DRAFT

Fish handling guidance for the Upper Colorado Endangered Fish Recovery Program

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Purpose and Need

The primary purpose of this document is to provide consistent and reliable techniques that reduce potential negative effects of capture and handling on fishes studied in the Upper Colorado River Basin (UCRB) while still allowing for data collection to increase our scientific understanding and conservation of these fish. It is intended for researchers of the Upper Colorado River Endangered Fish Recovery Program (Recovery Program). Catching and handling fish is necessary during research of their biology and ecology or during management through activities such as captive breeding, broodstock development or stocking. The initial focus of this document is on the capture, handling, tagging, and transport of wild fish handled during field studies. Field activities can potentially injure fish or induce stressors that indirectly decrease performance (e.g. reduced growth or reproduction) and either of these adverse handling effects can lead to death of the animal. Performing thoughtfully planned and executed procedures on study animals should reduce unnecessary injury and death to the animals and improve the quality of research.

Introduction

The techniques and procedures described in this document are based on the best available knowledge and experience, but are subject to change as new information becomes available. The attached Standard Operating Procedures (SOPs) are for critical and day-to-day tasks regarding procedures performed on fishes in the UCRB. Each procedure should focus primarily on fish health, human safety, and data accuracy. Each SOP should include the purpose of the procedure; a logical, chronological sequence of steps necessary to complete each task; and a material list that identifies all tools and supplies used in the procedure, including the manufacturer and where to purchase. Photos, diagrams, tables, and lists of formulas or conversions can assist in describing the procedure. Human health or safety considerations should be identified. Authors and approval date should be included in order to track changes. Finally, each SOP should have a list of references that support the technique.

SOPs should be peer reviewed by experts to insure conformity to standards and incorporation of best practices. Procedures should be provided to each field crew member and can be used as a consistent training tool to explain exactly how an activity is performed. Procedures can also serve as a handy reference for exacting tasks or to recall formulas, calculations or sources for supplies and should be used as a training aid and readily accessible in the field. Copies should be maintained on the Recovery Program website. Some procedures define a rigid standard technique and must be followed exactly as written to insure proper and uniform data collection. Other procedures provide guidelines that can be modified to fit different situations based on a researcher's best judgement. Any changes to guidelines should be documented.

Specific procedures that might warrant SOPs are presented below followed by a short discussion of general considerations for each procedure. Draft SOPs are attached and other SOPs will be added as needed.

The following draft SOPs are attached to this document.

- Fish transport
- Anesthesia
- Euthanasia
- Floy Tagging
- Passive Integrated Transponder (PIT) (Radio Frequency Identification; RFID) tagging

Additional SOPs needing development are listed below (Those marked with an asterisk are in development):

- Fish Handling *
- Electrofishing standardization *
- PIT Tag scanning-- Handheld scanner *
- PIT Tag scanner setup and data download *
- Fish resuscitation *
- State and Federal Collecting permit contacts
- Fish Preservation
- Data Collection and analysis
- Required data fields in Field Sheets–done by McAda for PIT Tag database
- Species codes- done by McAda for ISMP
- Surgical Implantation of a Radio Transmitter
- PIT Tag scanning with automated fixed-location scanner

General considerations for each procedure

Fish capture—Generally an SOP should be written for each type of sample gear to reduce injuries and deaths to target species or the by-catch. Electrofishing guidelines are provided in federal and state collecting permits which generally follow the recommendations of Snyder (2003). Types of electrofishing units to consider include boat, bank, backpack and seine. Current efforts to standardize the electrofishing boats used by UCRB researchers should guide the development of boat electrofishing SOPs.

Other capture gear include gill, trammel, fyke, and hoop nets, seines, dipnets, angling, fishways, weirs, and piscicides. Many of these gear may not require a specific SOP regarding their use but there is probably a need to specify check time intervals for nets, especially those that entangle fish to reduce injury or death from asphyxiation. In contrast, piscicides which are not currently used in UCRB fish sampling will require intensive permitting and process documentation including MSDS handling safety and detox procedures if they are ever considered.

