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RESEARCH PAPER

# Holding of juvenile salmonids for surgical implantation of electronic tags: a review and recommendations

**Eric W. Oldenburg · Alison H. Colotelo · Richard S. Brown · M. Brad Eppard**

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**Abstract** Many telemetry-based studies require that fish be sampled from the wild and then held both prior to and after the implantation of an electronic tag. However, the effects of such holding (or the lack thereof) have yet to be studied intensively. Presurgical holding often occurs to facilitate logistical needs of research projects and as an attempt to minimize negative physiological effects due to capture and handling stress. Further, post-surgical holding time and conditions greatly influence the physiological state of fish prior to being returned to the wild. This paper reviews pertinent studies pertaining to the effects of surgical holding on the behavior, physiology, and survival of fishes, with particular emphasis on juvenile salmonids. The effects of individual aspects of surgical holding such as handling, water quality, light conditions, holding density, metabolic scope, and duration of holding are reviewed. Recommendations regarding certain aspects of surgical holding are offered with a goal of reducing bias related to the surgical process.

E. W. Oldenburg (✉) · A. H. Colotelo · R. S. Brown
Ecology Group, Pacific Northwest National Laboratory, MSIN K6-85, P.O. Box 999, Richland, WA 99352, USA
e-mail: eric.oldenburg@pnl.gov

M. B. Eppard
US Army Corps of Engineers, Portland District, P.O. Box 2946, Portland, WA 97208-2946, USA

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## Introduction

Holding fish prior to and after surgical implantation of electronic tags is an important aspect of telemetry studies, yet little information exists regarding this topic. A primary assumption of telemetry studies is that surgically implanted fish are representative of the population of inference. However, the process of surgical implantation has the potential to introduce bias to the sample and alter aspects of fish swimming ability (Adams et al. 1998a; Wagner and Stevens 2000; Brown et al. 2006), growth (Martinelli et al. 1995; Adams et al. 1998b), physiology (Jepsen et al. 2001; Close et al. 2003), and survival (Adams et al. 1998a; Jepsen et al. 1998; Brown et al. 2010). Therefore, it is desirable to minimize these effects so that reliable inferences can be made regarding the population of interest.

Stressors, such as those associated with surgical holding, affect the physiology of fish in a predictable manner, including primary, secondary and tertiary responses. The primary response includes measurable increases in the circulation of catecholamines and corticosteroids (Barton 2002). If a stressor persists long enough, or is of sufficient severity, the stress response escalates to include changes in metabolism, hydromineral balance, and impairments in immune function (Barton 2002). Finally, whole-organism performance (e.g., behavior, growth, disease resistance) and survival may be affected. There are a wide range of stressors associated with the surgical process including but not limited to: handling, confinement, water quality, light intensity, and holding density.

## Factors to consider

### Handling

Fish are handled during the surgical implantation of electronic tags and while being removed from and returned to their environment. In addition, handling may include measuring length and weight, transportation, and fin clipping for identification purposes. All of these processes can elicit a stress response resulting from air exposure and physical contact (Strange et al. 1977; Ferguson and Tufts 1992; Davis and Schreck 1997). The duration of air exposure has been associated with the magnitude of the stress response and behavioral impairment of several species (e.g., Ferguson and Tufts 1992; Thompson et al. 2008). While the effects of air exposure specific to the surgical process have not been investigated, the effects of air exposure during catch-and-release angling have been extensively studied for several species such as rainbow trout *Oncorhynchus mykiss* (Ferguson and Tufts 1992), bluegill *Lepomis macrochirus* (Gingerich et al. 2007), and bonefish *Albula vulpes* (Suski et al. 2007), and it is generally recommended that the duration of air exposure should be minimized (Cooke and Suski 2005).

Removal of fish from and replacement to their original environment (e.g., natural systems or aquatic facilities) is often done with the use of nets. Nets, and any other surface that may come into contact with the fish, may be a source of injury to the epithelial layer covering the surface of the body (Barthel et al. 2003; Colotelo et al. 2009), and these injuries may result in infections by pathogens. Injuries and associated infections may impair the behavior and survival of the fish after release, making inferences regarding the population of interest difficult or impossible. Among studies focused on recreational angling it has been recommended that knotless rubber nets be used to minimize mucous loss resulting from abrasion with the net (adult bluegill, Barthel et al. 2003; adult trout, Dedual and Shorland 2006). Water-to-water transfer of juvenile salmonids has been shown to reduce mortality rates compared with those which are netted using traditional dip nets (Flagg and Harrel 1990); thus, the use of rubber or sanctuary nets is advised for handling fish throughout the surgical process (Fig. 1).

