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RESEARCH PAPER

Surgical implantation techniques for electronic tags in fish

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Abstract Intracoelomic implantation of transmitters into fish requires making a surgical incision, incision closure, and other surgery related techniques; however, the tools and techniques used in the surgical process vary widely. We review the available literature and focus on tools and techniques used for conducting surgery on juvenile salmonids because of the large amount of research that is conducted on them. The use of sterilized surgical instruments properly selected for a given size of fish will minimize tissue damage and infection rates, and speed the wound healing of fish implanted with transmitters. For the implantation of transmitters into small fish, the optimal surgical methods include making an incision on the ventral midline along the linea alba (for studies under 1 month), protecting the viscera (by lifting the skin with forceps while creating the incision), and using absorbable monofilament suture with a small-swaged-on swaged-on tapered or reverse-cutting needle. Standardizing the implantation techniques to be used in a study involving particular species and age classes of fish will improve survival and transmitter retention while allowing for comparisons to be made among studies and across multiple years. This review should be useful for researchers working on juvenile salmonids and other sizes and species of fish.

Keywords Telemetry · Surgery · Sterilization · Suture · Knot · Fish · Implantation

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Introduction

The general guidelines for surgical procedures on fish are to know the anatomy, minimize tissue damage and surgery times, and to not cut unknown tissues (Murray 2002). Adherence to these guidelines minimizes discomfort to the fish because small, clean wounds that are sutured properly heal quickly with minimal complications (Mulcahy 2003). Therefore, the size of the incision, suture material, and needles used are dependent largely on the size of fish involved and the subsequent size of transmitter to be implanted. Along with minimizing the amount of tissue damage, it is important to use general aseptic techniques to minimize the risk of infections (Mulcahy 2003; CCAC 2005). These techniques help to maximize healing rates, retain the transmitter in the body cavity of the fish, and reduce the chance of introducing pathogens into the body.

We review the literature available related to making incisions, incision closure, and other related surgery techniques. In some cases, the only available information pertains to studies performed on mammals. This review is intended to provide the best available scientifically based methods for implantation of acoustic transmitters in juvenile salmonids. In many cases, the optimal procedures are unknown, so suggestions for future research are provided as well.

Surgical instruments

Implantation of acoustic transmitters requires the use of surgical tools to create and close the incision. Although many tool options are available and personal preference may differ among surgeons, it is important to use tools that are appropriate for the application (Dunn and Phillips 2004). For implantation of juvenile salmon, the tools should be smaller and more delicate than those used on adults because the fish and transmitters are smaller.

Typically, hundreds of juvenile salmon smolts are tagged daily on some large projects in the Pacific Northwest; thus, choosing ergonomic and efficient tools is important. A scalpel, forceps, and needle holder are needed for creating and closing the incision. Scalpel blades that remain sharp after repeated use are ideal for both cost and time efficiency purposes. Due to the thin body wall of juvenile salmon and the small length of incision needed, the use of large surgical blades (e.g., size 10) is not suggested. A smaller blade, such as a size 15, is preferred, while a micro blade, such as the Becton–Dickinson Micro-Sharp Blade or Micro-Unitome Knife i (~1.5–3.0 mm cutting tip), is a good alternative. Some surgeons prefer to use the micro blade to start the incision (i.e., make a cut through the skin), followed by a surgical blade (e.g., size 15) to finish the incision through to the abdominal cavity. Surgeons implanting transmitters in juvenile salmonids typically use small forceps (~10 cm long) with blunt curved tips that are approximately 0.5 mm wide. In addition, relatively small (~12 cm long) needle holders with a small tip width (~1.5 mm) and clamping length (~8.5 mm) commonly are used. Having needle holders with integrated scissors (i.e., Olsen-Hegar needle holders) also reduces the need for additional tools.