Fish handling—Fish handling includes all of the activities and procedures performed on a fish from the time it is removed from its natural environment until it is returned to the water. Because different studies and different species often require a variety of different procedures it is most efficient to have a specific SOP for each activity. Specific SOPs are needed for fish handling, holding facilities, length and weight measurements, anesthesia,

tagging, surgery, resuscitation for fish in distress, transport, fish release, euthanasia, preservation, and disposal. Other specific SOPs can be developed as needed.

Workshop participants discussed the importance of carefully handling fish while landing them into the boat and the need to consider dip net material, mesh, and net depth (too shallow and the fish can flop out) in relation to reducing harm to the fish. Participants identified a need to document brands, sizes, and manufacturers of nets and other capture and holding gear. Fish condition and activity should be constantly monitored. Critical issues to reduce or prevent stress while holding a fish include water quality, water temperature, crowding, contaminants, and holding duration. Water quality can be maintained by changing water frequently, adding salts, electrolytes, or fish-cote type products to improve osmotic regulation. Holding tank material (metal or plastic) and color may be important. Metal, especially galvanized tanks is generally discouraged except when there may be a potentially harmful Faraday effect of electrical fields in non-metallic holding tanks during electrofishing. A light tub color will reduce solar heat accumulation. The most important factor in reducing fish stress is supplying adequate oxygen to the holding water. This is best performed by frequent water exchange with fresh river water. The use of oxygen tanks to provide supplemental oxygen was necessary during recent nonnative fish translocation studies, especially for sensitive species like smallmouth bass. Those oxygen tanks were also used for successful recovery of other species in duress; however, oxygen tanks require additional handling, space, and safety considerations. In place of oxygen bottles there is new technology that uses electrolysis to separate oxygen from water molecules to oxygenate holding water. While the process is practical for increasing oxygen in tanks holding a few fish, it appears inadequate for tanks containing large numbers of fish that quickly use the available oxygen. The oxygenation process also releases chlorine and carbon monoxide which must be removed by exchanging water.

Safe handling requires proper body support of the fish and techniques that prevent injury such as using wet hands (bare or gloved) and not holding fish by the eyes, jaws, or gills. Time out of the water should be minimized and all procedures should be done low and over the water tub so that if a fish slips off the measuring board it will fall gently into the tub of water and not on a hard boat surface. Hoisting endangered fish high and extended for a trophy photo is poor technique, especially when multiple photos are taken with each crew member. Photographs for scientific documentation are valuable but should be done with the fish low over the water tub and the fish returned to the water for a short period between shots. When releasing fish back to the wild, researchers should consider whether the fish needs acclimation to the release water if the temperatures are highly different between the holding tub and release water. Avoid throwing or stranding fish at release.

Measurements—Length measurements should be done on a clean, smooth, wet board that has easy to read gradations in mm. Digital scales are good for smaller fish but become less accurate if not level or during windy or unstable conditions. Keeping large fish restrained on a flat bed scale is extremely difficult and likely results in inaccurate weights. Hanging spring scales are practical for larger fish as long as fish are well

restrained in a wet holding net. Weights should be measured in grams. All scales should be calibrated and tared frequently.

