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1	Assessment of the effects of microPIT tags on the swimming
2	performance of small-bodied and juvenile fish.
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11 Abstract

12

13 Monitoring the movements of fish enables management decisions to be based on the ecological 14 requirements of the species in question. PIT tags are a cost effective, long-term method of tracking large numbers of fish. As this technology has improved, the size of PIT tags has decreased, enabling 15 16 smaller species, and younger fish to be tagged and tracked. There is limited information available 17 on the possible effects that these tags have on the survival rates and subsequent health of small fish. 18 Swimming performance is a physiological measure that is easy to quantify in the lab and directly 19 relates to an individual fishes health. We used swimming performance to assess the effect of 20 microPIT tags (8.4 mm) on five small-bodied, and juveniles of five large growing native Australian 21 fish species. In an initial trial to assess suitability for microPIT tagging, two of the small-bodied 22 species had high mortality and were categorised as unsuitable for tagging. Sample sizes were 23 increased for the remaining eight species to quantify potential effects of microPIT tags on swimming performance. For these eight species we found no significant effects of microPIT tags on 24 25 the swimming their swimming performance.

26 **1. Introduction**

27

28 Monitoring large and small-scale fish movements is vital for assessing fish population dynamics, 29 their utilisation of available resources, and to give insights into how human activities have 30 fragmented or restored connectivity of aquatic systems (Hussey et al., 2015). Temporal and spatial 31 data on fish movement allows fisheries management to reflect the specific requirements of different 32 ecosystems (Cooke et al., 2016). Knowing where different species are found, and when and if they 33 move between locations and habitats is key information for fisheries managers and research. A 34 multitude of methods exist for collecting fish movement data (see review by Cooke et al., 2013), the 35 cost of which generally trades negatively with the complexity of information acquired. Fish may 36 simply be tagged with external metal or plastic tags (Harris, 1988), have their otoliths chemically labelled (Baumgartner, 2016; Cameron et al., 2016; 2011), or be tattooed with a visible implantable 37 38 elastomer (VIE). These methods all provide cost-effective mark and re-capture data (Bangs et al., 39 2013; Hanson and Barron, 2017). At a larger cost, radio (Koehn et al., 2009; Meyer, 2017), 40 acoustic, GPS and satellite tagging provides greater resolution of individual fish movements 41 (Brown et al., 2010; Liss et al., 2016; Lyon et al., 2017), with tags often lasting the lifetime of the 42 fish. Likewise, passive integrated transponders (PIT) tags (Fraiola and Carlson, 2016) provide a 43 life-long tagging method, with low mortality rates (Grieve et al., 2018; Huusko et al., 2016; Wilder 44 et al., 2016), and are relatively inexpensive for large scale monitoring programs.

45

46 PIT tags are a widely used method for monitoring fish movements within and between waterways 47 (Meyer, 2017; Musselman et al., 2017; Smyth and Nebel, 2013). There are two types of 48 transmission and tag types, full-duplex (FDX) and half-duplex (HDX). HDX systems operate in an 49 alternating on-off read pulse. When a HDX antenna is on, the magnetic field charges the capacitator 50 in any HDX PIT tags within range. Then when the antenna is off, or in read mode, it detects the 51 signal emitted by the charged PIT tag. This stored energy gives HDX systems greater range than FDX systems, which transmit continuously and do not contain a capacitator in the PIT tag. This
lack of capacitator enables FDX tags to be smaller than HDX tags, allowing small-bodied and
juvenile fish to be tagged.

