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Abstract

Passive integrated transponder (PIT) tags are commonly used to monitor growth, habitat use, activity rates, and survival of individual fish. However, for successful completion of research objectives, the tags must be retained and must not affect fish growth or survival. We compared the effects of PIT tagging location on tag retention, growth, and survival of juvenile bluegills *Lepomis macrochirus* and yellow perch *Perca flavescens*. In total, 80 bluegills and 80 yellow perch from two size-classes (75–101 and 128–162 mm total length) were randomly assigned to a control or to one of three tagging location treatments: isthmus, body cavity, or dorsal musculature. Fish received daily *ad libitum* rations and were monitored for survival. On days 14, 28, and 42, the fish were measured, weighed, and checked for tag retention. Use of the isthmus as a tagging location resulted in lower tag retention for both species and both size-classes relative to the body cavity and dorsal musculature locations. Tagging location had no detectable effect on growth or survival responses for either species or either size-class. Thus, PIT tags that are implanted in the dorsal musculature of large juvenile bluegills and yellow perch and in the body cavity of small juvenile bluegills and yellow perch can have high retention with minimal adverse effects.

The use of tags in fisheries science has long been an important tool for collecting information on population parameters, including behavior, migration, growth, recruitment, and survival (Nielsen 1992). For tagging data to be reliable, two assumptions must be met: (1) tag loss must be minimal or known and (2) tags must not alter fish behavior, growth, or survival (Guy et al. 1996). Previous research has indicated that some tagging methodologies meet these assumptions better than others (McAllister et al. 1992; Mourning et al. 1994; Rikardsen et al. 2002). Failure to comply with these assumptions can compromise the validity of the conclusions obtained from a study (Robson and Regier 1966; McDonald et al. 2003; Rotella and Hines 2005), highlighting the need to select a tagging procedure that minimizes tag loss and negative effects on the study organism.

The use of passive integrated transponder (PIT) tags has gained considerable interest among fisheries biologists since the late 1980s. Passive integrated transponder tags can be used to monitor growth and survival of stocked fish for establishing or supplementing fisheries, are frequently utilized under experimental conditions, and can be applied in aquaculture settings (Baras et al. 2000; Ireland et al. 2002; Pirhonen et al. 2003; Cucherousset et al. 2007). In addition, information on movement or migration patterns can also be obtained by the use of PIT tags, especially when applied to small-bodied fish (Ombredane et al. 1998; Roussel et al. 2000; Cucherousset et al. 2005). Their small size, light weight, infinite life span, internal location, and almost unlimited number of individual codes make PIT tags particularly well suited for use with small-bodied fish (Nielsen 1992; Gibbons and Andrews 2004). The PIT tags are commonly inserted into the body cavity, dorsal musculature, or isthmus area of the fish (Brännäs and Alanärä 1993; Parker and Rankin 2003; Wagner et al. 2007; Isermann and Carlson 2008). Although there are several anatomical locations that can be used for tagging, few studies have examined tag retention, growth, and survival relative to multiple tagging locations within a single fish species.

Selecting the appropriate tagging location is important because for a given fish species, the rates of tag retention, growth, and survival often vary depending on the tagging location used (Navarro et al. 2006; Younk et al. 2010; Zaroban and Anglea 2010). For example, tag loss was higher for fingerling gilthead bream *Sparus auratus* (50–70 mm total length [TL]) that were tagged in the dorsal musculature than for fingerlings that were tagged in the abdominal cavity, and survival was lower for smaller individuals (Navarro et al. 2006). Tag retention was also higher for shorthead sculpins *Cottus confusus* (60–80 and 81–106mmTL) that were tagged in the body cavity than for fish that were tagged in the dorsal musculature; however, no differences in survival were observed between fish that were tagged at the two locations (Zaroban and Anglea 2010). Tag retention was lower for muskellunge *Esox masquinongy* (178–367 mm TL) that were PIT-tagged in the cheek than for

fish that were tagged in the dorsal musculature, but survival did not differ in relation to tagging location (Younk et al. 2010). For black rockfish *Sebastes melanops* (250–470mmTL), the isthmus provided the most appropriate PIT tag location among the several tagging locations examined, as tag retention was high and little to no adverse effects on the fish were observed (Parker and Rankin 2003).