Floy and PIT Tagging—Floy tagging requires coordination of tag colors and number sequence among agencies, and crews. Tag location is typically in the dorsal musculature under the dorsal fin. Tag retention should be examined by double-tagging a batch of fish to determine if there is tag loss. Pit tags (RFID tags) require more stringent aseptic conditions when fish are tagged in the abdominal cavity, because internal procedures are invasive procedures similar to surgery. Tags and needles should be as clean and aseptic as possible to prevent disease or bacteria transfer to the fish. Bull-trout researchers recently shifted from an abdominal insertion of PIT tags to an insertion into the dorsal sinus to reduce the potential for harm due to invasive abdominal tagging. The dorsal sinus is the dorsal musculature area below the dorsal fin. They have reported excellent retention. Another less invasive location for fish over 150 mm is the cheek musculature. CSU double-tagged northern pike in 2003 with Floy tags in the standard dorsal location and PIT tags in the cheek musculature and had good retention of PIT tags. Associated with PIT tags is the need to develop good techniques for scanning fish that potentially have a PIT tag. Storing scanned tag data and careful reading and transcribing of PIT tag numbers is critical for successful abundance estimate studies.

Transport—Transport of fish requires consideration of hauling distance, duration, and density of fish but most importantly maintaining high water quality, especially adequate oxygen. Use of salts and oxygen should help maintain water quality. Permits are necessary for moving fish to other waters.

Anesthesia—Anesthesia is necessary for some procedures. Tricaine (MS-222) is the only FDA approved anesthetic for use on fishes and requires a 30 day cleansing period prior to release or human consumption. MS-222 should be buffered to reduce harm to the fish due to its acidity. Dosages should be predetermined for each situation and premeasured. Detoxification procedures are necessary.

Surgery—Surgical procedures need to consider proper sanitation and sterilization, methods to provide oxygenated water and anesthesia to the fish during the procedure, implications of prolonged anesthesia exposure, suture material, surgical tools, and proper surgical technique.

Euthanasia—Euthanasia should be performed on fish that are severely injured or prior to preservation and can include an overdose of the anesthetic MS-222 or in some situations a sharp blow to the head of the fish. Lethal doses should be predetermined and premeasured. Disposal of dead fish should consider permitting, public relations issues and human health. When possible, dead fish could be retained for scientific use or salvaged for human consumption. Preservation procedures should be developed to match intended use of the specimens. Because of their high scientific value, dead endangered fish should have clear disposition policies. There is a need to develop preservation policies for some high profile alien or illicit species and the need for a chain of custody procedure for potential criminal investigations related to illicit stocking.

Fish resuscitation—Fish in extreme distress need immediate attention and resuscitation (or Fish CPR). This is especially important for endangered species. A protocol should describe the signs of distress and the emergency response should include separating the distressed fish from other fish, refreshing water, infusing oxygen, adding salts, and preventing temperature shock. There is also a CPR technique that should be taught to revive a fish by aerating the gills with rapid forward movements through either oxygenated tub water or flowing river water. A small oxygen tank with diffuser stone could be carried by each sample crew to provide emergency oxygen support for endangered fish in distress.

Resources

Web -Need to provide web-based resources here
People/ agencies - Need to provide important contacts for the Program and collecting permits here

References

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Childs, M.R. 2006. Protocol for Collection, Transport, Quarantine, Disease Treatment and Maintenance of the Grand Canyon Population of Humpback Chub: Refuge Establishment and Long-Term Genetics Management. Draft Arizona Game and Fish Report. 7 pp.

Emery L, and Wydoski R. 1987. Marking and tagging of aquatic animals: an indexed bibliography. U.S. Fish and Wildlife Service, Resource Publication 165:57 pages.

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Ward, D. L 2006a. Standardized Methods for Handling Fish in Grand Canyon. Report submitted to Grand Canyon Monitoring and Research Center. Arizona Game and Fish Department, Research Branch, Phoenix. 21pp.

Widmer, A. C. Carveth, and S. Bonar. 2005. Transport and care of small desert fishes. Fisheries Research Report 03-05. Arizona Cooperative Fish and Wildlife Research Unit.

Standard Operating Procedure

Transportation of Fish

Purpose

The purpose of this standard operating procedure (SOP) is to provide a safe and acceptable method of transportation of fish that reduces stress and mortality on the animals being moved.