### Water quality

It has been suggested that water quality during experimental holding periods is the most important factor for increasing survival and minimizing stress in fish (Klontz 1995; Casebolt et al. 1998). There are many factors which influence water quality including dissolved gas levels, water temperature, flow, and water exchange rate.

Westers (2001) stated that dissolved oxygen is the first limiting water quality factor for fish, with the recommendation that levels near saturation are desirable. For this reason, supplemental oxygen should be applied to surgical holding tanks whenever necessary in order to keep dissolved oxygen levels at or near saturation (i.e., 100%). This is particularly important for surgery related holding, as fish captured and held before and after surgery will be recovering from physiological stressors, and it has been shown that juvenile steelhead *Oncorhynchus mykiss* [mean weight ($\pm$SE) = 92 $\pm$ 7 g; Barton and Schreck 1987] and juvenile coho salmon *Oncorhynchus*

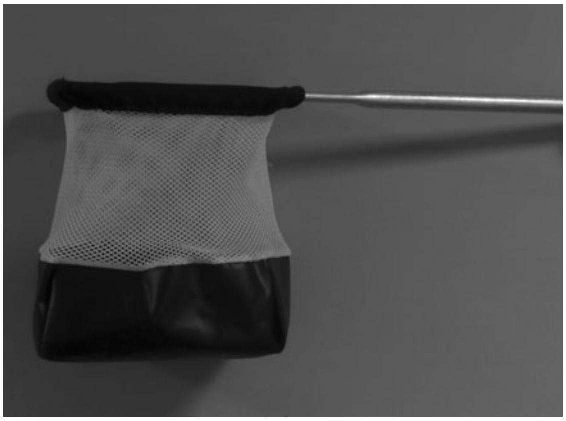

**Fig. 1** Sanctuary nets, used in fisheries research, are lined along the bottom with vinyl to retain water so fish are not air exposed and abrasion with net is minimized

*kisutch* (weight range = 67–206 g; Davis and Schreck 1997) increase oxygen consumption following handling. Despite the need for sufficient levels of oxygen, it is important not to over saturate the water with gases. High total dissolved gas concentrations (e.g., >120%) can lead to gas-bubble disease that occurs when nitrogen comes out of solution and creates gas emboli in the blood and tissues (Bouck 1980). Gas-bubble disease can interrupt organ function and ultimately alter behavior and result in mortality (Bouck 1980).

Water temperature is another important factor to consider, particularly because fish are poikilothermic. When exposed to water temperatures >20°C, salmonids may experience increases in stress, immune suppression, and susceptibility to infection (Klontz 1995). Lower than optimal water temperatures reduce the rate of physiological processes (Wedemeyer 1996a) which may decrease the rate of healing of surgical incisions. Therefore, we recommend that surgeries not be performed when water temperatures approach species specific thermal maximums or minimums. In addition to the effects of extreme temperatures, rapid changes in temperature (i.e., thermal shock) may also influence the physiology and behavior of fish. To avoid negative consequences of thermal shock, it is recommended that researchers hold and perform surgeries on fish at the ambient water temperature from which the fish were sampled, or at a temperature to which fish are acclimated (Jepsen et al. 2002; Portz et al. 2006).

Physiological processes such as healing and immune function are temperature mediated. Juvenile Chinook salmon *Oncorhynchus tshawytscha* (length range = 95–121 mm) held at higher water temperatures (i.e., 17°C) generally had greater irritation and inflammation at the incision site when compared to those held at lower water temperatures (i.e., 12°C) (Deters et al. 2010). Similarly, mortality rates were higher for juvenile Chinook salmon (length range = 95–121 mm) held at 20°C post-surgery compared to those held at 12°C (Panther et al. 2010). Electronic tag retention has also been shown to be influenced by temperature, with greater tag loss among juvenile Chinook salmon (tag burden range = 3.5–6.7%) held at higher water temperatures (i.e., 17°C) as compared to those held at lower temperatures (i.e., 12°C) (Deters et al. 2010). When appropriate, we recommend that water temperature be included as an independent variable in surgery related studies as it influences rates of processes such as recovery from stressors, healing, and pathogen propagation.