It is important to properly clean and dry surgical tools used in the aquatic environment to avoid corrosion and other physical damage. The use of a surgical instrument lubricant (commonly known as "surgical milk") can also extend life of the instruments and minimize corrosion. Also recommended is the veterinary practice of surgical instrument sterilization prior to and between surgical procedures on fish (CCAC 2005). Typically, the need for sterilization and disinfection is considered on a scale that depends on the risk of infection to the patient, based on use of a given instrument (Spaulding 1968). In the case of fish surgery, the risk of infection is considered high for any instrument used in the surgical procedure. As such, these tools are considered critical items and require the most stringent of sterilization procedures because any microbial contamination could result in disease transmission (Spaulding 1968). Sterilization is relevant not only to the surgical instruments but also to the transmitters being implanted (Mulcahy 2003) and possibly the surgical gloves (Korniewicz et al. 1991). Unlike human physicians and veterinarians, the majority of fish surgeons use nonsterile latex gloves, likely because these surgeries are performed in a nonsterile environment.

Heat and chemical sterilization can completely eliminate all microorganisms, including bacterial endospores and all viruses and bacteria that are resistant to disinfectants. The various sterilization and disinfection techniques, their limitations, and their efficacy are summarized in several reviews focused on human medicine (e.g., Rutala 1990, 1996; Favero and Bond 1991; Jacobs et al. 1998; Rutala and Weber 2004). Fish surgeries should entail as many sterile practices as possible within the limitations of the species and field conditions involved. As indicated by (AFS (American Fisheries Society) 2004), conducting surgical procedures on fish under completely sterile conditions is not always possible, but utilizing all precautionary methods available will help to minimize bacterial contamination of the incision and body cavity. Despite the insistence of some veterinarians to the contrary, the fact remains that fish produce a constantly streaming mucus layer containing high numbers of colonizing bacteria (Austin and Austin 1987) through which an incision must be made. Removal of this mucus layer using an antiseptic treatment is not recommended because wound healing may actually be hindered due to the loss of this physical barrier to pathogenic microorganisms that also contains lysozymes, proteolytic enzymes, and other compounds that inhibit bacterial colonization and infiltration (Alexander and Ingram 1992). Similarly, once a surgery is completed a sterile wound area cannot be maintained as with mammalian surgeries because fish must be returned to their water supply which is inherently unsterile regardless of its source (e.g. laboratory or natural environment). The only know study to examine the effects of water infiltration into the coelom of fish showed no adverse effects to bluegill at two temperatures (Chomyshyn et al. 2011).

Heat sterilization (e.g., using an autoclave) is the most effective way to kill 100% of all microorganisms. Typically, autoclaves should maintain a temperature of at least 246°C (475°F) for 30 min using either dry or steam heat. Dry heat autoclaves are used for surgical products and instruments that are sensitive to the moisture created in steam heat autoclaves that use heated water vapors. This process of sterilization is effective but energy-intensive and time-consuming, and may be impractical under field conditions when electricity is not available (e.g., when hiking into a remote site). Some large autoclaves are not easily moved, whereas others (such as those developed for piercing and tattoo parlors) can be easily transported to the field. Therefore, decisions regarding which type of autoclave to use must reflect the logistics of a given study. Another option involves purchasing sterile surgical instrument kits or sterilizing instruments in the laboratory in sterile packages that can be opened in the field. This approach requires multiple sets of surgical tools. A fundamental problem with heat, however, is the potential to damage the internal circuits and batteries of telemetry devices that are made primarily of plastics and resins that can melt. Therefore, electronic tags must be sterilized using other methods.

