

# Long Term Retention, Survival, Growth, and Physiological Indicators of Juvenile Salmonids Marked with Passive Integrated Transponder Tags

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**Abstract.**—To track individuals in situ, over 12 million salmon and trout have been marked with passive integrated transponder (PIT) tags in the Columbia River Basin, USA. However, few studies have examined long term tag retention as well as tag effects on juvenile salmon and trout. We marked juvenile coho salmon *Oncorhynchus kisutch* ( $N = 207$ ), steelhead (anadromous rainbow trout) *O. mykiss* ( $N = 221$ ), cutthroat trout *O. clarkii* ( $N = 202$ ) and bull trout *Salvelinus confluentus* ( $N = 180$ ) with 12, 19, or 23 mm PIT tags and examined tag retention, survival, growth, and physiological performance over a six month period in a laboratory environment. PIT tag retention rates were high for coho salmon (100%), steelhead (95%), cutthroat trout (97%), and bull trout (99%), regardless of tag size. Survival was also high for coho (99%), steelhead (99%), cutthroat trout (97%), and bull trout (88%) and did not vary among tag sizes. Short term individual growth rates for coho salmon marked with 12 mm tags were significantly higher than those marked with 19 mm and 23 mm PIT tags. Likewise, steelhead trout individual growth rates were lower for fish marked with 23 mm PIT tags followed by 19 and 12 mm tags. Conversely, long-term growth rates were positive and not affected by tag size. There were no significant effects of tag size or marking on coho gill  $\text{Na}^+$ ,  $\text{K}^+$ , -ATPase activity ( $\mu\text{mol ADP} \times \text{mg protein}^{-1} \text{h}^{-1}$ ) and plasma osmolality ( $\mu\text{mol kg}^{-1}$ ) or bull trout hepatosomatic indices. Our study suggests that marking juvenile salmonids with PIT tags results in high retention with little effect upon their survival, growth, and important physiological indicators regardless of tag size in a laboratory environment.

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## Introduction

Since the 1980s, passive integrated transponder (PIT) tags have been used to support the collection of various biological and population demographic data in a variety of animal models (Gibbons and Andrews 2004). For example, this technology has been used extensively in the Pacific Northwest for monitoring the behavior and survival of juvenile and adult salmonids in the Columbia River Basin (Zabel et al. 2005). Over 15 million salmon and trout have been PIT tagged in the Columbia River Basin since 1987 (Pacific States Marine Fisheries Commission 2005). Though 12-mm PIT tags are most commonly used, to maximize detection distance many researchers employ a variety of tag sizes including 19-mm and 23-mm PIT tags (Rousset et al. 2000; Zydlewski et al. 2001; Hill et al. 2006). The distance at which a PIT tag can be detected is particularly important in applications where fish are not recaptured but are remotely detected via a fixed or mobile antenna. Since data collected from PIT tag detections is often used to determine stock identity (Jenkins and Smith 1990; Achord et al. 1996), movements (Ombredane et al. 1998; Zydlewski et al. 2001), migration rates and routes (Achord et al. 1996; Kennedy et al. 2007a), abundance (Achord et al. 1996), growth (Peterson et al. 1994), mortality (Kennedy et al. 2007a), and stocking success (Wills 2006), it is important that the PIT tags themselves do not directly or indirectly alter these data.

Although PIT tag retention rates (Buzby and Deegan 1999; Gries and Letcher 2002; Peterson et al. 1994) and short term post tagging survival (Prentice et al. 1990; Peterson et al. 1994; Ombredane et al. 1998; Gries and Letcher 2002; Bateman and Gresswell 2006) and growth (Prentice et al. 1990; Peterson et al. 1994; Ombredane et al. 1998; Bateman and Gresswell 2006) have been examined and commonly reported in the literature, the