55

56 Fish species suitable for PIT tagging are limited by the ratio of tag size to body mass; this ratio is 57 called the 'tag burden'. The tag burden that a fish can carry without its health being compromised is 58 species-specific (Brown et al., 1999; Jørgensen et al., 2017; Schreck et al., 2004). Generally having 59 a dry tag mass of less than 2 % of fish body mass is considered standard (Hanson and Barron, 2017; 60 Childs et al., 2011; Winter, 1983), but tag burdens of up to 12 % have been reported (see Clark, 61 2016 and references within). Most commonly utilised PIT tags for fish tagging are 12, 23 or 32 mm in length (0.1 - 0.5 g). Consequently, PIT tagging has been limited to fish of body masses greater 62 than 5 g. Unfortunately, this means that a large number of small bodied fish species and juveniles of 63 64 larger bodied species are often excluded from tagging studies and this represents a significant gap in our understanding of fish movement and behaviour. The relatively recent advances in the 65 development of small (< 12 mm) microPIT tags enables us to include small-bodied (< 100 mm, 5 g) 66 species, and juveniles of larger growing species. While microPIT tags offer the potential to study 67 movement behaviours in small bodied and juvenile fish, there is a significant trade-off between PIT 68 69 tag size and tag detection range (Burnett et al., 2013). Tag detection range is the maximum distance 70 between the tag and the detecting antenna. Currently, the smallest commercially available PIT tags 71 are 6 x 1 mm in size (Nonatek RFID, Lutronic International, Rodange, Luxembourge, 72 www.nonatec.net), but are reported to have a limited detection range of 1 cm (Cousin et al., 2012). 73 By comparison, standard 12 and 23 mm HDX PIT tags have a detection distance of up to 1.5 and 3 74 m, respectively (Baker et al. 2017). Intermediate sized tags (8.4 mm, 0.03 g, Biomark) offer an 75 appropriate compromise, with detection ranges of up to 30 cm.

77 Although the development of microPIT tags has the potential to improve our understanding of a movement behaviours of greater range of species and juvenile life history stages, their long-term 78 79 impact on fish swimming performance of small (> 10 g) fish has not been well assessed. Survival 80 and tag retention rates are commonly reported in the literature (e.g. Baras et al., 2000; Dixon and 81 Mesa, 2011; Richard et al., 2013; Wagner et al., 2007; Ward et al., 2015; Weber and Flammang, 82 2017; Wilder et al., 2016), but impacts on swimming performance are scarce (Clark, 2016; Collins 83 et al., 2013; Knaepkens et al., 2007). There was no significant effect of 12 mm PIT tags on the 84 swimming performance of the Bullhead (Cottus gobio), which carried a tag burden close to 5 % 85 (Knaepkens et al., 2007). Likewise, there was no effect of either 8 or 12 mm PIT tags on the 86 swimming performance of the Blackspotted topminnow (Fundulus olivaceus) (Clark, 2016), while 87 Sockeye salmon (Oncorhynchus nerka) only showed a reduced swim performance with tag burdens 88 of greater than 8 % (Collins et al., 2013). This leads us to hypothesise that the 8.4mm microPIT tags 89 will not affect the survival rate or swimming performance of the small bodied, and juvenile fish of 90 species we test.

91

To assess the utility of microPIT tags for fish tracking studies of small-bodied species and juveniles of larger growing species of fish, we quantified the effects of implanted microPIT tags on both sustained and burst swimming performance (*Ucrit* and *Usprint*, respectively) of ten native Australian fish species. We hypothesised that the greatest impacts associated with the implantation and retention of the microPIT in the coeliac cavity of the fish would be evident early on, and that there would be no effect of the tags on the swimming performance once the fish were fully recovered from the procedure.

- 100 **2. Methods**
- 101
- 102 2.1 Fish husbandry

103 Fish were obtained from commercial hatcheries and maintained at The University of Queensland in 104 1000 L recirculation holding systems. Each system comprised of 12 x 37.5 L glass aquaria (L x W x 105 H, 60 x 25 x 25 cm) and a sump housing mechanical and biological filtration, and UV sterilisation. The temperature was controlled at 25 +/- 1 °C and photoperiod set to 12 h light: 12 h dark cvcle. An 106 107 automated freshwater flow through system delivered a constant supply of filtered Brisbane City tap 108 water. Tap water was filtered through a 10 µm particulate filter, followed by carbon filtration to remove chlorine, chloramines and dissolved contaminants. The flow through system replaced 25% 109 110 of the system volume each day. Ammonia, nitrite and nitrate were measured weekly and were 111 below the detection range of test kits (Sera, Germany, www.sera.de). Fish were fed a mix of Hikari Tropical Micro-wafers (Hikari, Kyorin Co. Ltd., Japan) and frozen bloodworms (Hikari, Kyorin Co. 112 113 Ltd., Japan), to satiation twice per day.