An alternative to heat is the chemical sterilization of surgical instruments and electronic tags. This process can be performed by soaking items in a chemical sterilant such as glutaraldehyde (e.g. Cidex®) or phthalaldehyde (Mulcahy 2003). Chemical sterilants kill all microorganisms by denaturing their proteins and dissolving their lipids (McDonnell and Russell 1999). Other chemicals, such as ethanol, benzalkonium chloride, and chlorhexidine, have been used to disinfect surgical tools, although disinfectants do not truly sterilize (i.e., completely eliminate all microorganisms). Bacterial endospores are most resistant to disinfectants; however, some viruses and bacteria also possess some tolerance. Proper use of chemical sterilants and disinfectants involves submerging surgical instruments in a full concentration bath for at least 10 min (Burger et al. 1994; CCAC 2005). Soaking tools in this manner must be followed by a thorough rinsing with sterile water or saline because these chemicals can be toxic to tissues. For multiple surgeries, several sets of instruments can be rotated among the chemical bath, the rinsing bath, and use in the surgical procedure. Disposal of such chemicals often is regulated, and appropriate personal protective equipment is required during their use (see discussion in Rutala and Weber 2004). Another option for chemical sterilization is the use of gases such as ethylene oxide or hydrogen peroxide vapor. These gases are not recommended for direct use by researcher, but local hospitals may be inclined to include transmitters in their regularly-scheduled gas applications. These sterile transmitters can then be placed into sterilized packages and used when required.

Chemical disinfection is the only option for telemetry transmitters. However, it is necessary to ensure the chemicals to be used do not degrade or destroy the sensors or encapsulating material on the transmitters.

Surgical incisions

Incision placement

Implantation of a transmitter within the abdominal cavity of a juvenile salmon (intracoelomic implantation) requires an initial incision approximately 5–10 mm in length, depending on the size of the transmitter relative to the fish. This incision usually is made on the ventral surface of the fish, anterior to the pelvic girdle. Two locations most often are used for incisions—on the ventral midline along the linea alba (dense fibrous structure composed mostly of collagenous connective tissue) or lateral and parallel to this region. Many researchers make the incision parallel to, and slightly off, the linea alba. However, midline incisions may reduce the amount of tissue damage and are a traditional approach for abdominal surgeries on both humans (Rath et al. 1996) and fish (Murray 2002), and other animals.

Various pros and cons are associated with these two commonly used incision locations. A possible negative consequence of making incisions on the linea alba of fish is the increased likelihood of the post-surgical fish coming into contact with objects, substrate, or debris on the bottom of a body of water, possibly leading to physical damage at the incision site (CCAC 2005). However, this placement has been shown to have the strongest suture-holding power and wound closure strength for abdominal incisions in humans (Tera and Åberg 1977). Fish are similar in anatomy to humans and other vertebrates at the midline of the abdomen where bilaterally symmetrical muscle groups are joined together by the connective tissue of the linea alba. An incision parallel to the midline would avoid the problem of direct contact with substrate, but in humans this location causes damage to muscles and has weaker holding power (Tera and Åberg 1977). Along with muscles not tolerating suturing well, cutting through muscle tissue can increase blood loss (Nygaard and Squatrito 1996) and interfere with important nerve function in comparison to cutting through the relatively avascular linea alba (Burger et al. 2002). Each abdominal muscle fiber in fish is innervated by multiple axons (i.e., polyneuronal) (Hudson 1969), which do not always heal as quickly as connective tissue that has a high prevalence of collagen and fibroblasts (Mutsaers et al. 1997). Although myofibrillar regeneration of muscles can act as fast as fibroblast and collagen reformation of connective tissue, muscle healing often is slower due to necrosis and inflammation (Anderson and Roberts 1975).

Although it has been well documented that the linea alba is the location of choice for abdominal surgeries in humans, research still is lacking as to which incision location is better for juvenile salmonids. Currently, there is widespread use of these two locations in fish telemetry surgeries, but there have been few documented studies comparing the effects of the two incision locations. Wagner and Stevens (2000) found no difference between the two incision locations in terms of inflammation or the behavior of freely swimming rainbow trout (average 306 mm and 338 g). However, fish with midline incisions swam farther than fish with off-midline incisions, indicating that incisions along the linea alba minimize behavioral changes in trout. Further research is needed, however, because the fish in this study had a tag burden of 1%, and it is typical for salmon smolts to have a tag burden between 7 and 10% when implanted with acoustic microtransmitters. It is unknown how this heightened tag burden affects incisions on the linea alba or off-line incision.

