

THESIS

IMPROVING ROCK RAMP FISHWAYS FOR SMALL-BODIED GREAT PLAINS FISHES

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ABSTRACT

IMPROVING ROCK RAMP FISHWAYS FOR SMALL-BODIED GREAT PLAINS FISHES

The growing global need to improve the longitudinal connectivity of lotic systems is often met by using fish passage structures (fishways). When designing fishways in the past, biologists and engineers focused primarily on strong swimming species such as salmonids. However, the majority of riverine species in the interior United States are not salmonids and may be excluded by fishways built using salmonid criteria due to lower swimming abilities and/or behavioral differences. I designed and built a 9.1-m long adjustable hydraulic research flume at the Colorado State University Foothills Fisheries Laboratory (FFL) to test fish passage and evaluate the effects of grade (slopes of 2 – 10%, in 2% increments) on the passage success of three Great Plains fish species: Flathead Chub *Platygobio gracilis*, Stonecat *Noturus flavus*, and Arkansas Darter *Etheostoma cragini*. A 6.1-m long rock ramp fishway was installed in the flume and four PIT tag antennas were used to detect full or partial passage success.

In order to test the key assumption that tagging does not affect fish performance, I evaluated the impacts of 8-mm PIT tags on Arkansas Darter and found no significant difference in the survival and swimming abilities of PIT tagged fish versus non-tagged fish. A review of the literature on small-bodied fish PIT tagging suggests that suturing incisions of surgically implanted PIT tags of small-bodied fishes should be avoided to reduce mortality. Prior studies had already demonstrated that Stonecats and Flathead Chub could be tagged without incurring performance losses.

I used the Cormack-Jolly-Seber (CJS) model in Program MARK to determine the probability of full and partial passage success over the fishway based on the PIT tag detection history of each fish at each antenna. Passage success to upstream antennas was highest at shorter distances and at lower slopes for all species. Probability of passage success was highest for Flathead Chub, followed by Stonecat, and then Arkansas Darter. The probabilities of Flathead Chub successfully ascending a 6.1-m rock ramp fishway at slopes of 2, 4, and 6% were 1.0. Probability of Flathead Chub passage success was very high (0.96) for a 4.06-m, 8% slope fishway. Flathead Chub were unable to ascend 4.06-m of a 10% slope fishway. Stonecats had a passage probability of 1.0 for a 6.1-m fishway at 2 and 4% slope, and a passage probability of 0.83 for a 4.06-m, 6% slope fishway. No passage was predicted for 10% slope fishways greater than 4.06-m and 8% fishways greater than 6.1-m. Arkansas Darters never achieved a probability of 1.0 for ascending a 6.1-m fishway. However, their probability of partial passage success was moderate for a 2.03-m, 4% slope fishway with a probability of 0.43, and for a 4.06-m, 2% slope fishway with a probability of 0.54. Passage probabilities for Arkansas Darters were 0.00 for 10% slope 4.06-m, 8% slope 4.06-m, and 6% slope 6.10-m fishways. Based on the results of this study, it is clear that fishway designs should consider the passage requirements of the species with the lowest performance both in terms of fishway slope and fishway length. For example, a rock ramp fishway with a slope of 4% and a length of 2.03 m would be passable by some Arkansas Darters and all of the Stonecats and Flathead Chub in the size ranges tested. The results of this study provide valuable design criteria by identifying fishway slope and length combinations that allow passage of this representative suite of small-bodied Great Plains fishes.

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CHAPTER 1: DESIGN, CONSTRUCTION, AND PRELIMINARY HYDRAULIC EVALUATION OF A MODEL ROCK RAMP FISHWAY

INTRODUCTION

Fish passage structures or “fishways” are used to improve or restore longitudinal connectivity of streams and rivers on a global scale. The design and construction of fishways dates back to at least 17th century France where regulations for their construction were already in place (McLeod and Nemenyi 1941). Early fishways were sometimes blasted out of bedrock to create primitive pool and weir structures to pass Atlantic Salmon *Salmo salar* over waterfalls (Berg 1973). Since then, multiple designs have been developed to accommodate a variety of fish species and project budgets (Katopodis and Williams 2012). These designs, including pool-and-weir, pool-and-weir-with-orifice, vertical slot, and rock ramp fishways, have been installed in waterways worldwide, with varying degrees of fish passage success (Clay 1995).