effects of tags on long term growth and important physiological processes for salmonids have only been described only preliminarily. Given the importance of these processes on salmonid growth and survival it is important to understand if PIT tag marking imparts negative effects upon smoltification, growth, and energy storage. In addition, the results from these earlier studies were often confounded by differences in environment, tag size, surgical and implantation technique, study duration, and species and fish life history stage (Bateman and Gresswell 2006) making it difficult to independently evaluate the effect of marking salmonids with PIT tags. As a result we examined the long-term retention and effect of different sized PIT tags (12, 19, 23 mm) on the survival, growth, and physiological performance of juvenile steelhead (anadromous rainbow trout) *Oncorhynchus mykiss*, coho salmon *O. kisutch*, cutthroat trout *O. clarkii*, and bull trout *Salvelinus confluentus* in a laboratory environment.

## Methods

Coho salmon and steelhead trout eggs were obtained from Washington Department of Fish and Wildlife (WDFW) Big Creek Hatchery, cutthroat trout eggs were obtained from WDFW Cowlitz Complex, and adfluvial bull trout eggs were obtained from the U.S. Fish and Wildlife Service's Creston National Fish Hatchery (NFH). Eggs were transported to Abernathy Fish Technology Center (AFTC) incubated and hatched in flow through trays maintained on well water (12.5°C) and a natural photoperiod. Juvenile coho salmon (N = 207), steelhead (N = 221), cutthroat (N = 202) and bull trout (N = 180) were reared on artificial salmonid feeds until they reached a desired size (range = 100–150 mm, Fork Length, Florida). Individual fish were anesthetized, measured, weighed, and randomly implanted with a 12, 19, or 23 mm

PIT tag (134.2 khz ISO; Destron Fearing Inc.) or left untagged (controls). The PIT tags were inserted by cutting a small ( $\leq 5$  mm) ventral opening into the body cavity just behind the pectoral fin insertion (Gries and Letcher 2002). A PIT tag was placed into the body cavity in a lateral ventral position behind the pectoral fin insertion but in front of the pelvic fin insertion. Coho and steelhead were marked with all three differently sized tags whereas bull trout were marked with 12 and 23 mm PIT tags and cutthroat trout with 23 mm tags. Equal numbers of fish were placed within three replicate circular tanks (1.2 m diameter) that were supplied with approximately 1 l/min well water at ambient temperature ( $10 \pm 2^\circ\text{C}$ ). Fish were fed Bio-Oregon dry pellets daily at 1% body weight. Feeding levels were adjusted according to total biomass and water temperature monthly. Tanks were examined daily for expelled tags and fish mortality. During each month, for up to 8 months, fish were anesthetized, measured, weighed, and identified via PIT tag code to determine growth rates. Growth rates were determined between all sampling dates by calculating specific growth (Busacker et al. 1990).

During the spring (March, April, and May) coho salmon gill  $\text{Na}^+$ ,  $\text{K}^+$ -ATPase (ATPase) activity and plasma osmolality ( $\text{mOsm} \times \text{Kg H}_2\text{O}^{-1}$ ) were determined as relative indicators of seawater readiness and smoltification. ATPase activity was determined via a small gill biopsy using the method of McCormick (1993) and immediately frozen on dry ice. Gill samples were stored at  $-80^\circ\text{C}$  and gill ATPase activity measured spectrophotometrically using the method of McCormick (1993). To measure the relative seawater tolerance, plasma osmolality was determined after 24 h salt water exposures (32 ppt). Fish were killed by an overdose of anesthetic, and blood collected using heparinized syringes. Plasma was obtained by centrifugation at 10,000 rpm,  $4^\circ\text{C}$  for 5 min. Osmolality was

determined using an Osmette II osmometer.