A recent study has been conducted comparing healing among incisions on the linea alba, off the midline, and using a muscle sparing technique (following the underlying muscle fibers) in juvenile Chinook salmon (95–121 mm, *Oncorhynchus tshawytscha*) implanted with acoustic microtransmitters (Panther et al. 2010). Results indicated incision closure was better for fish with incisions on the linea alba 28 days post surgery at 12°C and 21 days post surgery at 20°C. Transmitter loss was similar among all incision types during the first 28 days at either temperature. Another implantation study involving juvenile Chinook salmon held at 12 and 17°C corroborated these results, with transmitter loss through an incision in the linea alba at less than 0.5% for either temperature after 28 days (Deters et al. 2010). However, there may be an issue with long-term wound strength of incisions along the linea alba for fish held at colder temperatures. Panther et al. (2010) reported significantly higher transmitter loss by day 98 for fish held at 12°C with incisions on the linea alba. This loss possibly was due to dehiscing of weakly bonded wound edges due to internal transmitter pressure on the incision site. These results suggest the odds of losing a transmitter through an incision on the linea alba may decrease as temperature increases and the rate of healing increases.

Based on the above research, we suggest that incisions on the linea alba are likely to lead to better incision healing during short term (≤28 days) studies. However, more long-term studies are necessary to determine which incision location is optimal at different temperatures and tag burdens. In addition, more research is needed comparing the relatively new use of muscle sparing incisions with incisions off the midline and along the linea alba.

Soft tissue protection

While making the incision and during insertion of the transmitter, it is important to protect the soft tissues and organs inside the body cavity, such as the pyloric cecae and the spleen. In juveniles, perforation of the swim bladder is also an issue, as it occasionally is positioned immediately below the incision site. The viscera can be protected during the initial incision by proceeding slowly with the scalpel to extend a small shallow cut through the tissue layers. Use of good lighting (e.g., fiber optic or LED white light so that heat is not generated and directed toward the fish) makes it possible to discern when the peritoneum has been cut. Curved, blunt forceps can be inserted into the body cavity through this small opening, lifting the body wall gently with the forceps to create space above the viscera while the incision is lengthened to the desired size. Insertion of the transmitter involves passing one end through the incision into the abdominal cavity, then decreasing the angle of insertion while gently pushing the transmitter slightly anteriorly until the entire tag is within the abdominal cavity. In smaller fish, placement of the transmitter immediately below the incision can provide a barrier between the viscera and the needle when suturing. After closure, the transmitter can then be gently repositioned away from the incision by stroking the body of the fish. This repositioning may help prevent excess pressure on the incision site that can lead to pressure necrosis and transmitter expulsion (Lucas 1989; Knights and Lasee 1996). With larger juvenile salmon, the transmitter can be placed anterior to and to one side of the incision. In some projects, fish are implanted with a passive integrated transponder (PIT) tag in addition to the acoustic transmitter. In this case, the PIT tag should be placed anteriorly in the abdominal cavity prior to the insertion of the transmitter. Care should be taken to not accidently drive the PIT tag into any of the organs, especially the liver, when pushing the transmitter into the abdominal cavity.

Wound closure

Suture material

Several types of material are available for closing incisions, including synthetic monofilament and braided sutures (absorbable and nonabsorbable), silk (nonabsorbable), natural gut (absorbable), stainless steel suture and staples (nonabsorbable), and tissue adhesives (nonabsorbable). The most common suture material currently used for surgeries involving fish is synthetic monofilament (Wagner and Cooke 2005), which has been shown to minimize tissue inflammation compared to other suture types in several fish species. These suture comparison studies have included rainbow trout *Oncorhynchus mykiss* (range of 394–618 mm and 700–2,980 g, Kaseloo et al. 1992; mean 306 mm and 338 g, Wagner et al. 2000), tilapia *Oreochromis aureus* (range of 574–1,033 g, Thoreau and Baras 1997), Chinook salmon (range of 95–121 mm, Deters et al. 2010) and koi *Cyprinus carpio* (average 94 g, Hurty et al. 2002).