Discussion

Fish that are transported long distances in holding tanks often experience stressful conditions that may result in decreased fitness or increased mortality (Winkler 1987). These conditions can include low dissolved oxygen, extreme temperature, rapid temperature change, increased ammonia, and physical damage (Kelsch and Shields 1996). Conditions can be improved via mechanical, chemical, or procedural techniques to reduce stress on fish. Fish that are transported using these recommendations will experience reduced stress from handling, temperature and chemical changes. This transportation method is based on recommendations provided in Piper et al. (1982), Winkler (1987), and Carmichael and Tomasso (1988) for safe transport of fish.

Health and Safety Considerations

The hauling tank will be secured to prevent shifting of weight during transport which could cause injury to persons moving fish. Compressed Oxygen bottles must be securely fastened to the vehicle and capped with DOT approved safety cap during transport.(*Need to add Specs*).

Responsibilities

It is the responsibility of each principal investigator to ensure that all persons transporting fish have read and are following the applicable procedures.

Procedures

Fish will be transported in a 750-liter (200-gallon) tank. General guidelines for number of fish per unit volume of water call for 1 kg per 8 liters (one pound per gallon; Piper et al. 1982). Site water will be used to provide consistent water quality to fish. Sodium choride (NaCl) in the form of Solar Salt will be added to at a concentration of 0.5 % (0.005) of water volume to minimize physiological stress from osmotic loss (Piper et al. 1982, Carmichael and Tomasso 1988, Kelsch and Shields 1996). Avoid the use of

iodized and standard livestock salt because those form may contain impurities. Diffused oxygen will be provided through 25-cm airstones from a regulated, compressed oxygen tank. Water temperature will be monitored frequently on long distance (>30 minutes) stocking trips. If an increase in water temperature is detected, ice will be added to the tank. To reduce stress from temperature changes, fish will be tempered with water from receiving water prior to release if temperatures of tank water and receiving water differ by > 5.5° C (10° F).

Procedures

1. Rinse holding tank with site water.
2. Fill holding tank to 80% of capacity, about 600 liters or 160 gallons.
3. For every 380 liters (100 gallons) add and mix 200 grams NaCl.
4. Turn on oxygen from regulator and adjust until airstone releases a gentle, fine stream of bubbles. Do not over oxygenate, evidenced by rapid, large bubbles.
5. Add no more than 75 kg (165 lbs) of fish. This equals no more than 30 typical, 2.5 kg (5 lb) adult fish.
6. Water temperature of receiving water should be less than 10 F of holding tank water. If greater than 10 F, fish should be tempered by mixing water from receiving water into the holding tank. OR just make it SOP to always mix the two waters at some specified rate.
7. Other options: Mix water with agitator or bilge pump.
8. Check equipment and fish condition every 30 minutes (or 1 hr).

Material checklist

Solar Salt from Hardware or livestock supply store

Useful Formulas

1 gallon = 3.785 liters =0.1337 ft^3
1 liter = 0.2642 gallons = 0.001 m^3

Formula for adding salt at 5% volume rate: 2 g per gallon or 0.5 g per L

References

Carmichael, G. J. and J. R. Tomasso. 1988. Survey of fish transportation equipment and techniques. Progressive Fish Culturist 50:155-159.

Kelsch, S. W. and B. Shields. 1996. Care and handling of sampled organisms. Pages 121-145 *in* B. R. Murphy and D. W. Willis, editors. Fisheries techniques, 2nd edition. American Fisheries Society, Bethesda, Maryland.

Piper, R. G., I. B. McElwain, L. E. Orme, J. P. McCraren, L. G. Fowler, and J. R. Leonard. 1982. Fish hatchery management. Department of the Interior. Fish and Wildlife Service. Washington, D.C..

Winkler, P.. 1987. A method to minimize stress during fish transport. The Progressive Fish Culturist 49:154-155.

Other Recommendations

Air tank should be mounted to the truck to prevent movement.

Holding tank should be incrementally marked in 50 (100) gallon increments.