A drawback to many past and contemporary fishway design efforts is that innovation in design often relies upon the evaluation of completed fishways, a process that requires the post-construction monitoring of passage success of the novel fishway. This iterative design, construction, and monitoring process consumes a considerable amount of time, delaying the speed with which new innovations can be tested and brought into general use. Modern fishways tend to be expensive, with cost estimates ranging widely (\$10,000 to over \$30,000 USD per vertical foot of dam) based on the specific project (Maloney et al. 2000). Overall, this iterative process of build-and-evaluate is expensive, time consuming, and potentially risky when new approaches or innovations are being tested under field conditions.

An alternative to this approach is to use physical models of fishways to rigorously evaluate design features under controlled conditions before building them in the field (Cheong et al. 2006; Bestgen et al. 2010; Ficke et al. 2011; Ficke 2015; Dockery et al. 2017). Refinement of these lab-tested designs comes from further laboratory research into fish physiology, fish behavior, hydraulics, and monitoring of the design's performance under field conditions from both fish and hydraulic points of view.

In response to the need for an experimental apparatus that would allow testing of a full-scale rock ramp fishway, a hydraulic research flume was designed and built to specifically conduct controlled laboratory experiments on fish passage design and function. This chapter describes the design and construction process. Additionally, basic hydraulic data describing operational conditions within the fishway are presented.

METHODS

Flume Design

The development of a modular research flume and fishway was the first phase of a larger project that focused on identifying the ideal slope for rock ramp fishways for successfully passing a variety of small-bodied Great Plains fishes. To complete this project, a variable slope (0 to 10%) flume that could hold a full-scale rock ramp fishway was needed. Additionally, it was deemed desirable to be able to control as many environmental variables as possible, including water temperature and light level, so that water quality conditions would approximate those of the target system (e.g., Great Plains warmwater stream or cold high elevation stream).

This requirement, along with the projected high levels of water use, made it necessary to design a recirculating water system (Figure 1-1).

The recirculating flume system consisted of a modular fiberglass flume box, a metal brace for the flume, a steel superstructure, an apparatus for adjusting the height and slope of the flume, the water delivery system, and a temperature control system. These are described in greater detail below.

Fiberglass flume: The fiberglass flume was made of two 4.55-m long, 1.2-m wide, and 0.6-m high segments that bolted together to form a 9.1-m long flume. The flume was built by OpenChannelFlow Co. and had plastic reinforcing bars inserted every 1.55 m to minimize buckling of the walls. Each flume section included a pair of 0.61 x 0.30-m Plexiglas windows to allow observations.

In addition to the two straight flume segments, pre-fabricated curved sections of fiberglass flume (one with a 45° bend and two with a 90° bend) were acquired from the same vendor. The curved sections could be added individually or together to create a flume with a 45 to 180° bend in the middle, or, if desired, at either the flume entrance or exit.

Flume support brace: The flume was directly supported by a metal brace constructed of 12-gauge zinc coated steel Unistrut (9.1-m long x 1.4-m wide). The brace consisted of two 4.55 x 1.4 m sections that were bolted together, each supporting one section of the fiberglass flume. The brace also incorporated a wood-decked walkway (9.1-m long x 0.6-m wide) with safety railing to allow easy access to the flume. The brace fit the flume snugly and acted as a load arm to which the flume lifting system was connected, allowing users to raise and lower the flume and walkway.

Flume superstructure: The flume was supported by a two-section steel superstructure. Each section consisted of a 4.62 x 1.50 x 3.68 m (L x W x H) frame that held one of the flume segment, brace, and walkway assemblies. The design load of each section was 3402 kg. The superstructure was constructed of 14-gauge zinc coated steel Unistrut bolted together with 13-mm diameter steel bolts with a shear strength of 413,685 kPa each. The frame also included stops for setting the slope of the flume from 0 to 10%. The downstream end of flume brace was bolted to the downstream end of the support structure to act as a fulcrum when the slope was being adjusted.

Flume hoisting system: A pair of 907-kg capacity manual chain hoists (Dayton model 1VW51) suspended from an overhead I-beam were used to raise and lower the flume brace. Each hoist was positioned over the upstream end of one of the 4.55-m long flume segments, and, when operated simultaneously, could be used to raise and lower the flume. The hoists were connected to the flume support brace with 1088-kg capacity nylon cargo straps.

Water delivery system: Water was delivered to the flume by a 15-hp (11.19 kW) pump located in an outdoor sump that pumped water to a head tank. Water would then flow through the flume, discharge into the tailwater collecting tank, and then flow back to the sump.