114

115 *2.2 Implantation technique*

PIT tags can be injected with a specialised needle or surgically implanted, both of which have high 116 117 survival rates (Baras et al., 2000; 1999; Cousin et al., 2012). The PIT tagging procedure we used 118 followed the surgical protocol outlined in Cousin et al. (2012). In brief, the fish were lightly anesthetised with 17 mg L⁻¹ Aqui-S (Aqui-S, Lower Hutt, New Zealand, www.aqui-s.com) and 119 120 their total length and total mass recorded. Tags (8.4 mm, 0.03 g, Biomark) and surgical equipment 121 were sterilised with 70% ethanol prior to use. To insert the PIT tag, a 19-gauge needle (Livingstone 122 International, New South Wales, Australia) was used to carefully pierce the posterior abdominal 123 skin. The diameter of this hole was stretched with fine forceps, and the 8.4 mm PIT tag gently 124 inserted into the coelemic cavity using locking forceps. The elasticity of the skin was enough to 125 naturally close the insertion point. The fish were placed into an oxygenated recovery tank, before 126 being returned to the holding system once normal swimming behaviour was observed.

127

128 2.3 Study Species – Suitability for tagging trial

7

129 The study species comprised a diverse range of morphologies and ecological niches, including five

130 small-bodied fish species: Blue eyes (*Pseudomugil signifier*), Crimson-spotted rainbowfish

131 (Melanotaenia duboulayi), Empire gudgeon (Hypseleotris compressa), Glassfish (Ambassia

132 *agassizii*), and Southern pygmy perch (*Nannoperca australis*) (see Table 1 for size ranges). The

133 populations of both the Glassfish and the Southern pygmy perch are listed as endangered in the

134 Murray-Darling basin (Lintermans, 2009). Juveniles (< 100 mm) of five additional large bodied

135 species were also chosen with diverse morphologies and different life cycle migrations. These

136 species were Australian bass (Macquaria novemaculeata), Golden perch (Macquaria ambigua),

137 Murray cod (Maccullochella peeli), Silver perch (Bidyanus bidyanus) and Eel-tailed catfish

138 (*Tandanus tandanus*). The PIT tag treatment groups of fish were implanted with 8.4 mm PIT tags 139 using the above technique and their survival and tag retention rates recorded for three weeks. The 140 sample sizes in this initial experiment were kept small to identify any species that were unsuitable 141 for microPIT tagging, without causing unnecessary mortalities.

142

143 2.4 Species included to quantify the effect of PIT tags on swimming performance

144 N. australis and P. signifer were excluded from the performance experiment due to their low 145 survival rates in the initial survival and tag retention trial. The three small-bodied species included 146 were A. agassizii, M. Duboulavi and H. compressa, along with juveniles of M. Novemaculeata, M. peeli, M. ambigua, B. bidyanus and T. tandanus (see Table 2 for size ranges). Individual fish were 147 148 randomly assigned to the PIT tagged or control groups, with more fish allocated to the PIT tag 149 group to account for mortalities and ensure sample sizes were sufficient for swimming tests (see 150 Table 2 for respective size range distributions). The control group were anesthetised (Aqui-S), 151 weighed (g) and measured (TL). Fish were left to recover for a minimum of three weeks before 152 being swum. Individual fish were only swum once in each Ucrit and Usprint trial. If a fish refused 153 to swim, the trial was stopped and the fish was not included in the analysis.

154

155 2.5 Swimming performance

156	To quantify the effect that PIT tagging has on the species included in this study, two measures of
157	swimming performance were measured. Both Ucrit (Brett, 1964; Rodgers et al., 2014) and Usprint
158	trials were performed on each fish after a recovery period of three weeks. For Ucrit trials, fish were
159	placed into a flow controlled recirculating flume (Loligo, Tjele, Denmark) and left for 5 min to
160	recuperate from handling stress. After the 5 min recovery period the water velocity was set to 0.1 m
161	s ⁻¹ . Then the velocity was increased by 0.05 m s ⁻¹ every 5 min until the fish fatigued, defined by the
162	fish resting on the back screen for 3 seconds. For the Usprint tests, individual fish were transferred
163	to the 185 L recirculating flume and left for 5 min at 0.1 m s ⁻¹ to orientate with the flow. After the 5
164	min, the velocity was increased by 0.05 m s ⁻¹ every 10 s until the fish fatigued as defined for Ucrit.
165	After swimming each Ucrit and Usprint test, fish were measured (total length), and after being
166	lightly dried with paper towel to remove excess water, weighed. Both Ucrit and Usprint were
167	calculated as in Brett (1964):