Anesthesia of Fish

Purpose

The purpose of this standard operating procedure (SOP) is to provide a consistent and acceptable method for anesthesia of fish.

Discussion

Research involving fishes benefits from efficient, safe, and conscientious handling of study animals. Handling methods should reduce stresses that invoke physiological and behavioral changes in the study animals (Barton and Iwama 1991). During captivity, stress is reduced by providing optimal environmental conditions that allow captive animals to maintain homeostasis. Stress may increase from nociception, the detection of injurious stimuli by the nervous system, especially when the fish is removed from water, is handled, or is subject to a study procedures. General anesthesia is recommended to reduce nociception of fishes during intrusive procedures. The following method of anesthesia induction follows procedures recommended by Summerfelt and Smith (1990) and is intended to be consistent with recommendations of UFR (2004).

Tricaine (MS-222) is recommended for fish anesthesia because of its wide use, relative safety, and ease of use. It is also the only federally approved drug for anesthetic use with food fishes. The Food and Drug Administration (FDA) has approved the use of two tricaine compounds for fish, Finquel tm and Tricaine-S tm (Bowser 2001).

Human Health and Safety Considerations

Technical tricaine is a white, crystalline powder that is readily soluble in water. When preparing stock solutions, care should be taken to avoid breathing, and contact with any material that may become suspended in the air as a result of handling. Fish that have been euthanized with tricaine are not safe for human consumption for 21 days. Chronic exposure to dilute MS-222 may cause retinal toxicity (Bernstein et al. 1997). Nitrile or waterproof gloves and eye protection should be worn when using MS-222.

Responsibilities

It is the responsibility of each principal investigator to ensure that individuals participating in anesthesia of fish have read and are following applicable procedures and have received field training with live fish.

Procedures

Fish should be sedated and anesthetized with a treatment bath of site water mixed with a stock solution of Tricaine (40 mg/L). Do not use tricaine in galvanized or brass containers unless sealed to prevent dissolution of zinc. Buffer the dilute, acidic Tricaine solution to pH 7.0–7.5 with sodium bicarbonate ($NaHCO_2$). Buffering prevents lowered blood pH and reduces respiratory stress (Summerfelt and Smith 1990). Uptake of the chemical is through the gills during respiration. Efficacy is affected by water temperature, water hardness, and fish number, species, and size (Bowser 2001). Dosage or immersion time may be increased to offset reduced efficacy, but immersion time should be reduced as dose increases. A short induction time (2–5 minutes) is desired to effect a short recovery. Fish that are anesthetized with tricaine will experience progressive stages of anesthesia including: decreased respiration and reactivity, partial loss of equilibrium, total loss of equilibrium, loss of reflex reactivity, and if overdosed, medullar collapse.

In the event of anesthetic overdose causing respiratory arrest, evidenced by lack of opercula movement, gills should be immediately irrigated with fresh, preferably oxygenated, site water until fish recovery. Fish should be transferred post-procedure to a container of fresh site water with a 5–10 mg/L solution of stockman s salt (NaCl) and held 5–10 minutes until orientation and muscular control are regained. Recovery is identified by the fish regaining equilibrium, muscle tone, and reaction to external stimuli. Fish should be released into calm water at site of capture as soon as possible to reduce captivity stress.

For larval fish, a tricaine concentration of about 150 mg/L is recommended for collections (Snyder 1997).

Source of Materials:

Tricaine, suppliers: Finquel [tm]
Argent Chemical Laboratories,
8702 152nd Avenue,
Redmond, Washington
(800) 426-6258
www.Argent-labs.com

Tricaine-S [tm]
Western Chemical, Inc.
1269 Lattimore Rd.
Ferndale, WA 98248
(800) 283-5292 (360) 384-5898

References:

Barton, B. A. and G. K. Iwama. 1991. Physiological changes in fish from stress in aquaculture with emphasis on the response and effects of corticosteroids. Annual Review of Fish Diseases 1:3-26.

