is evidence of stream drying, identifying a reason for an absence of fish may be difficult. However, undertaking assessment of habitat extent and threat assessment (see above), particularly focussing on the quantity and depth of instream pools, shading, etc. and water persistence, may provide a helpful insight.

A fishless stream reach is rated lower in priority than one with fish due to the potential risk of loss of habitat (i.e. drying), and one with poor habitat (e.g. shallow, little shading, high abundance of instream sediment, etc.), rates lower than one with pools, shading, little sediment.

A lack of fish may also be due to occasional predator incursion, potentially leading to galaxiid loss and subsequent loss of the predators due to random events (e.g. fires, drought, storms, etc.). Therefore, existing downstream barrier assessment (see below), may assist.

#### Existing barrier assessment

Addresses point 4 and 5, above. The degree of security of a location from the upstream incursion of predatory fish is related to the presence of one or more instream barriers (e.g. waterfalls), and their effectiveness in preventing upstream fish movement at all flow levels. This assessment may already have been achieved during Task 2, but if further assessment is required, particularly further upstream or downstream, the same method should be followed (see 'Predator Barrier' under 3.2.1, above). The detection of instream barriers is therefore critical and may need to be undertaken a considerable distance further downstream if no barriers are located within 1 km of the assessment location. Remote sensing for barriers using drones should be given consideration (see below), as it may reduce effort, at least in the initial stage of locating barriers.

The effectiveness of an existing barrier can be indicated by the fish species found occurring upstream. For example, if galaxiids are present upstream in the absence of trout, but the converse is found below the barrier, it can be considered effective. However, if trout are found upstream, this may be due to the barrier being ineffective, partially effective (e.g. at low to moderate flow levels) but not during rare high flow events, or trout have been translocated upstream of the barrier.

Therefore, if trout are present upstream of an instream barrier, a careful assessment is required, focussing on the following attributes (e.g. Easton 2015) and considering varying flow levels from baseflow to overbank flows:

- Barrier type:
  - i. Waterfall can be tiered or stepped (treat as separate units if several falls occur over a distance but are clearly separated by > 50 m distance).
    - plunge vertical and often undercut, water does not touch the bedrock.
    - acute wall angle 40° < 90°, water in contact with the bedrock.
    - slide wall angle 20° < 40°, water in contact with the bedrock.
  - ii. Cascade stepped waterfall of numerous vertical steps.
- Effective height (m).
  - i. Minimum height of the top of the waterfall, measured from the water surface on the downstream side.
  - ii. Overall height of all segments of a cascade.
- Approximate vertical angle of the front face of waterfall.
- Dimensions (length, width, depth (m)) of pool immediately below barrier for waterfalls < 3 m in height.
- Degree of extent of waterfall across the stream channel (full, partial).
- Degree of extent of waterfall across the valley (full from valley side to valley side; partial).
- Slope of sides of waterfall in riparian zone (as an indication of providing a potential pathway for fish passage during periods of flood-flows).
- Description of features of the waterfall face (e.g. smooth, eroded, presence of low flow path, etc).
- Estimated slope of stream downstream of the barrier.

Digital images and sketches (front elevation and plan) of barrier features, including the span of the barrier across the stream channel and valley width, should also be taken to help with interpretation.

Whilst it may be difficult to rate the effectiveness of a barrier following an inspection at one time of year and flow level, the following conditions, alone or in combination, should be taken into consideration.

- Multiple sequential barriers are better than a single barrier.
- Barriers extending across the entire valley width (for narrow valleys) are better than barriers extending across the stream channel.
- Waterfalls are better than cascades.
- High waterfalls are better than lower waterfalls.
- More vertical waterfalls can be better than lower angle waterfalls.

An ideal barrier would be one which was vertical, of reasonable height (3+ m), extended uninterrupted across a narrow valley, did not have a pool immediately below and a steep stream gradient downstream; more than one of these is better. This type of barrier has height to prevent fish jumping over, a lack of a pool below which would allow fish to gain speed with which to jump, sufficient height and downstream stream gradient to minimise potential for drown-out during floods, a narrow valley to reduce floodplain habitat and increase drainage, a sufficiently wide barrier to prevent alternate by-pass pathways to form during overbank/flood flows, and wall steepness to reduce fish pathways up the barrier (e.g. jumping between small pools).

**Drones.** The use of aerial drones for detecting instream barriers, as well as remotely collecting water samples, is in the early stages of development (Lally et al. 2019; Timm et al. 2019; Giordan et al. 2020). A small pilot study to develop the method to detect instream barriers in smaller catchments was recently successfully undertaken in the upper Murrumbidgee catchment (Allan and Lintermans 2021), where a drone was used to successfully locate waterfalls and other potential barriers (e.g. cascades, etc.), and capture data to assess barrier integrity in blocking fish passage (Allan and Lintermans 2021). Whilst one main set of barriers detected was also visible using Google Earth imagery, the value of drone technology is in locating smaller barriers, however, a current drawback is that the pilot needs to be present nearby in the catchment.