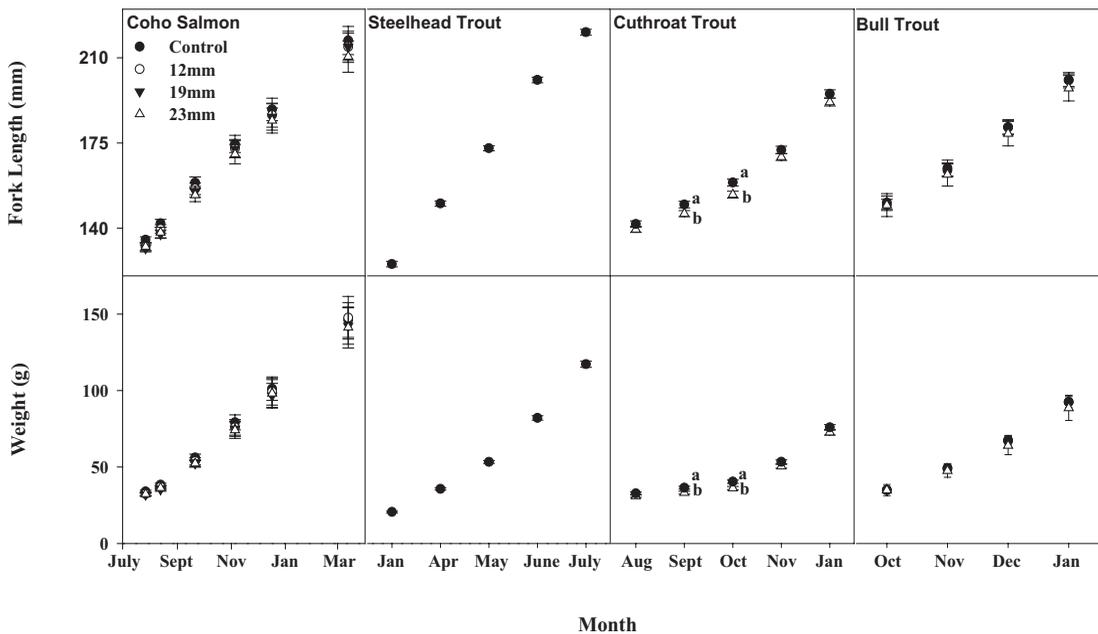
We determined the hepatosomatic index (HSI) as an indicator of glycogen stores of adfluvial bulltrout. At the termination of the experiment fish were measured (FL), weighed, and livers were dissected, removed from the viscera, weighed, and HIS was calculated as:  $\text{HIS} = (\text{fish liver weight (g)}/\text{fish body weight (g)}) \times 100$ .

Analysis of Variance (analysis of variance (ANOVA)) and *t*-tests were used to test for differences among the treatments for the response variables (FL, weight, growth rate, ATPase, plasma osmolality, and LR). Chi-square tests were used to compare differences ( $P < 0.05$ ) in tag retention and mortality among treatments for each species. Statistical comparisons of growth rates included only fish marked with PIT tags because untagged control fish could not be individually identified. Data were log transformed to meet assumptions of normality and homogeneity of variances. Significant ANOVAs ( $P < 0.05$ ) were followed by Fisher LSD mean separation tests for pairwise comparisons.

## Results

All fish increased in FL and gained weight during the experiment regardless of species or PIT tag size (Figure 1). There were no significant differences in FL or weight among treatment groups for any species, excluding cutthroat trout. Although cutthroat trout marked with 23 mm PIT tags were similar in FL and weight at the onset and completion of the experiment they differed during September (FL,  $P = 0.047$ ; weight,  $P = 0.016$ ) and October (FL,  $P = 0.014$ ; weight,  $P = 0.007$ ). Percent weight gain did not significantly differ between the control fish and those marked with different sized tags, in spite of species (Table 1).