Bernstein, P. S., K. B. Digre, and D. J. Creel. 1997. Retinal toxicity associated with occupational exposure to the fish anesthetic MS-222. American Journal of Opthamology 124 (6): 843–844.

Bowser, P.R. 2001. Anesthetic Options for Fish. In: Gleed R.D. and Ludders J.W. (Eds.), Recent Advances in Veterinary Anesthesia and Analgesia: Companion Animals. Ithaca: International Veterinary Information Service (www.ivis.org), 2001; Document No. A1412.0701. http://www.ivis.org/advances/Anesthesia_Gleed/bowser/chapter_frm.asp?LA=1

Snyder, D. E. 1997. Effects of the fish anesthetic tricaine on larval and early juvenile razorback sucker, *Xyrauchen texanus*. Final report of the Colorado State University Larval Fish Laboratory to U.S. National Parks Service, Fort Collins, Colorado.

Summerfelt, R. C. and L. S. Smith. 1990. Anesthesia, surgery, and related techniques. Pages 213-272 in Methods of fish biology, S. B. Schreck and P. B. Moyle, editors. American Fisheries Society, Bethesda, Maryland.

UFR (Use of Fishes in Research) Committee. 2004. Guidelines for the use of fishes in research. American Fisheries Society, Bethesda, Maryland.

Euthanasia of Fish

Purpose

The purpose of this standard operating procedure (SOP) is to provide a consistent and acceptable method for euthanasia of fish.

Discussion

This method is based on procedures recommended by Summerfelt and Smith (1990) for inducing general anesthesia in fish, and is intended to be consistent with the U.S. Department of Health and Human Services (1985) *Guide for the Care and Use of Laboratory Animals*. Fish that are euthanized following this SOP will experience a progression of stages of anesthesia including: light sedation, deep sedation, partial loss of equilibrium, total loss of equilibrium, loss of reflex reactivity, and medullary collapse. During the final stage of anesthesia, opercular (breathing) movements cease and cardiac arrest occurs.

Use of tricaine (also called MS-222 and Finquel) is recommended for this procedure because it is a widely used for fishery research, and is relatively safe and easy to employ.

Health and Safety Considerations

Technical tricaine is a white, crystalline powder that is readily soluble in water. When preparing stock solutions, care should be taken to avoid breathing, and contact with any material that may become suspended in the air as a result of handling. Fish that have been euthanized with tricaine *are not* safe for human consumption.

Responsibilities

It is the responsibility of each principal investigator to ensure that individuals participating in euthanasia of fish have read and are following applicable procedures.

Required Materials

Tricaine, which may be purchased from:

Argent Chemical Laboratories,
8702 152nd Avenue,
Redmond, Washington 98052
Phone: 1-800-426-6258

Procedures

Prepare a stock solution of tricaine in water of sufficient concentration to achieve a final concentration of 200 mg/L tricaine in the vessel containing fish to be euthanized. Anesthetized fish should experience total loss of equilibrium in 0.5 to 2 minutes. Exposure to tricaine should continue for a minimum of 5 minutes after opercular movement ceases. Following euthanasia, fish may be preserved as needed for subsequent studies.

References

Summerfelt, R.C., and L.S. Smith. 1990. Anesthesia, surgery, and related techniques. Pages 213-272 *in* C.B. Schreck, and P.B. Moyle, editors. Methods for Fish Biology. American Fisheries Society, Bethesda, Maryland.

U.S. Department of Health and Human Services. 1985. Guide for the care and use of laboratory animals. NIH publication 86-23. Animal Resources Program, Division of Research Resources, National Institutes of Health, Bethesda, Maryland.

Floy® Tagging fish

Purpose

The purpose of this standard operating procedure (SOP) is to provide a consistent and acceptable method for attaching Floy®,T-bar anchor tags on fish.