The effects of PIT tags on bluegills *Lepomis macrochirus* and yellow perch *Perca flavescens* have not been examined despite the ecological and recreational importance of these species. In addition, many PIT tagging studies have focused on a single species and a single anatomical tagging location (Baras et al. 2000; Dare 2003; Ruetz et al. 2006; Isermann and Carlson 2008; Knudsen et al. 2009), whereas fewer studies have examined multiple tagging locations on multiple species. The response of fish to tagging and stress may differ depending on species and size (Winter 1983; Summerfelt and Smith 1990; Baras et al. 2000). Therefore, our objective was to compare PIT tag retention, growth, and survival of two size-classes (small and large) of juvenile bluegills and yellow perch that received tags at three implantation sites (dorsal musculature, isthmus, and body cavity).

Methods

Forty small (mean TL = 88 mm; range = 75–101 mm) and 40 large (mean TL = 146 mm; range = 128–162 mm) yellow perch and bluegills were collected from Lakes Cochrane and Sinai, and Gustafson Lake in eastern South Dakota by using C-phase, pulsed-DC electrofishing, angling, and cloverleaf traps. The selected sizes represented ages 1 and 2 for bluegills and ages 0 and 1 for yellow perch in South Dakota water bodies (St. Sauver et al. 2009), but they also represent sizes of mature and older adults observed in bluegill and yellow perch populations within other geographic regions (Carlander 1977). Furthermore, these sizes correspond to the smaller size ranges for these species and thus are of greater concern because the high tag : body mass ratios and small implantation sites may ultimately affect physiological and behavioral processes (Brown et al. 1999; Baras et al. 2000; Ruetz et al. 2006). Fish were transferred to a 674-L, flow-through raceway at South Dakota State University–Brookings and were fed a combination of chironomids and a commercial diet (Silver Cup Fish Feed) ad libitum for 2 weeks to allow for acclimation prior to the initiation of experiments. Water temperature was maintained at 19 °C, and overhead lighting provided a photoperiod of 14 h light : 10 h dark.

Fish from each species and size-class were randomly assigned to a control or to one of three tagging treatments (one intraperitoneal site and two intramuscular sites; 10 replicates/ treatment): (1) isthmus, extending forward

and between the gill openings; (2) dorsal musculature, adjacent to and 1 cm below the anterior portion of the dorsal fin; and (3) and body cavity, just anterior to the anus. All fish were anesthetized with tricaine methane sulfonate (MS-222; 50 mg/L), measured (nearest 1 mm TL), and weighed (nearest 0.1 g). Treatment fish were tagged by use of a 12-gauge hypodermic needle with a spring-modified syringe. The hypodermic needle and PIT tags (12.0 × 2.1 mm, 0.08 g in air; Biomark, Boise, Idaho) were disinfected with ethanol prior to each tagging event to reduce the likelihood of infection (Wagner et al. 2011). Control fish and the fish in the three tagging location treatments were given a unique fin clip to allow for external identification in the event of tag loss. Duration of handling (i.e., anesthetization, fin-clipping, weighing, measuring, tag insertion, and fish placement into the treatment tank) was recorded for all individuals. Handling of control fish was similar to that of treatment fish except that the control fish did not receive puncturing or tag insertion with a hypodermic needle. After tag implantation, fish received an ad libitum ration of a commercial diet (Silver Cup Fish Feed) and were monitored daily for survival. To allow for gut evacuation and reduce the possibility of bias in growth measurements, fish were unfed for 36 h before being measured for TL (mm) and weight (g) and monitored for tag retention on days 14, 28, and 42. Fish that died during the experiment were dissected to determine the cause of mortality (i.e., ruptured organs, infection, etc.).