Species specific growth rates of fish marked with PIT tags may be negatively af-



**FIGURE 1.** Fork length (mean  $\pm$  1 SE) and weight (mean  $\pm$  1 SE) of coho salmon, steelhead, cutthroat, and bull trout unmarked or marked with a 12, 19, or 23 mm passive integrated transponder tag. Letters denote significant ( $P < 0.05$ ) differences between fish marked with different sized PIT tags on a respective date.

ected following a marking event (Figure 2). The individual growth rates for coho salmon during the first growth interval were significantly lower for the fish marked with 19 mm and 23 mm PIT tags as compared to those marked with 12 mm PIT tags. Individual growth rates did not differ between coho salmon marked with 19 mm and 23 mm PIT tags during the first growth interval. Likewise, steelhead trout individual growth rates were slowest for fish marked with 23 mm PIT tags followed by 19 and 12 mm tags. During the third growth interval (May to June) steelhead marked with 23 mm PIT tags grew significantly faster than those marked with 19 or 12 mm tags that did not significantly differ from each other. Bull trout individual growth rates did not differ among fish marked with different sized PIT tags.

All PIT tagged coho salmon subjected to 24 h seawater challenges survived. Although ATPase activity was significantly higher ( $P$

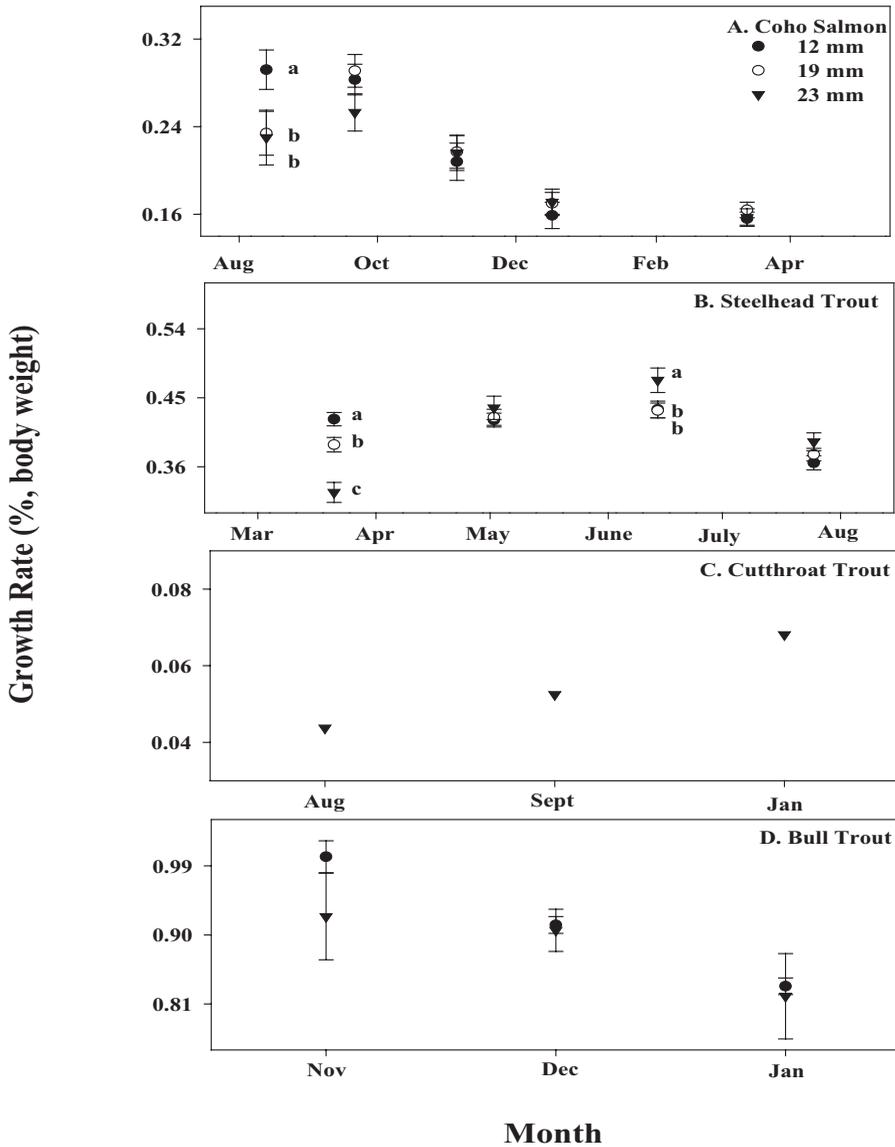
$= 0.003$ ) in April when compared to March and May, it did not differ among the groups marked with different sized tags (Figure 3A). Plasma osmolality was significantly higher in later months (Figure 3B). Nevertheless, there were no significant differences ( $P = 0.16$ ) in plasma osmolality among the coho salmon marked with PIT tags of various sizes.