## Holding density

Holding density can influence the metabolic scope, health and behavior in salmonids and is directly related to water quality parameters (Wedemeyer 1996a). Excessively high rearing densities can result in increased mortality and decreased final weight, length, condition factor, and food conversion efficiency among salmonids (Fagerlund et al. 1981; Poston and Williams 1988; Procarione et al. 1999). Although it has been suggested that increased dissolved oxygen levels may support increased stock densities (Westers 2001), higher densities of fish may promote accelerated depletion of dissolved oxygen levels (Portz et al. 2006) and can increase the rate of propagation and magnitude of infection (Wedemeyer 1996a). Holding density also influences social behaviors such as fin nipping, ramming, and aggression, which can result in fin degradation, scale loss and other injuries (Wedemeyer 1996a). This factor is important with respect to water quality as well as social assemblage and should be further investigated relative to pre- and post-surgical holding.

## Light conditions

The behaviors of animals, including fish, are often influenced by light intensity and photoperiod (Boeuf and Le Bail 1999). For fish being held in artificial settings (e.g., indoors) this can be an important factor for conserving natural behaviors. For fish being held outdoors, often in direct light, fish behavior (e.g., inability of fish to find photic refuge in deep water or shade) and water conditions (e.g., fluctuating water temperature, increased algal growth) can be affected. Therefore, tank covers are often used to prevent these influences. The benefits of using tank covers have not been extensively studied; however, Atlantic salmon parr *Salmo salar* (length range = 95–126 mm) and brook trout *Salvelinus fontinalis* (length range = 117–212 mm) have demonstrated an affinity for covered areas in holding tanks (Gibson 1978). Reduced light intensity has also been successful in mitigating transport stress. Chinook salmon had a

25% reduction in hyperglycemia and hyperchloremia (measures of physiological stress) under darkened conditions (Wedemeyer 1996b). Light conditions in holding tanks should be considered when designing studies and we recommend that the effects of light conditions be further evaluated with respect to stress levels and the behaviors of fish during holding.

## Metabolic scope

Metabolic scope is another factor that should be considered when planning for surgical holding. Specific dynamic action (SDA) is the portion of metabolic scope allocated to processing and assimilating food. Studies on salmonids have demonstrated that SDA remains remarkably constant during exercise and that the energetic demands of swimming are met only after the energetic demands of SDA are met (Alsop and Wood 1997; Thorarensen and Farrell 2006). Thus, it is possible that energetic needs of other processes (e.g., compensatory stress responses, immune function) may also be met only after the energetic needs of SDA. Therefore, fish should not be fed throughout the surgical holding process. Further, it may be beneficial to hold fish long enough to allow SDA to cease prior to surgery. However, increasing the duration of holding for the purpose of allowing SDA to cease may increase the magnitude of other stressors resulting in a net physiological effect that is detrimental to the fish. Research is needed to investigate the relationship between SDA, metabolic scope, physiological stress and holding duration.

Gastric evacuation is an important component of SDA. Throughout the literature, estimates of time until complete gastric evacuation are highly variable among and within species depending on study-specific variables (Table 1). Numerous studies have demonstrated that gastric evacuation rates increase with increasing water temperature (Windell et al. 1976; Doble and Eggers 1978; Brodeur and Pearcy 1987; He and Wurtsbaugh 1993; Principe et al. 2007). However, it is important to note that individuals may increase consumption with increasing water temperatures because of greater metabolic demands at these temperatures (Kolok and Rondorf 1987). Fish size also influences gastric evacuation rate, with larger fish having reduced metabolic rates and therefore slower evacuation. For example, juvenile sockeye *Oncorhynchus nerka* and Chinook salmon have demonstrated decreasing evacuation rate with increasing fish size (Table 1) (Doble and Eggers 1978; Kolok and Rondorf 1987; Principe et al. 2007); however, no significant difference was detected in evacuation rate among different size classes of brown trout *Salmo trutta* (He and Wurtsbaugh 1993).