However, it is possible some species variation may occur with respect to the effects of suture material on wound healing. Recently, absorbable braided suture has been shown to reduce transmitter loss and improve healing in brown trout *Salmo trutta* compared to nonabsorbable monofilament (average weights of the two groups—78 g and 194 g, Jepsen et al. 2008). This result is contrary to those of Deters et al. (2010), who studied seven suture types used to close incisions in juvenile Chinook salmon (range of 95–121 mm, 10–24 g) at two water temperatures (12 and 17°C). The two monofilaments (absorbable and nonabsorbable) generally had higher tag and suture retention and lower inflammation and ulceration than most of the braided suture types through 14 days post-surgery.

When choosing the type of monofilament to use, it is important to consider the temperature of water in the study area because absorbable sutures may lose tensile strength and be lost more quickly at higher temperatures. Walsh et al. (2000) found absorbable suture losses in fish held at high temperatures (range of 22–29°C) began at 7 days post-surgery and were reduced to two-thirds of their original number by 30 days. The absorbable sutures of fish held at lower temperatures (range 12–18°C) were not affected.

Although the use of quickly placed materials such as staples can decrease surgery times, proper placement in fish skin is difficult (Murray 2002; Wagner and Cooke 2005). Additionally, there have been no reports of researchers using staples to close incisions on fish less than 150 mm long, which is typical of juvenile salmon smolts. As well, the wound closure success and healing rates associated with staples have been variable and sometimes negative. For example, Haeseker et al. (1996) reported high mortality rates (40%) in striped bass *Morone saxatilis* (505–847 mm, 2,000–3,000 g average), and Starr et al. (2000)

reported increased transmitter loss in rockfishes (350–580 mm) when staples were initially used instead of synthetic sutures to close incisions. Similarly, the use of cyanoacrylate adhesives for wound closure is not recommended due to the high incidence of tissue necrosis and dehiscence (i.e., opening of the wound). Dehiscence occurs when cutaneous goblet cells produce mucus that removes the adhesive from the incision (Murray 2002).

Suture size

When choosing the suture material to close an incision, it is a generally accepted practice to use the smallest-diameter suture with adequate tensile strength to hold the mending tissue. However, it is suggested that prior to performing a research study, fish be tested with the proposed suture to ensure the material is not so small that it unduly cuts through the skin at the incision site. Using sutures with minimum tissue resistance and maximum strength will ensure effective wound closure and faster wound healing with a minimum mass of foreign material left within the body (Turner and McIlwraith 1989). An important consideration when choosing the smallest effective suture diameter is that thinner sutures have less tensile strength (Dunn and Phillips 2004) and are more easily damaged (Sutton et al. 2004). This consideration translates into selecting a suture size range between 4–0 and 6–0 for smaller fish such as salmonid parr and smolts that weigh less than 200 g (Table 1). All these sizes likely would maintain suture-holding power. However, because the bulk of a 4–0 suture may slow healing and a 6–0 suture may be so thin that it cuts the tissue, it is suggested that 5–0 sutures be used (similar to Deters et al. 2010) when implanting juvenile salmonids, unless negative effects attributed to suture diameter are observed.

Suture needles

The smallest, least-invasive needle should be selected, to easily penetrate the skin and underlying tissues while leaving the smallest possible hole and minimizing tissue cutting (von Fraunhofer and Chu 1997). Although cutting needles typically are used to suture tougher tissues in humans such as skin (von Fraunhofer and Chu 1997; Sherbeeny 2004), tapered needles (Fig. 1) minimize tissue damage and have been used in fish surgeries on parr and smolts (Table 1). Conventional cutting needles have the cutting edge on the side closest to the incision (the concave side of the needle), which provides a location for the tissue to tear if there is any tension on the suture. Reverse-cutting needles are shaped such that the cutting edge is away from the incision (on the convex side of the needle) and a flat edge is nearer the incision (Fig. 1). Therefore, tearing of the skin is reduced because the suture rests against a flat piece of tissue (Dunn and Phillips 2004).