Bull trout HSI did not differ among controls and the two marked groups (Figure 4). Fish marked with 23 mm PIT tags had a larger variance of relative liver weight than fish marked with either 12 mm PIT tags or controls.

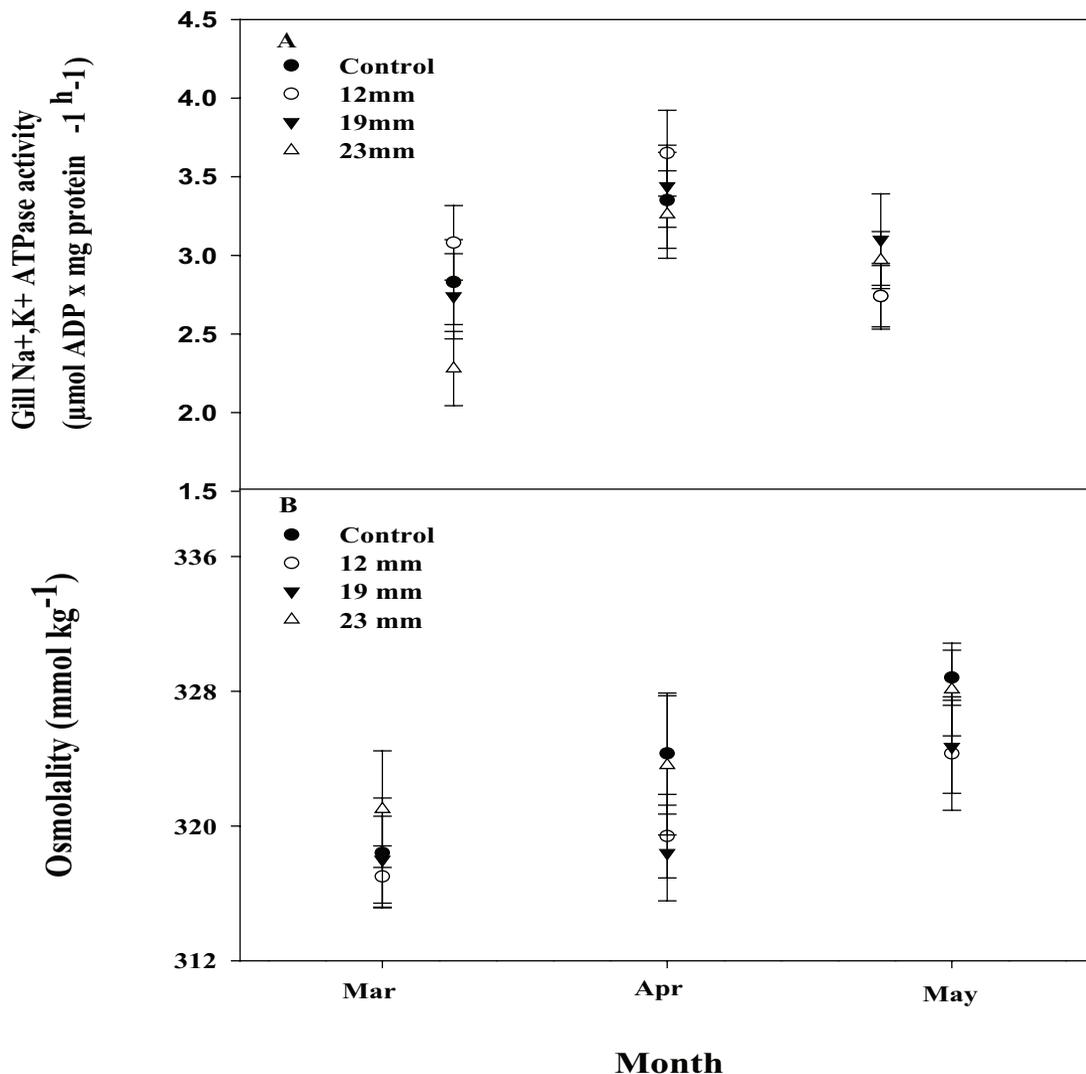
We observed low mortality and high PIT tag retention for all the species, regardless of tag size (Table 1). Mortality did not significantly differ within each species marked with different sized PIT tags. Mortality was higher for bull trout (range = 10–15%) than for the other species (range = 0–5%) we examined. Likewise PIT tag retention was high and did