Our statistical approach followed that of a similar study by Weimer et al. (2006), who compared two external transmitter types on two sizes of bluegills and yellow perch. Tag retention was compared among tagging locations within a species and size-class by use of a repeated-measures logistic regression (GENMOD procedure in the Statistical Analysis System [SAS]; SAS Institute 2003) sequentially through time (i.e., days 14, 28, and 42) with fish as the experimental unit. Relative daily growth rate (RDGR; $g \cdot g_{-1} \cdot d_{-1}$) was calculated to evaluate the effect of tagging location on fish growth. The RDGR on days 14, 28, and 42 was calculated as

$$\text{RDGR} = [(W_{\text{final}} - W_{\text{initial}})/W_{\text{initial}}]/\Delta t$$

where W = weight (g) and t = time (d). A mean initial weight was used as W_{initial} for the control treatment (Wagner et al. 2007). To achieve normalized residuals, the RDGR was $\log_{10}$ transformed prior to analysis. Differences in RDGR among tagging location treatments were compared through time by use of repeated-measures analysis of variance (MIXED procedure in SAS) with a variance components covariance structure. Tukey's post hoc comparisons were used to identify differences between tagging locations within a species and size-class. Life tables were constructed from survival data, and survival functions were fitted (LIFETEST procedure in SAS) for each species and size-class in relation to PIT tag location. A Wilcoxon chi-square (χ^2) test

was used to analyze cumulative survivorship among tagging location treatments within each species and size-class. The covariance matrix from the Wilcoxon statistics allowed the calculation of *Z*-scores for conducting individual pairwise comparisons. All comparisonwise differences were deemed significant at *P*-values less than 0.05.

Results

There was no difference in handling time among tagging location treatments for small or large bluegills (small: $F_{3, 35} = 0.18$, $P = 0.91$; large: $F_{3, 36} = 0.58$, $P = 0.63$) or for small yellow perch ($F_{3, 36} = 1.82$, $P = 0.16$; Table 1). Handling time was different among tagging locations for large yellow perch ($F_{3, 36} = 3.30$, $P = 0.03$); handling time for fish tagged in the isthmus was significantly greater than handling time for the control fish ($t_{1, 36} = -2.85$, $P = 0.03$; Table 1). Tag retention did not differ among tagging locations for large bluegills ($\chi^2 = 4.81$, df = 2, $P = 0.09$) and remained constant through time ($\chi^2 = 5.00$, df = 3, $P = 0.08$; Figure 1). Tag retention in small bluegills differed among tagging locations ($\chi^2 = 7.09$, df = 2, $P=0.03$); tag retention was lower for the isthmus treatment than for the body cavity treatment ($\chi^2 = 6.99$, df = 1, $P = 0.008$) but did not differ between the dorsal musculature treatment and the other two treatments ($\chi^2 = 2.65$, df = 1, $P = 0.10$). For small bluegills, tag loss was highest initially at days 14 and 28 and was lower by day 42 in comparison with day 14 ($\chi^2 = 5.38$, df = 1, $P = 0.02$; Figure 1).

Tag retention in large yellow perch was affected by tagging location ($\chi^2 = 7.71$, df = 2, $P = 0.02$) and was lower for the isthmus treatment than for the dorsal musculature treatment ($\chi^2 = 7.71$, df = 1, $P = 0.01$) or the body cavity treatment ($\chi^2 = 7.71$, df = 1, $P = 0.01$). Most of the tag losses in large yellow perch occurred by day 14 ($\chi^2 = 5.00$, df = 1, $P = 0.03$; Figure 1), and tag retention remained unchanged thereafter ($\chi^2 = 1.00$, df = 1, $P = 0.32$).

Table 1. Mean (SE in parentheses) handling time (s) for juvenile bluegills and yellow perch (two size-classes; small: 75–101 mm total length; large: 128–162 mm total length) that were passive integrated transponder tagged at one of three anatomical locations or that were not tagged (control). Asterisks denote significant differences between treatments ($P < 0.05$).

	Bluegills		Yellow perch	
Treatment	Small	Large	Small	Large
Control	59.3 (4.7)	58.5 (1.5)	76.7 (4.3)	63.8 (3.1)*
Dorsal musculature	60.3 (5.3)	62.5 (3.3)	67.1 (4.9)	65.9 (3.9)
Body cavity	62.2 (4.3)	60.8 (3.3)	83.3 (8.4)	72.8 (6.4)
Isthmus	63.7 (4.9)	63.8 (4.0)	87.2 (8.7)	84.8 (7.4)*