Discussion

Tagging of fish for individual identification is an essential technique for the study of fish ecology, behavior, age, growth, mortality rates, abundance, population dynamics, migrations, stock identification, and stocking success (Wydoski and Emory 1983; Guy et al. 1996). Floy tags are small (40-mm long x 2-mm diameter) flexible plastic (polyolefin) visual tags that are attached through the musculature and anchored between bony pterygiophores. The preferred tagging location is below the dorsal fin with the T-bar imbedded between dorsal, fin-ray, pterygiophores (Waldman et al. 1990; Guy et al. 1996). Groups of tags can have unique colors and each tag a unique, alpha-numeric code or additional text for contact information if anglers or other researchers capture a tagged fish. Advantages of Floy tags compared with other external tags are quick and easy attachment, ability to contain considerable information that is easily observed without elaborate equipment, and potential for long-term, mark-recapture studies. Disadvantages of anchor tags result primarily from incorrect application and may include tag loss or potential infection resulting in increased mortality or decreased growth (Guy et al. 1996). The method described is based on procedures recommended by Guy et al (1996) and operating procedures from Floy Manufacturing. The methodology is intended to be consistent with the *Guidelines for the Use of Fishes in Field Research* (UFR 2004).

Human Health and Safety Considerations

Care should be taken with storage and handling of needles used for tag injection.

Responsibilities

It is the responsibility of each principal investigator to ensure that individuals participating in tagging of fish have read and are following applicable procedures and have received field training with live fish.

Procedures

1. Cleanse needles and tags in 90% alcohol.
2. Insert clip of Floy-tags into barrel of tagging gun and disinfect both by stirring in 90% alcohol solution.
3. Anesthetize fish with tricaine when necessary.
4. Restrain fish by holding upright with head and body underwater and raise dorsal surface out of water. Do not restrict operculi movement.
5. Tag location is left side, under dorsal fin, about 10 mm from dorsal midline.
6. Tag should trail to the posterior at an acute angle to the body axis (to reduce swimming drag on fish).
7. Insert needle between scales, into musculature, pushing it through the dorsal pterygiophores past the midline.
8. Do not penetrate the opposite side of the fish.
9. Compress trigger to insert tag.
10. Rotate tag gun 90° with trigger depressed to dislodge and lock tag behind pterygiophores.
11. Release trigger and remove needle from fish.
12. Gently tug on tag to set and test anchor.

Maintenance

1. Replace or sharpen needles regularly.
2. Lubricate moving internal gun parts with "WD-40" type lubricant.
3. Store needles, tags, and tag gun in a clean, dry location.

Required Materials

1. Mark II, "Pistol L" tagging gun.
2. Stainless steel, heavy duty needles for Mark II.
3. Floy T-bar anchor tags (FD-94)
4. Disinfectant: isopropyl alcohol (90%) in small plastic vial or dish.
5. Diamond needle sharpener.

Source of Equipment

Floy Tag and Manufacturing, Inc.
4616 Union Bay Place N.E.
Seattle, WA 98105
Phone: (206) 524-2700
(800) 843-1172
http://www.halcyon.com/floytag/fish/floyfish.htm/

References

Guy, C. S., H. L. Blankenship, and L. A. Nielsen.1996. Tagging and marking. Pages 353-383 Murphy, B. R. and D. W. Willis, editors. Fisheries Techniques, Second edition. American Fisheries Society, Bethesda, Maryland.

UFR (Use of Fishes in Research) Committee. 2004. Guidelines for the use of fishes in research. American Fisheries Society, Bethesda, Maryland.
http://www.fisheries.org/html/Public_Affairs/Sound_Science/Guidelines2004.shtml

Waldman, J. R., D. J. Dunning, and M. T. Mattson. 1990. A morphological explanation for size-dependent anchor tag loss from striped bass. Transactions of the American Fisheries Society 119: 920-923.