**TABLE 1.** Mean ( $\pm$  1 SE) initial fork length (FL), initial weight, final weight gain, mortality, and passive integrated transponder (PIT) tag retention for coho salmon and steelhead, cutthroat, and bull trout marked with different sized PIT tags. There were no statistical differences among fish marked with different sized PIT tags regardless of species.

Species	Treatment	N	FL (mm)	Weight (g)	Weight Gain (%)	Mortality (%)	PIT tag Retention (%)
Coho salmon	Control	69	135 (1.24)	34 (0.82)	329.7	5.8	*
	12 mm	69	132 (1.58)	32 (1.04)	355.5	0.0	100
	19 mm	69	131 (1.47)	31 (1.00)	345.7	1.5	100
	23 mm	69	132 (1.72)	32 (1.09)	337.2	2.9	100
Steelhead trout	Control	75	125 (1.15)	20 (0.61)	468.9	1.3	*
	12 mm	75	124 (1.17)	20 (0.59)	492.5	1.3	98.7
	19 mm	75	124 (1.11)	20 (0.60)	477.7	0.0	97.3
	23 mm	75	123 (1.20)	20 (0.64)	477.6	1.3	89.0
Cutthroat trout	Control	101	136 (1.22)	31 (0.82)	131.9	5.0	*
	12 mm	*	*	*	*	*	*
	19 mm	*	*	*	*	*	*
	23 mm	101	141 (1.29)	32 (0.96)	134.5	3.0	100
Bull trout	Control	60	150 (2.56)	34 (1.80)	165.6	12.0	*
	12 mm	60	149 (2.20)	34 (1.60)	164.3	15.0	99.5
	19 mm	*	*	*	*	*	*
	23 mm	60	149 (2.56)	34 (1.83)	153.9	10.0	100



**FIGURE 2.** Individual growth rate (mean  $\pm$  1 SE) of coho salmon (A), steelhead (B), cutthroat (C), and bull trout (D) marked with a 12, 19, or 23 mm passive integrated transponder tag (PIT). Letters denote significant ( $P < 0.05$ ) differences between fish marked with different sized PIT tags.