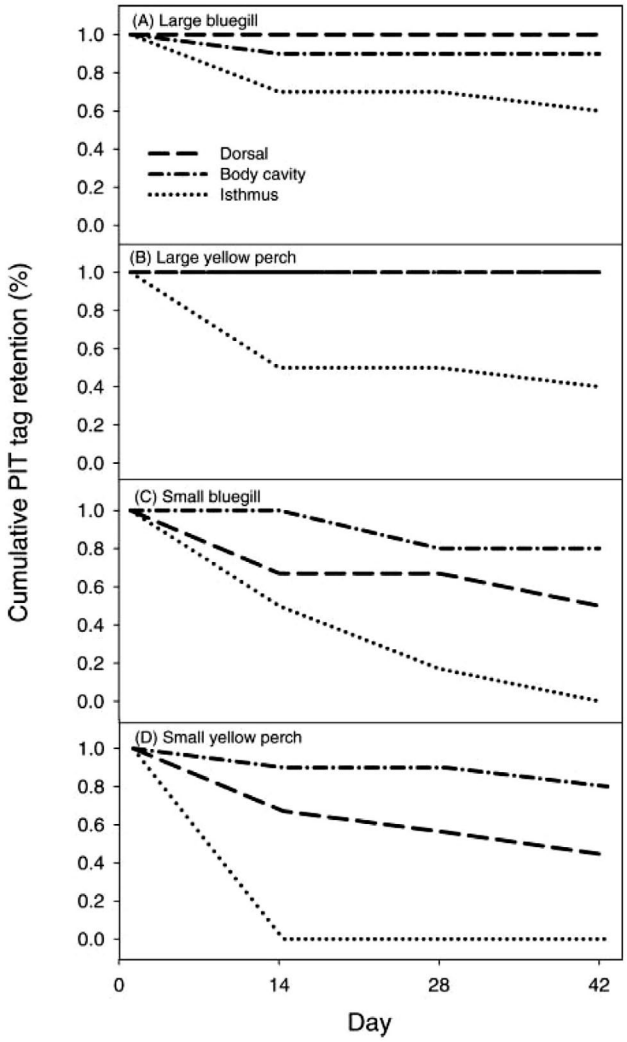

Figure 1. Cumulative tag retention over 42 d for **(A)** large bluegills (128–162 mm total length), **(B)** large yellow perch, **(C)** small bluegills (75–101 mm total length), and **(D)** small yellow perch that were passive integrated transponder (PIT)-tagged in the dorsal musculature, body cavity, and isthmus.

Tag retention in small yellow perch also differed among tagging locations (χ^2 = 13.88, df = 2, P = 0.001). Small yellow perch that were tagged in the isthmus lost more tags than those that were tagged in the body cavity (χ^2 = 13.99, df=1, P<0.001) or dorsal musculature (χ^2=7.13, df= 1, P = 0.008). Small yellow perch lost a significant number of their tags by day 14 (χ^2 = 13.96, df = 1, P < 0.001; Figure 1), and few tags were lost thereafter (χ^2 = 1.00, df = 1, P = 0.32).

Growth rates of fish were unaffected by PIT tag location. For large bluegills, there were no differences in RDGR among tagging location treatments