168

169 $Ucrit/Usprint = Uf + [Ui \times (Tf / Ti)]$

170

where Uf is the highest sustained water velocity (m s⁻¹), Ui is the velocity increase increments (0.05 m s⁻¹ for both *Ucrit* and *Usprint*), *Tf* is time swum during the final velocity increment, and *Ti* is the time increment (300 and 10 s for *Ucrit* and *Usprint* respectively). The Loligo flume was calibrated using a Prandtl-pitot tube (Kern et al., 2017) and the temperature was controlled at 25 ± 1 °C.

175

176 2.6 Statistical analysis

177 All statistical analyses were conducted using R (version 3.4.0) within the RStudio environment

178 (version 1.0.143). Ucrit and Usprint values were Box-Cox transformed (car package) (Fox and

179 Weisberg, 2011) and fitted to a Generalised Least Squares linear model (nlme package) (Pinheiro et

- al., 2012) with treatment (PIT tag or control), fish length (TL) and time between procedure and
- 181 testing (days) as interacting variables. Fish mass (g) was excluded due to its high correlation with

body length (Rodgers et al., 2014). Statistical significance was defined as P < 0.05. Survival data
were analysed (Chi-square test) and visualised using the following packages: survival (Therneau,
2015), ggfortify (Horikoshi and Tang, 2016), and survminer (Kassambara et al., 2017). The dplyr
(Wickham et al., 2017), ggpubr (Kassambara, 2017) and ggplot2 (Wickham, 2009) packages were
used to visualise data and create figures.

- 187
- 188 **3. Results**
- 189
- 190 *3.1 Suitability for microPIT tagging trial*

191 To determine each species' suitability for PIT tagging, we performed a trial to identify species not 192 suitable for microPIT tagging. *M. novemaculeata* (n = 10), *M. peeli* (n = 10), *B. bidyanus* (n = 10) 193 and T. tandanus (n = 10) displayed 100% survival after three weeks (Fig. 1). Thirty percent of A. 194 agassizzii (n = 10) died within 24 hours of the procedure, with no subsequent mortalities within the three-week timeframe. H. compressa (n = 10) and M. ambigua (n = 18) respectively displayed 195 196 40.0% and 16.7% mortality, with deaths occurring within five days of undergoing the implanting 197 procedure. M. duboulayi also had 30% mortality over the 48 hours post-procedure, with one 198 additional death after 15 days (40% end mortality). Finally, both N. australis (n = 10) and P. 199 signifer (n = 10) displayed 100% mortality. A Chi-squared test showed there was no effect of body 200 length on survival rate for *M. duboulavi*, *H. compressa* and *A. agassizii* (see S1 for length vs mass plot showing individual mortalities). There was a positive effect of body mass on the survival rate 201 202 of *M. duboulayi* (p = 0.032), and a positive effect of both body length (p = 0.047) and body mass (p203 = 0.017) on *M. ambigua* survival rate.

204

Across all species, the tag burden ranged from 0.3 to 7.5 % body mass (Table 1), with the tag

206 burden being highest for P. signifier and M. ambigua. H. compressa, A. agassizii, N. australis and

207 *M. novemaculeata* all contained individuals with tag burdens above 2%. There was 100% retention

of PIT tags throughout the experiment, with most species showing no signs of declining health. *H. compressa* appeared susceptible to fungal infection after undergoing the PIT tagging procedure.

210

211 3.2 Survival rates

M. duboulayi, H. compressa, A. agassizii and M. ambigua tagged for the subsequent performance experiment had a reduced mortality rate compared to individuals in the initial survival trial (Tables 1 and 2). M. peeli, B. bidyanus and T. tandanus had 100% survival in both experiments and both treatment groups of the performance experiment. M. novemaculeata mortality was higher in performance experiment, although the three deaths that occurred in the PIT tag group occurred three months after undergoing the PIT tag procedure, a much longer timeframe than the three weeks of the initial survival trial and were possibly caused by long term tagging-related health impacts.