Therefore drone technology has some capacity to speed up the process of searching for instream barriers, including defining barrier and site characteristics (e.g. height, reach gradient), as well as potential locations for artificial barrier construction. However, on-site inspection of the highest priority barriers and locations will need to be undertaken to examine finer details.

#### Barrier modification or construction

Addresses point 6, above. Barriers found to be partially or fully ineffective, should be assessed to see if they can be modified to improve their ability to prevent fish passage. The above attributes should be assessed against the attributes for an effective barrier to understand the nature of modification required, and engineering solutions sought. This may entail increasing the height of an existing barrier (or low section on a barrier), removing material from the face of a barrier to increase its vertical angle, removing a control point of a large, deep pool on the downstream side to remove or decrease pool level, removal of large obstructions on the downstream side to increase water flow and drainage during flood events, infilling of low-flow channels adown the barrier face to eliminate potential fish passage pathways, etc.

Where barriers do not exist, an alternative is to construct one or more artificial instream barriers. These can be made of reinforced concrete or other durable materials, and depending on the physical characteristics of the location, may need to be small or large in width and/or height, and either constructed onsite, or prefabricated off-site, delivered and installed. As artificial barriers can be expensive, and difficult to construct in remote areas, optimal site characteristics, which also achieve the necessary barrier characteristics required to be effective in preventing upstream fish passage are needed required to make the feasible. These can include:

- A narrow, steep valley reduces barrier width.
- Relatively steep stream gradient reduces barrier height required.
- Stable substrate such as bedrock.
- Ease of access for construction.

If these were to be optimised, a single artificial barrier may be sufficient. However, the remoteness of much of the terrain in the project area may constrain barrier design (technical feasibility) and construction (access logistics). As such, depending on options and an assessment of the criteria above, an alternative may be to install more than one barrier along a less-optimal reach.

#### Feasibility of predator removal

If predators (trout) are present at a site with good conditions for fish persistence (e.g. effective barriers present, good habitat and water security), the removal of all trout can improve the location to meet the criteria to be considered a potential translocation site for Stocky Galaxias. These sites should be assessed for the feasibility of fish removal, based on the following:

- Area (length x width) of stream to be treated.
- Stream complexity (e.g. single channel, braided channel, multiple tributaries).
- Water depth and complexity of instream habitat (with respect to difficulty of detecting fish/efficacy of removal treatment to be applied).
- Site accessibility (remoteness from vehicle access points).
- The time of year.
- Applicability of mechanical (physical removal via electrofishing) or chemical (piscicide) removal.

A short reach, or short reaches between multiple barriers, of a single stream channel will be easier and more effective to treat than long continuous reaches or complex channels and tributaries. Similarly, effort will increase and likelihood of success will decrease with increasing water depth and habitat complexity, as well as distance from access, particularly to achieve full effectiveness, which is a 100% removal of predatory fish. The time of year relates to fish detection, as fish recruited from the previous breeding period are the largest, and more easily detected, in autumn just before the next breeding season.

The amount of effort can also be influenced by the efficiency of the removal technique (mechanical or chemical), and whether it needs to be repeated. For example, chemical removal can be undertaken efficiently in one treatment if the stream channel is not braided, whereas successive electrofishing runs will need to be undertaken to maximise fish detection and removal, usually to three successive runs of zero detection to be confident (Raadik et al. 2015; Raadik 2017). If removal has been less than 100%, additional removals would need to be undertaken, and any fish left in the system over winter may spawn and increase the number of fish requiring removal the following year; small individuals (< 140 mm in length) are more difficult to detect than older, larger individuals. Each method has its benefits and limitations.

## 4 Links to other activities

The data generated from completion of the catchment survey will be fundamental to other Stocky Galaxias management activities, such as informing aspects of the:

- Species Monitoring Plan number of Stocky Galaxias populations which require monitoring.
- Captive Breeding Activities e.g. need for captive breeding, number and genetic quality of source populations and individuals, conservation breeding protocol, etc.
- Translocation Strategy e.g. where to place captive bred fish, number of wild hatcheries, reinforcement, or translocation sites, establishing new populations, etc.
- Pest Fish Monitoring updated information to a degree on pest fish distribution/abundance in the catchment, though only from sampled locations, potentially inform site selection for pest fish monitoring.

More broadly, catchment survey data may also contribute to Stocky Galaxias population or habitat modelling, which is important to guide translocations and other aspects of conservation of Stocky Galaxias.

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