Either reverse-cutting or tapered needles should be used for acoustic transmitter implantation. When choosing the needle type, it is important to take into account that tapered needles dull faster than cutting needles when used on fish with larger scales. However, this dulling effect is not likely relevant when working on juvenile salmonids that have very thin skin and scales. Currently there is no scientific evidence to suggest there is a difference between reverse-cutting needles or tapered needles when suturing incisions on juvenile salmonids.

Using as small a needle as possible is most important, as this will minimize the hole left in the body wall. A needle with a curvature of three-eighths of a circle typically is used for skin closure because it requires only slight movement of the wrist (Dunn and Phillips 2004). Needles with a one-half circle design also can be used but require more hand movement, which can cause faster discomfort and fatigue among surgeons.

Suture pattern

Simple interrupted sutures (Fig. 2) typically are used to close abdominal incisions in fish because they sufficiently appose all tissue layers while minimizing tissue trauma. In addition, they leave less foreign material in the fish compared with vertical mattress patterns (Wagner et al. 2000), although horizontal mattress patterns have been shown to work for some species with thin dermal layers (Lowartz and Beamish 2000). Epithelial cells migrate rapidly across a properly apposed wound margin, with the speed dependent upon water temperature, to protect the fish from infection and the aquatic environment (Ream et al. 2003). It is important to not tie the sutures too tightly because this can cause strangulation of the tissue, leading to ischemic necrosis (Whipple and

Table 1 Suture properties and needle types used to close abdominal incisions in salmonid parr and smolts

Fish species	Length (mm)	Weight (g)	Suture properties		Needle type	Source
			Type	Size		
Atlantic salmon	64–94	6–16	Braided silk	6–0	C-1 tapered	Roussel et al. (2000)
Chinook salmon	95–121	10–24	Absorbable monofilament	5–0	PS-2 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Non-absorbable monofilament	5–0	FS-2 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Non-absorbable braided	5–0	PC-1 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Braided silk	5–0	FS-2 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Absorbable, braided	5–0	FS-2 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Absorbable braided	5–0	P-3 cutting	Deters et al. (2010)
Chinook salmon	95–121	10–24	Absorbable braided	5–0	RB-1 tapered	Deters et al. (2010)
Chinook salmon	95–160	10–45	Absorbable braided	5–0	RB-1 tapered	Adams et al. (1998)
Chinook salmon	122–198	22–99	Absorbable braided	5–0	SH tapered	Anglea et al. (2004)
Atlantic salmon	144	32	Braided silk	4–0	C-13 cutting	Robertson et al. (2003)
Atlantic salmon	146	36	Non-absorbable monofilament	4–0	FS-2 cutting	Lacroix et al. (2004)
Atlantic salmon	161	46	Braided silk	4–0	C-13 cutting	Robertson et al. (2003)
Atlantic salmon	165	39	Braided silk	4–0	N/a	Connors et al. (2002)
Atlantic salmon	187	60	Non-absorbable monofilament	4–0	FS-2 cutting	Voegeli et al. (1998)
Brown trout	N/a	78	Absorbable braided	5–0	FS-2 cutting	Jepsen et al. (2008)
Brown trout	N/a	194	Non-absorbable monofilament	5–0	FS-2 cutting	Jepsen et al. (2008)