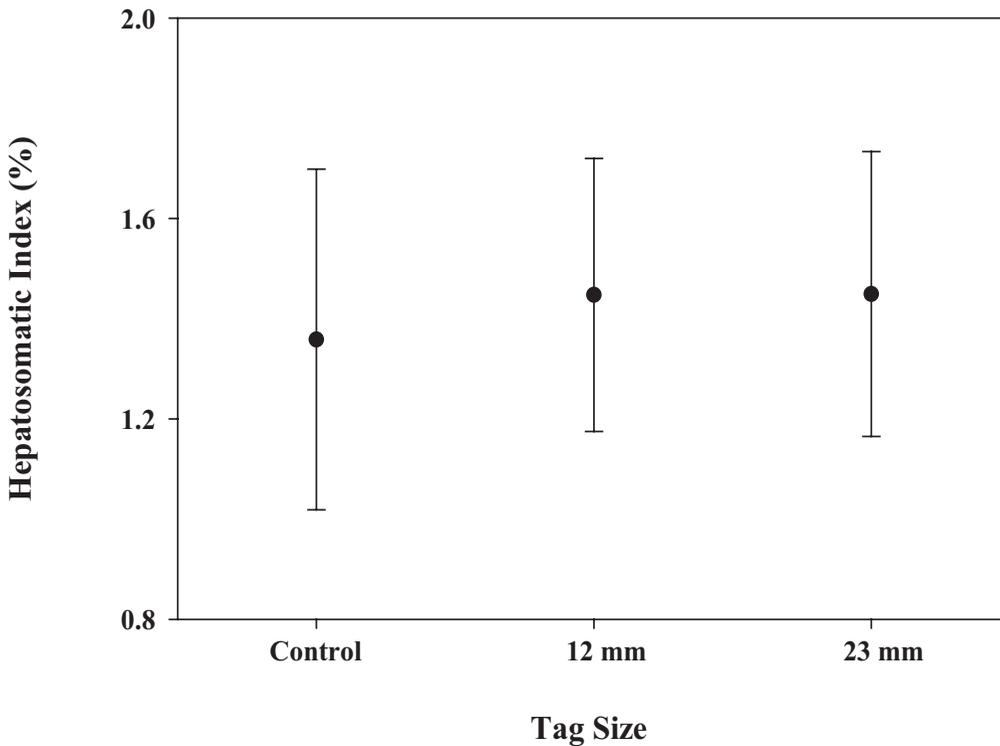
**FIGURE 3.** Mean ( $\pm 1$  SE) Gill Na<sup>+</sup>, K<sup>+</sup> -ATPase activity (A) and plasma osmolality (B) of coho salmon marked with a 12, 19, or 23 mm passive integrated transponder tags (PIT).

not differ among the treatments, regardless of species. PIT tag retention was the lowest for steelhead marked with 23 mm PIT tags (89%) as compared to all other treatments (range = 97–100%).

## Discussion

Our results suggest that there appears to be little effect of tagging on long term juvenile fish growth, physiological indicators,

mortality, and tag retention in a laboratory environment. Individual growth rates appear to be somewhat influenced by tag size shortly after marking. Therefore, researchers examining growth rates soon after tagging need to be cognizant of this potentially negative effect. As technology continues to improve and these tags become smaller, even short term effects on individual growth will likely become inconsequential. Our results suggest that the use of PIT tagged fish as indicators of stock identity (Jenkins and



**FIGURE 4.** Relative liver weight (mean  $\pm$  1 SE) for bull trout marked with either 12, or 23 mm passive integrated transponder tags (PIT). The length specific standard weight (Ws) developed by Hyatt and Hubert in (2000) was used to determine bull trout relative liver weight.

Smith 1990; Achord et al. 1996), movements (Ombredane et al. 1998; Zydlewski et al. 2001), migration rates and routes (Achord et al. 1996; Kennedy et al. 2007a), abundance (Achord et al. 1996), growth (Peterson et al. 1994), mortality (Kennedy et al. 2007a), and stocking success (Wills 2006) do not directly or indirectly influence or bias these data during juvenile life history stages. Nevertheless, it is important to note that PIT tag loss may be much higher during later life history stages (e.g., saltwater phase). PIT tagged fish appear to have higher mortality and lower smolt-to-adult recruit survival (SARS) than nonPIT tagged fish (see Knudsen et al. 2009; Williams et al. 2005). Thus our study results should be restricted to or cautiously extrapolated beyond juvenile life history stages.