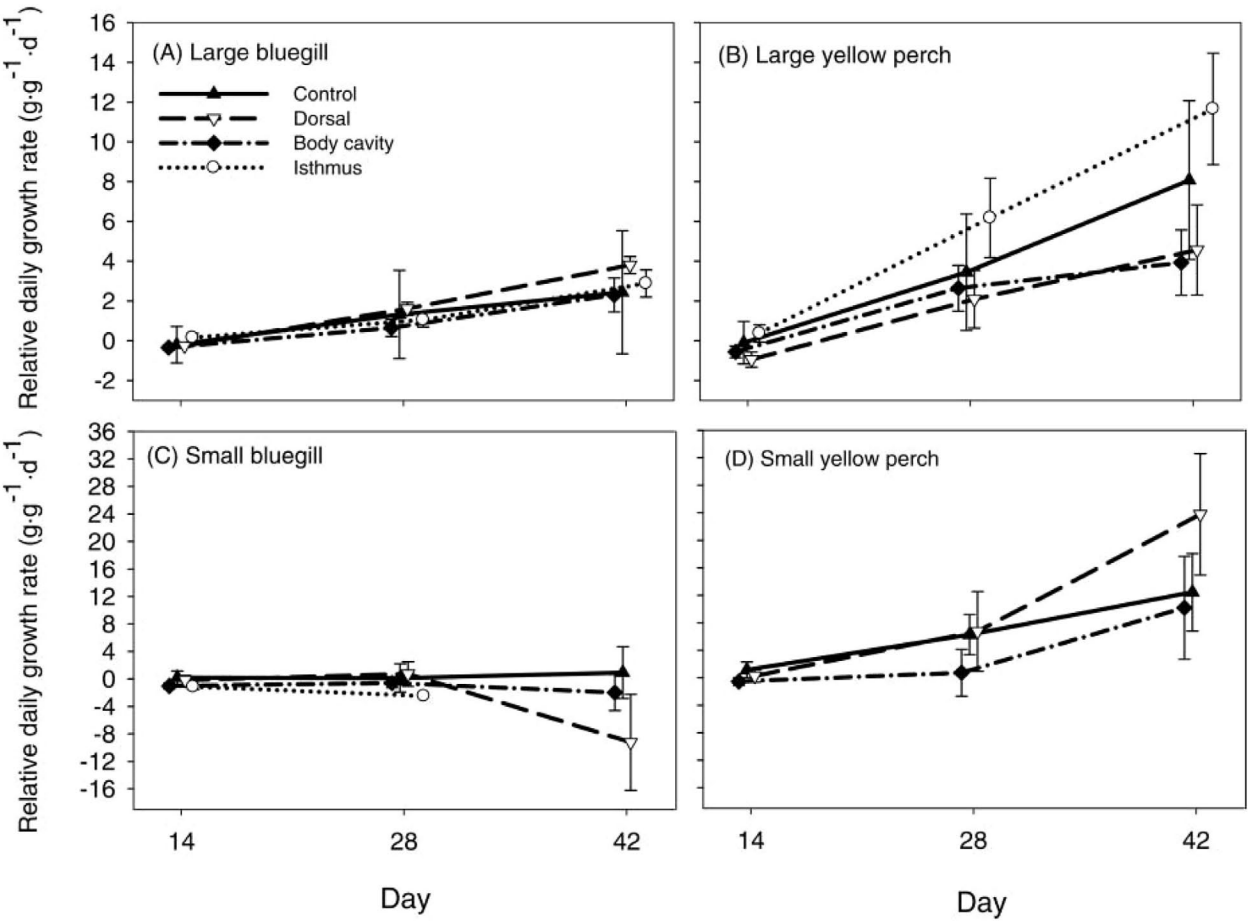

Figure 2. Mean (±SE) relative daily growth rate ($g \cdot g^{-1} \cdot d^{-1}$) over 42 d for **(A)** large bluegills, **(B)** large yellow perch, **(C)** small bluegills, and **(D)** small yellow perch that were passive integrated transponder tagged in the dorsal musculature, body cavity, or isthmus or that were not tagged (control).

($F_{3, 92}$ = 0.26, P = 0.85), but fish were larger on day 42 than on day 14 ($t_{1, 92}$ = −3.39, P = 0.003; Figure 2). Despite the difference in tag retention among small bluegills that were tagged in different locations, RDGR did not differ among tagging location treatments ($F_{3, 36}$ = 1.05, P = 0.38) and the fish were similar in size throughout the experiment ($F_{2, 36}$ = 1.67, P = 0.20; Figure 2).

The RDGR of large yellow perch was similar among tagging location treatments ($F_{3, 91}$ = 1.91, P = 0.13), and all fish were larger by day 42 in comparison with day 14 ($t_{1, 91}$ = −4.66, P < 0.0001; Figure 2). Small yellow perch exhibited no differences in RDGR among tagging location treatments ($F_{2, 58}$ =1.51, P= 0.23), but growth increased significantly from day 14 to day 42 ($t_{1, 58}$ = −4.13, P < 0.001) and from day 28 to day 42 ($t_{1, 58}$ = −2.88, P < 0.02; Figure 2).