219

All *M. peeli, B. bidyanus* and *T. tandanus* in the PIT tag and control treatments survived (Table 2). *M. duboulayi, H. compressa, A. agassizii* displayed low mortality rates of 11.8, 8.8 and 12.5% in
the PIT tag groups, with all fish in their control groups surviving (Table 2). *M. novemaculeata*displayed high survival rates in both the PIT tag (6.7%) and control groups (2.5%). *M. ambigua* had
a higher mortality rate in the control group (16.7%) than the PIT tag (5.4%). Out of the five *M. ambigua* that died in the control treatment, four did so within 24 hours of being anesthetised,
measured and weighed.

227

228 3.3 The effect of PIT tags on swimming performance

There was no significant effect of PIT tagging on the critical swimming speed (*Ucrit*) of any of the eight species tested, when accounting for the effects of body length and time since tagging. When visually inspected, the presence of outliers indicates that some individual fish may have had their performance impaired by the PIT tag (Fig. 2). *M. duboulayi*, *H. compressa*, *M. ambigua* and *T. tandanus* all show individuals that appear as outliers within the PIT tag group (Fig. 2). There was

- for all species, once the effects of body size and time since tagging were accounted for. The
- apparent reduced sprinting capacity of *M. peeli* is explained by the larger size of the control fish

237 (Table 2).

238

Species	Life Stage	n	Body Mass (g) [range]	Body Length (cm) [range]	Min tag burden (% body mass)	Max tag burden (% body mass)	Mortality ¹ rate (%) ₂
P. signifier	Adult	10	$0.52\pm 0.1[0.4-0.7]$	$3.5\pm0.2\;[3.1-3.8]$	4.3	7.5	¹⁰⁰ 243
M. duboulayi	Adult	10	$2.9\pm1.8\;[1.6-6.1]$	6.7 ± 1.2 [5.5-8.7]	0.5	1.9	40 244
H. compressa	Adult	10	$2.8\pm 1.8\;[1.1-6.3]$	$6.2\pm1.1\;[4.8-7.9]$	0.5	2.7	40 245
A. agassizii	Adult	10	$1.9\pm 0.5\;[1.1-2.6]$	$5.4 \pm 0.4 \; [4.5 - 5.8]$	1.2	2.7	30 247
N. australis	Adult	10	$1.8\pm 0.6\;[1.1-3.2]$	$5.2\pm0.5\;[4.2-6.2]$	1.0	4.3	248 17 249
M. novemaculeata	Juvenile	10	$1.7\pm0.5[1.0-2.7]$	5.1 ± 0.5 [4.4 - 5.8]	0.6	1.6	$\begin{smallmatrix}&250\\&251\end{smallmatrix}$
M. ambigua	Juvenile	18	$1.6\pm 0.5\;[0.7-2.2]$	$5.1\pm 0.5\;[4.1-6.3]$	0.9	2.7	100252
Macc. peeli	Juvenile	15	$3.0\pm0.7\;[1.9-4.8]$	$6.8\pm0.5\;[6.0-7.9]$	0.4	0.7	0
B. bidyanus	Juvenile	10	$6.0\pm1.4\;[4.1-7.5]$	$8.2 \pm 0.6 \ [7.3 - 8.8]$	0.3	1.2	0
T. tandanus	Juvenile	10	$4.8\pm1.9\;[2.6-9.1]$	$8.6 \pm 1.3 \; [6.2 - 10.6]$	1.1	3.0	0

Table 1. Size distribution, tag burdens and mortality rates of the fish included in the suitability for tagging trial.

254 **Table 2**. Size range of individual fish allocated to the PIT tagging and VIE control group for the swim trials.