Growth rates of fish, particularly coho salmon and steelhead, were negatively affected by PIT tagging shortly after tagging. Our results are similar to those observed for Chinook salmon *Oncorhynchus tshawytscha* (Prentice et al. 1990), brown trout *Salmo trutta* (Ombredane et al. 1998), Eurasian perch *Perca fluviatilis* (Baras et al. 2000), mottled sculpin *Cottus bairdii* (Ruetz et al. 2006), and steelhead trout (Bateman and Gresswell 2006). Furthermore, our results suggest that the negative influence on short term growth rates appears to be exacerbated as PIT tag size to body weight ratios increase. Growth rates have been reported to be negatively affected when PIT tag to body weight ratios exceed 4% (Baras et al. 2000; Ruetz et al. 2006). In our study, PIT tag to body weight ratios were less (range = 2% to 0.5%) than those previ-

ously reported, yet individual growth rates were still negatively effected in observations shortly after tagging. Fish that were implanted with heavier and larger sized tags had significantly slower growth shortly after tagging than controls or those implanted with smaller tags. This suggests that tag size, as well as the tagging event (Kennedy et al. 2007b) contributes to the observed growth patterns we observed following marking. Interestingly, it appears that once fish have recovered they are able to compensate for short term reductions in growth and resume growth patterns similar to that of control fish. Nevertheless, our results taken together with those of past studies suggest that a short period of reduced growth after tagging may be expected and should be considered when response variables are collected and measured shortly after tagging.

Our results suggest that PIT tagging does not impart significantly negative effects on coho smoltification or juvenile adfluvial bull trout glycogen stores. There were no significant differences in ATPase activity or plasma osmolality among coho salmon controls and those marked with different sized tags. In our study coho salmon were tagged prior to the initiation of smoltification. We suspect that the stress of the tagging event and the tag itself were minimal and therefore did not significantly affect the parr–smolt transformation. Our results for ATPase activity and plasma osmolality were similar to those reported for other salmonids reared under hatchery conditions (Hoar 1988; Hill et al. 2006; Kennedy et al. 2007a). Although PIT tag marking fish did not appear to affect smoltification, fish that are marked at the onset or during the smoltification process may be negatively influenced possibly slowing emigration, encouraging residualism, or increasing mortality (Kennedy et al. 2007b); however, additional research will be needed to examine this hypothesis. Likewise, bull trout HSI did not differ among controls and the two treatment groups. Nevertheless, the increased variance

associated with fish PIT tagged with 23 mm tags suggests that a proportion of our treatment population may have fared worse than the rest. Given bull trout's aggressive nature, the effect of social status combined with tag size may explain the observed variance. Thus PIT tagging conducted during critical periods of bull trout energy storage or utilization such as migratory or overwintering periods (Berry 1994; Ratcliff et al. 1996) may affect survival rates following marking events. We encourage researchers to consider the timing of PIT tag marking events as to avoid potentially affecting critical physiological processes.

Juvenile fish survival and PIT tag retention rates in this laboratory study were high and comparable to those reported in the literature. Although most studies on juvenile fish have been much shorter in duration than our study, our mortality rates were similar to those reported for Atlantic salmon *Salmo salar* (5.7%, 12 mm PIT tags, Gries and Letcher 2002; 21.2%, 23 mm PIT tags, Roussel et al. 2000), Chinook salmon (1%, 12 mm PIT tags, Achord et al. 1996; 12 mm PIT tags, 3.6%, Prentice et al. 1990), sockeye salmon *Oncorhynchus nerka* (3.6%, 12 mm PIT tags, Prentice et al. 1990), and steelhead (2%, 23 mm PIT tags, Hill et al. 2006; 14%, 23 mm PIT tags, Bataman and Gresswell 2006). Like our results, previously reported tag retention rates (range = 30–100%) were relatively high for juveniles (Prentice et al. 1990; Peterson et al. 1994; Ombredane et al. 1998; Bataman and Gresswell 2006; Knudsen et al. 2009). Collectively, these results and our study indicate that low mortality and high retention rates will occur if good fish handling practices are employed and fish are allowed to fully recover prior to release.

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