While time to complete gastric evacuation is often used as justification for pre-surgical holding times, SDA is primarily a post-absorptive effect, consuming a portion of the metabolic scope even after gastric contents have been evacuated (Fitzgibbon et al. 2007). Consequently, time until gastric evacuation alone may not provide the optimal estimate for pre-surgical holding time. Rainbow trout (mean weight = 808 ± 47 g) have been studied to examine the timing of SDA through measures of gastrointestinal blood flow, heart rate and oxygen consumption. Postprandial gastrointestinal blood flow peaked at 136% above baseline 11 h after feeding, heart rate peaked at 110% above baseline after 14 h, and oxygen consumption peaked at 96% above baseline at 27 h (Eliason et al. 2008); however, these timelines may differ depending on water temperature, species, age class, and fish size.

## Holding duration

Considerations for determining fish holding duration include physiological stress responses to the surgery as well as to pre- and post- surgery holding conditions. Pre-surgery holding duration is generally dictated by SDA and minimizing other stressors, while post-surgery holding duration is dictated by surgical recovery (e.g., minimizing stress levels, eliminating the effects of anesthesia, detecting immediate surgery-related mortalities).

Pre-surgery holding should allow the energetic demands of SDA to be reduced while minimizing the magnitude and duration of stressors [e.g., confinement (Strange et al. 1977, 1978)]. Recommended practice for the fasting of salmonids prior to transportation is 48–72 h (Wedemeyer 1996b), while many studies report fasting fish for 24–48 h prior to surgery (e.g. Adams et al. 1998a; Murray 2002). Based on the literature (see Table 1), 48 h would provide sufficient time for SDA to cease in most salmonids in water >5°C. However, temperature and species specific effects should be evaluated. For example, brook trout require 72 h post-feeding for

**Table 1** Evacuation rate and time to complete (90–100%) gastric evacuation and mean length for salmonids with varying water temperatures and prey types

| Species | Length (mm) | Prey | Evacuation rate (% $h^{-1}$) | | | | | Time to evacuation (h) | | | | | Source |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | <5°C | 6–10°C | 11–15°C | 16–20°C | >20°C | <5°C | 6–10°C | 11–15°C | 16–20°C | >20°C | |
| BRK | 152 | Larval insects | 0.6 | 1.3 | 1.4 | 1.5 | | | | | | | Sweka et al. (2004)[b] |
| BRN | 404 | Fingerling RBT | 7 | 10 | 15 | 18 | 29 | >34 | 34 | 24 | 20 | 14 | He and Wurtsbaugh (1993)[c] |
| RBT | 150 | *Asellus militarus* | | | | | | | 16 | | | | Kionka and Windell (1972)[d] |
| RBT | 150 | Pellets | | | | | | 72 | 44 | 27 | 16 | | Windell et al. (1976)[e] |
| RBW | 150 | Oligochaetes | | | | | | 59 | 38 | 25 | 16 | | Windell et al. (1976)[e] |
| CHK | 129 | Mealworms | | 14 | 9 | | | | | | | | Kolok and Rondorf (1987)[b] |
| CHK | 35–70[a] | Chironomids | | 21 | 29 | 33, 37 | | | | | | | Principe et al. (2007)[b] |
| CHO | 147 | Euphausiids | | | 8 | | | | | 29 | | | Brodeur and Pearcy (1987)[c] |
| CHO | 206 | Euphausiids | | | 11 | | | | | 14 | | | Brodeur and Pearcy (1987)[c] |

*BRK* brook trout, *BRN* brown trout, *RBT* rainbow trout, *CHK* Chinook salmon, *CHO* coho salmon

[a] Length range, [b] percent evacuated not reported, [c] time to 90% evacuation, [d] time to 98% evacuation, [e] time to 100% evacuation

oxygen consumption to begin to decrease (Beamish 1964) while rainbow trout oxygen consumption peaked at 27 h (Eliason et al. 2008). Wedemeyer (1996a) reported that for rainbow trout metabolic rate began to decline only after 48 h post-feeding. Increasing holding duration prior to surgery may reduce the portion of metabolic scope that is needed for SDA at the time of surgery. Thus, we recommend holding hatchery origin fish long enough to allow SDA to cease. When all factors (e.g., holding-related stressors) are considered, the net physiological effect of increasing holding duration for wild-caught fish in order to allow SDA to cease remains unknown and warrants further investigation.