Species	Treatment	n	Body Mass (g) [range]	Body Length (cm) [range]	Min tag burden (% body mass)	Max tag burden (% body mass)	Mortality rate (%)	
	PIT tagged	32	$3.3 \pm 1.7 \ [1.1 - 6.1]$	7.0 ± 1.2 [4.8 – 8.7]	0.5	2.7	11.8	
M. Duboulayi	Control	26	$3.5\pm2.7\;[1.6-14.4]$	$6.8\pm1.2\;[5.2-10.5]$			0	
	PIT tagged	34	$2.5\pm1.1\;[1.0-4.8]$	$6.0\pm0.8\;[4.5-7.9]$	0.6	3	8.8	
H. compressa	Control	30	$1.7\pm0.8\;[0.7-4.9]$	$5.4\pm0.7\;[4.3-7.9]$			0	
<i>,</i>	PIT tagged	32	$2.2\pm0.7\;[1.0-3.9]$	$5.7\pm0.7\;[4.2-6.8]$	0.8	3	12.5	
A. agassizii	Control	27	$2.4\pm0.9\;[0.8-4.1]$	$5.7\pm0.7\;[4.1-6.9]$			0	
	PIT tagged	37	$1.8\pm0.5\;[1.0-3.0]$	$5.3\pm 0.5\;[4.2-6.3]$	1	3	5.4	
M. ambigua	Control	30	$2.0\pm0.8\;[0.8-4.7]$	$5.6\pm0.6\;[4.4-7.5]$			16.7	
	PIT tagged	35	$4.8\pm2.1\;[1.9-9.1]$	$7.6\pm1.0\;[6.0-9.9]$	0.3	1.6	0	
M. peeli	Control	30	$9.9\pm2.6\;[6.7-15.8]$	$9.2\pm0.7\;[8.2-10.9]$			0	
	PIT tagged	55	$2.9 \pm 1.7 \; [1.0 - 7.5]$	$6.3\pm1.1\;[4.5-8.8]$	0.4	3	0	
B. bidyanus	Control	40	$3.0\pm1.1\;[1.1-5.4]$	$6.2\pm0.9\;[4.6-8.1]$			0	
	PIT tagged	21	$5.7\pm2.5\;[2.6-11.9]$	$9.3 \pm 1.4 \; [6.2 - 12.2]$	0.3	1.2	0	
T. tandanus	Control	18	$5.7 \pm 1.2 \; [3.1 - 7.4]$	$9.3\pm0.6\;[7.9-10.1]$			0	
	PIT tagged	45	$1.8\pm 0.5\;[0.9-2.7]$	$5.1\pm 0.5\;[4.2-5.9]$	1.1	3.3	6.7	
M. novemaculeata	Control	40	$2.5\pm 0.8\;[0.8-4.6]$	$5.6\pm0.6\;[4.1-7.1]$			2.5	

257 **4. Discussion**

258

259 PIT tags present a long term, cost effective method for monitoring fish movements and have been 260 shown to have negligible impact on the survival, growth rate, foraging behaviour and reproduction 261 of many fish species (Bangs et al., 2013; Bunt et al., 2011 and references within; Clark, 2016; 262 Dixon and Mesa, 2011; Fraiola and Carlson, 2016; Knaepkens et al., 2007; Ward et al., 2015). This generalisation cannot be uniformly applied to all fish species as significant mortality and potential 263 264 sub-lethal effects have also been reported in the literature (Dawson et al., 2015; Hanson and Barron, 265 2017; Wilder et al., 2016). As the swimming capacity of a fish is directly related to its survival 266 (Starrs et al., 2011), we used this metric to quantify the potential utility of PIT tags for monitoring 267 small-bodied and juvenile native Australian fish. We show that while microPIT tagging had no significant effect on two measures of swimming performance in eight fish species, a further two 268 269 species were identified as not suitable for microPIT tagging, displaying significant mortality within 270 48 hours of the implant procedure.