The polyethylene head tank (1.4-m wide x 0.78-m wide x 0.45-m deep) received water from the pump through a 0.20-m diameter bulkhead fitting in its base. Water flowed into the flume through a 1.02 x 0.26-m wide opening on its side. The head tank was covered with sheet PVC to reduce splashing and a Coroplast splash guard was installed on the opening to help guide water into the flume. A Sensaphone FGD-0222 float switch installed in the head tank connected to a Sensaphone Express II Monitoring System monitored water levels in the head tank. If water levels dropped below the sensor, the alarm was triggered, sending voice alerts to project staff and

also activated a recirculating pump that drew water from the collecting tank to provide any fish in the tailwater pool of the flume with sufficient water to survive.

Water exiting the flume flowed into a 4300-liter welded aluminum collecting tank (3.04-m long x 1.22-m wide x 1.18-m deep). A Coroplast channel funneled water into the collecting tank and reduced splashing. Water in the collecting tank then drained back to the outdoor sump through 0.20-m diameter pipe.

The majority of the water for the flume was stored in an outdoor, underground 22.7 m³ concrete sump (3.04 m x 3.04 m x 3.04 m). The sump was fitted with 0.20-m diameter inlet and outlet pipes, and included a mount for the pump.

A 15-hp Vertiflo model 832 vertical column pump was mounted in the sump to deliver water to the head tank. The pump was controlled by a Teco model N3-415-C Variable Frequency Drive (VFD), giving it the capability to deliver a range of flows to the flume up to a maximum of 0.0823 m³/s (2.92 cfs). To reduce electronic interference emitted from the VFD, a TCI model KDRB2H drive reactor and a TCI model KRF0025ATB electromagnetic interference (EMI) filter were also installed. The VFD, drive reactor, and EMI filter were all mounted inside of a Hoffman model WF10LP steel enclosure, to minimize electromagnetic interference with PIT tag antennas and other laboratory systems. Sacrificial magnesium anodes were attached to the pump to reduce corrosion. The pump was fitted with a shutoff float switch to stop the pump if the water level in the sump was too low.

Temperature control system: The temperature control system was designed to provide the flume with water ranging in temperature from 10°C to over 20°C, depending upon the experimental objectives. Temperature control was provided by counterbalancing the output of a 4,800W heater and a large chiller connected to the aluminum collecting tank. Both heater and chiller had

their own temperature sensors and recirculating pumps in the collecting tank. An additional recirculating pump (13,600 liters per hour) pulled water from the base of the outdoor sump to the collecting tank, allowing the system to regulate temperature even when the 15-hp primary pump was not operating.

Fishway Design

A trapezoidal cross section rock ramp fishway was designed to fit inside of the research flume described above. The fishway was modeled after the one used successfully by Ficke (2015) to measure the effects of roughness element spacing on the passage success of Longnose Sucker *Catostomus catostomus*, Longnose Dace *Rhinichthys cataractae*, and Johnny Darter *Etheostoma nigrum*.

The fishway was constructed with 6-mm thick PVC sheet, supported by an epoxy-coated wood frame that fit inside the 1.2-m wide flume channel. The fishway was trapezoidal in cross-section with 30° side slopes and included a 0.61-m wide center section that rested on 0.064-m high wooden support studs (Figure 1-2). The downstream fishway entrance consisted of a small sloped section of PVC sheet (0.30-m long at an approximately 30° angle) that connected the bottom of the flume to the floor of the fishway to improve entrance conditions for benthic fishes. A removable 3-mm mesh screen at the downstream end of the flume prevented fish from getting entrained in the collecting tank. Slots for flashboards were installed just downstream of the screen to regulate the water depth at the downstream end of the flume.

The smooth profile of the trapezoidal fishway channel was broken up by the use of regularly-spaced roughness elements. The roughness elements in the fishway were identical roughly-hemispherical polyethylene rocks (95 mm in diameter; 55 mm high; Figure 1-3) molded in the shape of a natural cobble particle found in the Poudre River, Fort Collins, CO, USA. The

polyethylene rocks screwed into the PVC floor of the fishway (Figures 1-1, 1-2, and 1-3). Roughness elements were arranged in a chevron pattern using a spacing of one diameter following recommendations in Ficke (2015) (Figure 1-4). This design and spacing allowed passage of all species tested by Ficke (2015). The flume, the PVC sheet used for the rock ramp, and the roughness elements were all gray in color to provide a subdued substrate color to minimize behavioral changes or avoidance behaviors that could be induced by using a bright substrate color such as white (Casterlin and Reynolds 1977; Houtman and Dill 1994; Ryer et al. 2008).