Wydoski, R., and L. Emery. 1983. Tagging and marking. Pages 215–238 in L. A. Nielsen and D. L. Johnson, editors. Fisheries techniques. American Fisheries Society, Bethesda, Maryland.

Standard Operating Procedure

PIT Tagging Fish

Purpose

The purpose of this standard operating procedure (SOP) is to provide a consistent and acceptable method for implanting PIT (Passive Integrated Transponder) tags in fish larger than 200-mm total length.

Discussion

Tagging of fish for individual identification is an essential technique for the study of fish ecology, behavior, age, growth, mortality rates, abundance, population dynamics, migrations, stock identification, and stocking success (Wydoski and Emory 1983; Guy et al. 1996). A Passive Integrated Transponder (PIT) tag is a small (11 mm x 2.1 mm) glass enclosed Radio Frequency Identification (RFID) computer chip and antenna. Each tag contains a unique 10-digit alphanumeric code that is read with an external scanner/reader that interrogates the tag with a radio signal. PIT tags are typically inserted with a 12 gauge needle into the musculature or body cavity. The body cavity is usually preferred over potentially edible musculature of fish (Prentice and Park 1984). Advantages of PIT tags compared with other unique numbered tags are easy identification of live fish, small size, high retention rates, minimal trauma, minimal infection, maximum retention rates, and minimal growth, survival, behavior, or physiological effects (Prentice et al. 1990, Burdick and Hammon 1993). Disadvantages are high costs of tags and scanning equipment, and the inability to detect the tag without equipment. The method described is based on procedures recommended by Burdick (1991) and is intended to be consistent with Guidelines for the Use of Fishes in Field Research (UFR 2004).

Human Health and Safety Considerations

Care should be taken with storage and handling of needles used for tag injection.

Responsibilities

It is the responsibility of each principal investigator to ensure that individuals participating in tagging of fish have read and are following applicable procedures and have received field training with live fish.

Procedures

Injection of PIT tag into fish body cavity:

1. Cleanse hypodermic needles and PIT tags in 90% isopropyl alcohol for 5 - 10 minutes.
2. Load PIT tag into barrel of hypodermic needle and disinfect both by stirring in 90% alcohol solution.
3. Interrogate the tag with tag reader and record the identification code.
4. Hold fish at an angle with head underwater and belly and tail out of water. Do not restrict operculi movement. Orient fish with ventral surface up so internal organs gravitate down away from ventral surface.
5. Implant location is 10-15 mm posterior to pelvic girdle and 5 mm lateral to mid-ventral line. Direction of needle is ANTERIOR.
6. Inject PIT tag:
    a. Face the needle with bevel UP to prevent the needle from sliding on the scales
    b. Angle the needle 20–40° with larger fish having a greater angle.
    c. Push the needle carefully but firmly through abdominal musculature into body cavity and push plunger to inject tag into body cavity.
    d. CARE MUST BE TAKEN TO PREVENT PUNCTURE OF INTERNAL ORGANS.
    e. PIT tag final position should be underneath or dorsal to the pelvic girdle.
7. Use Scanner to read, record, and verify the PIT tag code.

Required Materials

1. PIT tag scanner (Model FS2001F-ISO for 134.2 kHz tags or HS5105-L18K for 400 kHz tags) and spare fuses (2A 3AG)
2. PIT tags **134.2 kHz** (preferred; Model TX1400ST) or 400kHz (Model TX1400)
3. Disinfectant: Isopropyl alcohol (90%) in small plastic vial or dish.
4. Tag Injector: 12 gauge needle modified with stainless steel plunger.

Source of Equipment:
PIT tags and Tag Scanner
Biomark, Inc. http://www.biomark.com
West Riverside Drive
Boise, Idaho 83714
Phone: (208) 275-0011
Fax: (208) 275-0031
E-Mail: biomark@biomark.com

12-gauge needle: Veterinarian Supply Store

References

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