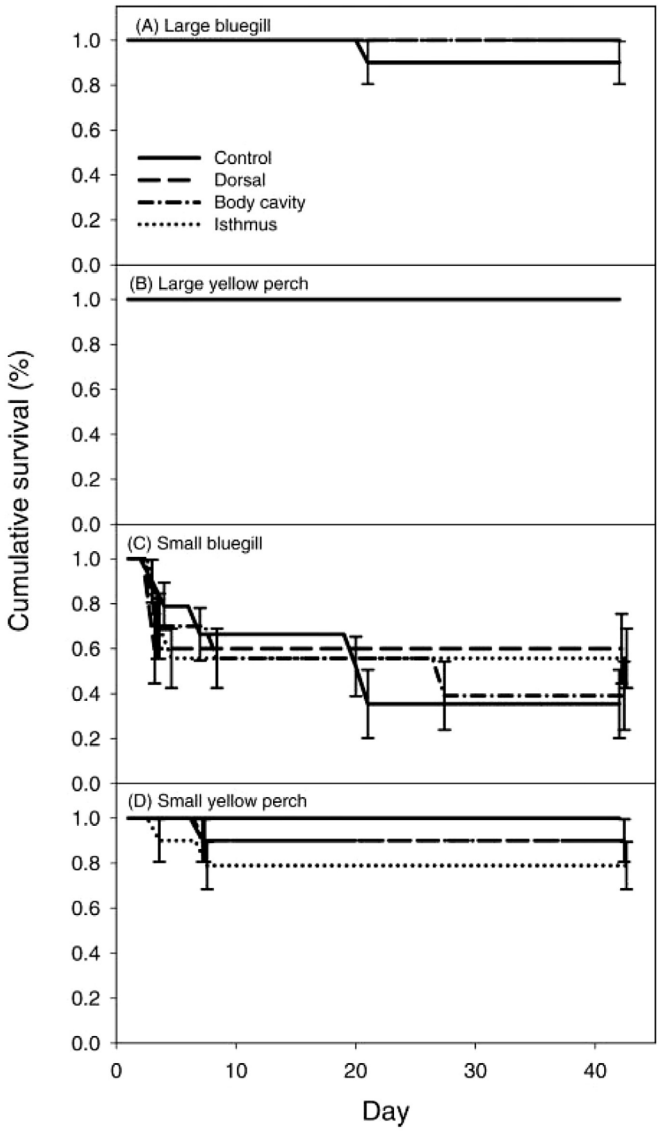

Figure 3. Mean (±SE) cumulative percent survival over 42 d for **(A)** large bluegills, **(B)** large yellow perch, **(C)** small bluegills, and **(D)** small yellow perch that were passive integrated transponder tagged in the dorsal musculature, body cavity, or isthmus or that were not tagged (control).

Fish survival was also unaffected by PIT tag location. Survival was similar among tagging location treatments for large bluegills (Wilcoxon $\chi^2 = 0.08$, df = 3, $P = 0.99$; Figure 3) and small bluegills (Wilcoxon $\chi^2 = 0.02$, df = 3, $P = 0.99$; Figure 3). No large yellow perch died during the experiment (Figure 3), and survival of small yellow perch was similar among all tagging location treatments (Wilcoxon $\chi^2 = 0.25$, df = 3, $P = 0.97$; Figure 3). Necropsies of the fish that died during the study provided no visual indication of bacterial infections, and there were no overt signs of ruptured organs or internal bleeding at any of the tagging locations.

Discussion

We evaluated the effects of three anatomical tagging locations on PIT tag retention, growth, and survival in two size-classes and two species. The main difference observed among tagging location treatments was related to tag retention: small and large juvenile yellow perch and small juvenile bluegills that were tagged in the isthmus retained fewer tags than fish that were tagged at the other locations. Differences in PIT tag retention among tagging locations have been demonstrated in previous studies (Navarro et al. 2006; Younk et al. 2010; Zaroban and Anglea 2010). Other studies have documented high PIT tag retention rates for fish that were tagged in the isthmus (Brännäs and Alanärä 1993; Parker and Rankin 2003); however, the fish in those studies were generally much larger (Arctic char *Salvelinus alpinus,* 48–328 g: Brännäs and Alanärä 1993; black rockfish, 250–470 mm: Parker and Rankin 2003) than the individuals that were tagged in our study (75–162 mm; 4–93 g). We chose to evaluate the isthmus tagging location because (1) this location is commonly used (Brännäs and Alanärä 1993; Parker and Rankin 2003); (2) it provides an alternative to the application of tags in the dorsal musculature, as such tags may be incidentally ingested by anglers that harvest the fish and consume the fillet; and (3) it provides an alternative to tagging in the body cavity, which may cause organ damage during implantation or may result in tag expulsion (Ward et al. 2008). However, low tag retention at the isthmus site for the fish sizes and species we examined makes it an unsuitable site for broad application in PIT tag studies.