271

272 The two small-bodied species that displayed significant mortality following tagging were N. 273 australis and P. signifier. This high mortality occurred despite the tag burden for some N. Australia 274 being below the 2% body weight recommendation (Hanson and Barron, 2017; Winter, 1983). This supports the growing literature asserting that maximum tolerable tag burdens are species-specific 275 276 (Brown et al., 1999; Jørgensen et al., 2017; Schreck et al., 2004), with morphology and degree of 277 stress response to implantation also being important in determining a species' suitability and 278 minimum tag-able fish size. The 2% fish to tag mass ratio may serve as an initial guide but it is not 279 comprehensive across all fish species. For example, the Oregon chub (Oregonichthys crameri) is a 280 small-bodied endangered North American fish with similar body morphology to N. australis, yet 281 individuals of the same size class showed high survival rate after tagging with 8 mm microPIT tags 282 (Bangs et al., 2013). Although P. signifier were the smallest species included in this study, they

283 have proportionally large abdominal cavities, with the potential to accommodate the microPIT tags, 284 yet the mortality rate was high. This high mortality rate of both P. signifier and N. australis may be 285 due to an elevated stress response that could have been reduced or avoided by using an alternative 286 anaesthetic. Fish species have been shown to respond to different anaesthetics with varying stress response blood profiles (Berlinsky et al., 2016). Specific to Aqui-S, it has been shown to 287 288 immediately irritate juvenile Pacific Lamprey to the point that many individuals remove themselves 289 from contact (Christiansen et al., 2013). Trialling the use of alternate anaesthetics may help resolve 290 mortality rates in N. australis, P. signifier, and other untested species that display high mortality 291 within 24 hours of being PIT tagged when anesthetised using Aqui-S. 292 293 We allowed a minimum of three weeks for fish to recover before performing any swimming 294 performance test to ensure that the fish had finished healing from the tag insertion procedure. The

295 lack of significant effect of PIT tagging on swimming performance indicated that shortening the recovery timeframe after the implanting procedure might be acceptable, improving the logistics of 296 297 capture-tag-release programs. In Australia, M. peeli, M. novemaculeata, B. bidyanus and M. 298 ambigua are popular freshwater recreational angling species, the juveniles of which are produced in 299 commercial hatcheries and stocked into natural water-bodies. Our results indicate that stocking 300 programs may incorporate PIT tagging of juveniles, enabling life-long tracking of movements 301 throughout large waterways with existing monitoring stations, such as the Murray-Darling River 302 Basin.

303

304 Challenging water velocities are often associated with anthropogenic stream modifications, such as 305 dams, weirs and culverts (Goodrich et al., 2018; Watson et al., 2018). Fishways are structures 306 designed to facilitate fish passage around or through such barriers. To be classified as a successfully 307 remediated barrier, the fishway must pass a high percentage of fish that attempt passage that will be 308 site and monitoring agency specific. However, the effectiveness of most fish passage structures is 309 assessed by monitoring the movement of medium to large individuals that are of a size suitable for 310 conventional tagging approaches. Virtually nothing is known about the movement behaviours of the 311 most vulnerable size class of Australia native fish: small-bodied and/or juvenile fish less than 100 312 mm long. Our results give confidence that microPIT tags are suitable for evaluating the 313 effectiveness of fish passage remediation efforts for smaller-bodied and juvenile Australian fish 314 species, helping inform fisheries management and advance research in this field. Such data will be 315 more informative than simply upstream collection of successfully traversed fish, as this does not 316 give information on failed passage rates.

There are limited data on the impacts of PIT tags on swimming performance of small fish. Although our results indicate that if a species has high survival rates, then its swimming performance is likely to be unaffected, further work is needed to support this. It was important to quantify the effects on swimming performance in addition to the survival and retention rates, as a reduced capacity to swim would lead to a range of sublethal effects including the ability to find food, evade predation, reproduce and move against challenging water velocities.

323

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499 **Figure captions**

500

Figure 1. Survival curves for ten native Australian fish species that underwent implantation of an 8.4mm PIT tag. An asterisk denotes censored data points where fish survived the three-week time period of the trial. The initial number of individual fish for each species follows the species name, while the number of individuals that survived three weeks is noted in the bottom right corner of each species respective survival curve.

506

Figure 2. The effect of PIT tags on the critical swimming speed (*Ucrit*) and sprint swimming
performance (*Usprint*) of eight Australian freshwater fish species. For all species there was no
statistically significant effect of the PIT tags on *Ucrit* when compared to the control group, once
body size, mass and time between procedure and performance test were included into the analysis
S1. The length to body mass relationships of the six species with mortalities in the initial survival

513 trial. The four species with 100 % survival are not shown. Individuals that survived for at least 3

514 weeks post PIT tagging are denoted with blue and mortalities in red.