N/a = Information not available

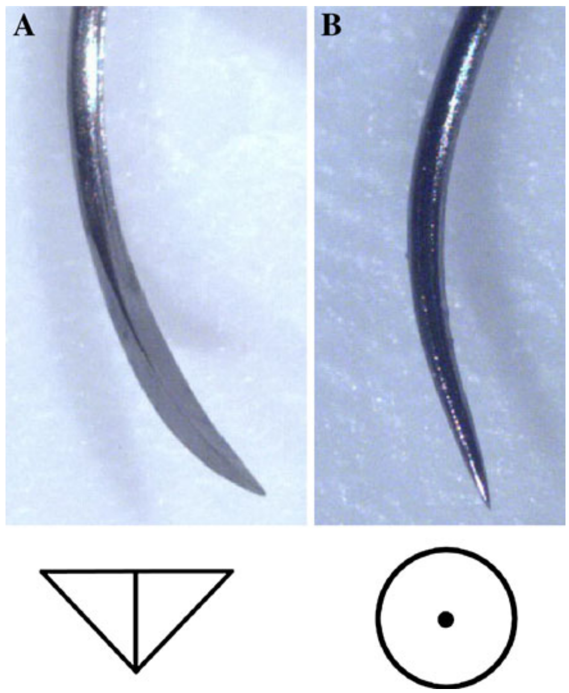

Fig. 1 Two common types of surgical needle used to suture wounds: a reverse cutting needle (**a**) and a tapered needle (**b**)

Elliott 1938) as well as a lack of apposition of the tissue layers. Tight sutures also are a common error in human abdominal wound closure, resulting in lower wound strengths compared to wounds in which the edges are approximated (Nelson and Dennis 1951; Haxton 1965; Sanders et al. 1977).

Suture numbers and spacing

The number of sutures required to close an incision typically depends on its length. At a minimum, the wound margins of the incision should be touching and be in close apposition (i.e., alignment of the tissue layers) to speed epithelial migration across the wound margins (Wagner et al. 1999; Ream et al. 2003). Sutures spaced too far apart do not provide adequate apposition and lead to dehiscence of the wound (Swaim 1980). Sanders et al. (1977) found sutures placed 6–7 mm apart with 5-mm facial bites (i.e., entry point of the needle from the wound margin) created the best apposition and subsequent abdominal wound strength in rats with 6-cm incisions, 4–6 days post-surgery. In general, sutures should be spaced

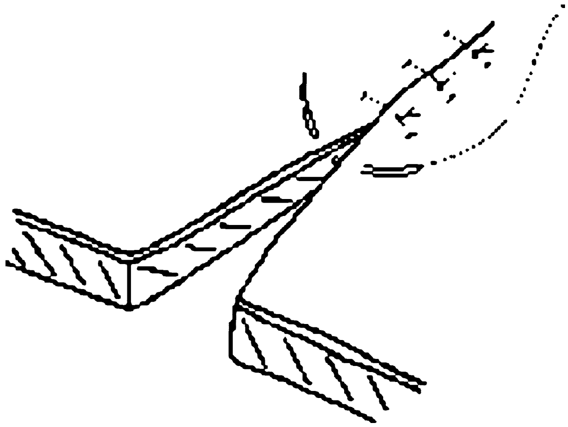

Fig. 2 Simple interrupted suture pattern used to close surgical incisions in fish. Redrawn from Fossom (1997)

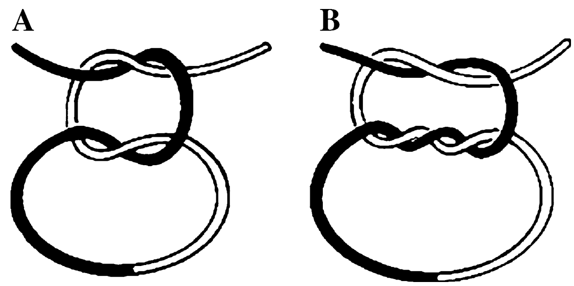

Fig. 3 Two typical knots used to tie sutures. A square knot (**a**) is a secure knot made by a single throw of one of the two ends over the other, then a return and another single throw. A surgeon's knot (**b**) is the same as a square knot except that the first throw is a double. It is more secure than a square and holds its position better for the second throw if wound tissue is pulling apart. It is often reinforced by a third single throw. Redrawn from Toombs and Clarke (2003)

the same distance apart as each suture is wide (Swaim 1980). In addition, the size of the tissue bite is dependent on the thickness of the tissue, with smaller bites for thinner tissue (Turner and McIlwraith 1989). Tissue bites of 2 mm on either side of the incision result in a 4-mm-wide suture. Therefore, in this case the sutures should be spaced 4 mm apart. When implanting acoustic microtransmitters for which the incision may be as small as 5 mm in length, use of one suture may suffice, but there is no research to confirm this. Suture spacing for abdominal incisions in fish to properly appose the wound edges likely will depend on the size of the fish and transmitter, fish species, and thickness of the abdominal wall.