The duration of post-surgery holding is important for telemetry studies, because this period allows fish to recover from stressors related to surgery and return to as near a representative state as possible. Post-surgery holding duration for migration studies involving wild fish have ranged from release within 30 min of recovery [brown trout (Ovidio et al. 1998); juvenile Atlantic salmon (Moore et al. 1998)], to overnight [juvenile brown trout and Atlantic salmon (Jepsen et al. 1998)] and up to several days [juvenile Atlantic salmon (Voegeli et al. 1998); juvenile Atlantic salmon and sea trout *Salmo trutta* (Aarestrup et al. 2002)]. However, recommending a best practice is difficult because little work has been conducted to evaluate the magnitude and duration of the stress response subsequent to surgical procedures. Following an acute disturbance (e.g., confinement, handling, air exposure), oxygen consumption and plasma cortisol concentrations (primary stress response) in fish increase within a few minutes of the stressor (Wendelaar Bonga 1997). In juvenile Chinook salmon [length range = 100–142 mm (Strange et al. 1978); mean weight range = 17–31 g (Barton et al. 1986)] and juvenile coho salmon [weight range = 67–157 g (Davis and Schreck 1997)] plasma cortisol concentrations return to basal levels 3–6 h after an initial increase. However, stress can be cumulative in fishes (Barton et al. 1986) and the surgical process exposes fish to multiple stressors. For juvenile Chinook salmon (mean weight = 11 g) exposed to multiple handlings or agitations, plasma cortisol and lactate concentrations peaked 0.5–1.0 and 0.5–1.5 h, respectively, after the final stressor (Mesa 1994). These plasma concentrations returned to baseline levels 6 h after the final stressor, while

**Table 2** Summary of recommendations for pre- and post-surgery holding factors for juvenile salmonids

| Factor | Recommendation |
|---|---|
| Air exposure | Minimal |
| Net type | Rubber or sanctuary nets |
| Dissolved oxygen | Near saturation (e.g., 100%) |
| Water temperature | Hold and perform surgeries at temperature to which fish are acclimated |
| Holding density | Dependent on water quality, species, etc. |
| Light condition | Consider shaded or shielded tanks or natural light conditions |
| Feeding | Withhold feed pre- and post-surgery |
| Holding duration pre-surgery | Dependent on study and species |
| Holding duration post-surgery | Dependent on study and species |

plasma glucose levels peaked 3 h after the final stressor and returned to baseline levels 24 h after the final stressor (Mesa 1994). These results suggest that juvenile salmonids should be held for a minimum of 24 h post-surgery in order for stress levels to return to baseline; however, species specific holding times should be investigated in future research. In addition, holding of fish for 24 h after surgery may allow for the detection of any immediate, surgery-related mortalities.

Holding durations may be influenced by the facilities available to the researcher. Recovery characteristics presented in this review relate to ideal holding conditions, in which density and water quality are optimal for fish survival. Many of the studies cited also included the use of hatchery or laboratory reared fish, which may not be as affected by confinement as wild fish. Wild fish held in laboratory settings may respond to stressors related to holding and surgery differently than hatchery-reared fish. Therefore, fish origin (i.e., hatchery versus wild) should be carefully considered when determining holding duration and future research should be conducted to investigate the effects of pre- and post-surgery holding for both hatchery-reared and wild fish.

### Holding location

Many of the elements of pre- and post-surgery holding of fish are affected by the environment in which surgeries are performed. A review of studies utilizing the surgical implantation of telemetry tags (not limited to juvenile salmonids) reported that 59.5% of surgeries were performed in the field, while only 13.4% of studies occurred in a hatchery or laboratory setting and 2.8% reported a combination of field and laboratory settings (24.3% of studies investigated did not report this information) (Thiem et al. 2010). Holding location influences many of the aforementioned factors (e.g., water quality, holding duration, etc.) and should be considered when planning for the surgical process; albeit, logistical constraints may exist.

## Conclusions

The current review highlights important factors that should be considered when holding fish pre- and post-surgery, as well as some general recommendations (summarized in Table 2). It has been suggested that water quality factors, including water temperature, are the most important considerations for pre- and post-surgical holding of fish as poor water conditions can increase the effect of stressors. Although all of the factors outlined in this review have been investigated in the literature, there is a paucity of information specific to holding as it relates to surgical processes. There is also a lack of understanding on how these factors are related and influence each other. This review identifies important research needs that will aid in a better understanding of the effects of surgery and related holding on juvenile salmonids as well as provide information that will assist in the creation of reliable recommendations for pre- and post-surgical holding conditions.

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