Similar to other studies (Parker and Rankin 2003; McCormick and Smith 2004; Ruetz et al. 2006; Wagner et al. 2007), we found high tag retention rates for all sizes of juvenile bluegills and yellow perch that were tagged in the body cavity. The body cavity provided the highest tag retention for small-sized fish (75–101 mm TL) of both species; however, we caution that tag expulsion from the body cavity may be higher for smaller fish than for larger fish (expulsion rate = 20% and 5%, respectively; Navarro et al. 2006). In contrast, the dorsal musculature appears to be the most suitable tagging location for large juvenile yellow perch and bluegills, as indicated by 100% tag retention for each of these groups. Tag retention was also high for muskellunge fingerlings that were tagged in the dorsal musculature (Wagner et al. 2007; Younk et al. 2010). Among brook trout *Salvelinus fontinalis* (99–302 mm TL) and brown trout *Salmo trutta* (122–511 TL), tag retention was greater for fish that were tagged in the dorsal musculature than for those tagged in the body cavity (Dieterman and Hoxmeier 2009). Thus, PIT tagging in the body cavity may be most appropriate for small-bodied fish, whereas the dorsal musculature may be the more appropriate tagging location for large-bodied individuals, in which tag retention is maximized.

We found that PIT tagging had minimal effects on fish growth and survival in this study, regardless of fish species or size. Other tag types are often observed to affect fish growth and survival (i.e., Paukert et al. 2001; Rikardsen et al. 2002; Strand et al. 2002; Weimer et al. 2006). In contrast, PIT tags in general appear to have minimal effects on fish growth rates (brown trout, 55–127 mm fork length: Ombredane et al. 1998; mottled sculpin *C. bairdii*, 55–59 mm TL: Ruetz et al. 2006; muskellunge, mean TL = 284 mm: Wagner et al. 2007; zander *Sander lucioperca*, 188 mm standard length: Hopko et al. 2010) or fish survival (largemouth bass *Micropterus salmoides*, mean TL = 259 mm: Harvey and Campbell 1989; brown trout, 55–127 mm fork length: Ombredane et al. 1998; European bullhead *C. gobio*, 70–105mmTL: Bruyndoncx et al. 2002; mottled sculpin, 55–59 mm TL: Ruetz et al. 2006; round goby *Neogobius melanostomus*, <105 mm TL: Cookingham and Ruetz 2008; black crappie *Pomoxis nigromaculatus*, 175–328 mm TL: Isermann and Carlson 2008; zander, mean standard length = 188 mm: Hopko et al. 2010). Survival of small bluegills in all treatments, including the control, was lower than expected in comparison with the survival of larger conspecifics and both yellow perch size-classes. Lower survival among small bluegills could have been related to interspecific or intraspecific competition for food resources (although the fish were fed ad libitum) or agonistic behaviors from larger bluegills or both sizes of yellow perch; however, the lower survival in small bluegills was not attributed to PIT tag effects, as no differences in survival were observed among the treatments. Thus, in contrast to other tagging options, PIT tagging appears to have minimal effects on growth or survival of fish regardless of species, size, or anatomical tagging location.

Due to high tag retention and negligible effects on fish growth and survival, PIT tags appear to be an appropriate tool for fisheries researchers and managers to use in monitoring behavior, migration rates, growth, recruitment, and survival across a wide range of fish sizes and species. Our results show that PIT tags have minimal effects on growth and survival of juvenile bluegills and yellow perch. Tag retention was highest for small bluegills and yellow perch that were tagged in the body cavity and for large bluegills and yellow perch that were tagged in the dorsal musculature, suggesting a potential size-related difference among tagging locations for these two species. Thus, to maximize tag retention, managers should carefully select the proper PIT tagging location for the species and size-class of interest. However, if only one tagging site must be selected, the body cavity provides the most optimal tagging location for both sizes of juvenile bluegills and yellow perch.

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