Using more sutures than are required may have a detrimental effect on healing processes because of extra tissue trauma and extra foreign material (i.e., the suture itself) that incites a foreign body response (Uff et al. 1995). The resultant increase in chronic inflammation can delay the coordinated onset of healing and results in reduced wound strength (Edlich et al. 1979). However, it is important to relieve as much strain as possible from the middle of longer incisions because sutures in this location can experience greater tensile forces and tissue inflammation, especially if the transmitter is pressing against the wound (Wagner et al. 2000).

Suture technique

The method used to tie sutures also is important. Excessive tension should be avoided because it can cut the tissue or lead to ischemic necrosis, as previously mentioned. Ideally, sutures are tied loose enough to allow for post-operative edema (Dunn and Phillips 2004) but tight enough to lock in the individual throws (i.e., a single wrap of the suture material around the surgical tool) to produce a secure knot. The two most common knots used in small animal (including fish) surgery are square knots and surgeon's knots (Fig. 3). Some basic knot-tying techniques include alternating the direction of each throw toward and away from the surgeon, laying down each throw parallel to the previous tie (accomplished by alternating the direction in which the two strands are pulled and by applying equal tension to the strands when tightening) and keeping the throws as close to horizontal as possible (Toombs and Clarke 2003).

The number of throws used to secure a knot depends on the amount of pressure on the incision, the suture material being used, and the knot type. The amount of pressure on the wound exerted by the transmitter may be considerable, depending on the transmitter size in relation to the fish size. A surgeon's knot may be preferred when excessive pressure on the wound prevents the first throw of a square knot from remaining tied (Turner and McIlwraith 1989). In cases where a square knot is sufficient, however, Turner and McIlwraith (1989) note the minimum amount of resistance can be attained by using three separate throws, although four or five throws may be required. Dunn and Phillips (2004) suggest that monofilaments in particular may require additional throws to maintain

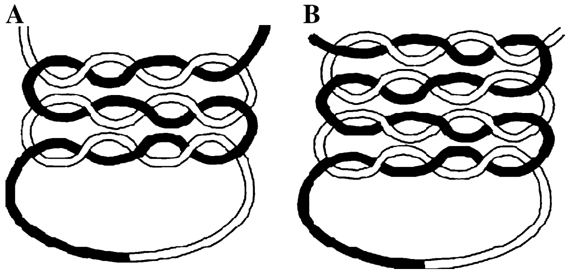

Fig. 4 Two surgeon's knots used by Deters et al. (2010). A surgeon's knot consisting of three double throws was used for all braided sutures (**a**). Monofilament sutures were tied using a surgeon's knot consisting of four double throws (**b**)

knot security. As such, Deters et al. (2010) recently used knots consisting of four double throws (i.e. each throw consists of two wraps of the suture material around the surgical tool) when suturing juvenile Chinook salmon with monofilaments (Fig. 4). Suture retention generally was higher using the monofilaments than using most braided sutures that were tied with three double throws. It is well known that monofilaments have poor knot security; therefore, it is important to be precise when tying these sutures. It is also important to know that excessive numbers of throws do not add to knot security, but do add bulk to the knot that can aggravate tissues and act as a surface for colonizing bacteria and molds.

Summary

The use of sterilized surgical instruments properly selected for a given size of fish will minimize tissue damage, minimize infection rates, and speed the wound healing of fish implanted with transmitters. For the implantation of transmitters into small fish, the optimal surgical methods include making an incision on the linea alba, protecting the viscera (by lifting the skin with forceps while creating the incision), using 5–0 absorbable monofilament suture with a small swaged tapered or reverse-cutting needle. Standardizing the implantation techniques to be used in a study involving particular species and age classes of fish will improve the survivorship and transmitter retention results while allowing for comparisons to be made among studies and across multiple years. Ongoing and future research will provide a basis for refining the surgical methods outlined above.

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