The system was designed to use four PIT tag antennas in the fishway at 2.03-m intervals (fishway entrance and exit, and at two other locations within the fishway) to allow tracking of partial or complete passage success of individual fish, and to gain more information on the rates and timing of movements. The PIT tag antennas were connected to dual mode (full and half duplex) long range readers (Oregon RFID, Portland, OR, USA). The dual-mode antenna and reader system were selected for their greater flexibility in terms of PIT tags that could be used in the fishway. Small-bodied fishes such as Arkansas Darters *Etheostoma cragini* can be safely tagged with 8-mm PIT tags which were only available in full duplex (FDX) at the time of this study, but they, and larger FDX tags are more susceptible to EMI (W. Leach, Oregon RFID, personal communication). However, the read ranges of the larger (12 – 23 mm) half duplex (HDX) tags were less affected by EMI caused by the VFD, so these tags were used preferentially when the subject fish were sufficiently large to accept them. The PIT tag readers were wired in a “master-slave” configuration with the most upstream antenna being the “master” to prevent the readers from interfering with each other.

The initial design used a pass-through configuration for the antennas but the high levels of EMI caused by the VFD prevented PIT tag detection under this orientation. Further experimentation with antenna design and orientation demonstrated that pass-over antennas installed below the fishway floor and oriented with their long axis perpendicular to the direction of flow in the flume had acceptable detection ranges for the tags used in the study (8-mm FDX, 12-mm FDX and HDX, and 23-mm HDX). These antennas were constructed of ten wraps of 20-gauge copper wire inserted into strips of Coroplast® sheet to maintain constant wire spacing, housed in air-tight 51-mm diameter schedule 40 PVC pipe to form rectangular swim-over antennas (1.01-m wide x 0.25-m long).

Hydraulic Measurements

Mean water column velocity (m/s) in the flume was measured with a Marsh McBirney flowmeter and mean water surface elevations were measured with a point gauge at 58 points in 19 cross-sections throughout the fishway at five gradients (2 – 10% in 2% increments; Figures 1-4, 1-5, and 1-6) to gain a general understanding of the fishway hydraulics. Measurements were taken upstream, downstream, and between roughness elements to characterize the hydraulic conditions inside the fishway. Water velocity was measured at approximately 60% depth. Flows to the fishway at each slope treatment (2, 4, 6, 8, and 10%) were set to 0.0074, 0.0096, 0.0229, 0.0316, and 0.0363 m³/s (0.26, 0.34, 0.81, 1.12 and 1.28 cfs) to maintain a nominal average water surface elevation (a surrogate for depth) of approximately 51 mm at each slope treatment, as measured at two transects (1.58 and 5.69 m below the upstream end of the flume). These transects were chosen to characterize depth conditions at upstream and downstream portions of the fishway. Flashboards installed at the downstream end of the flume regulated the downstream holding pool depth to ensure that the fishway entrance was submerged. While a Marsh

McBirney flowmeter gives relatively coarse measurements of velocity and also in only one direction, attempts to generate a higher-resolution velocity field with an Acoustic Doppler Velocimeter (ADV) and a Pitot Tube were unsuccessful. The water was sufficiently shallow that the available ADVs did not function, while the turbulent and aerated nature of the flow caused the Pitot Tube to give unreliable readings. Therefore, the hydraulic measurements presented in the results section should be viewed as approximations of the actual conditions.

RESULTS

The process of designing, building, and testing a recirculating research flume and full-scale rock ramp fishway proved challenging, yet ultimately was successful. The rough hydraulic data collected showed that I could create consistent conditions as the flume slope was changed, and provided conditions that were very similar to those in some existing fishways in Colorado Front Range streams (Figures 1-5, 1-6, 1-7, and 1-8).

Flume Assembly and Operation

The fishway could easily be raised or lowered from 0 – 10% slope and was safely operated under a variety of flows. The two flume pieces could be separated in the middle to perform maintenance or to install the curved sections with relative ease. Technicians conducting velocity measurements could easily access the flume via the walkway. The temperature control system worked effectively between 9 and 25°C with an accuracy of $\pm 0.5^\circ\text{C}$.

While the pump could deliver $0.082 \text{ m}^3/\text{s}$ (2.9 cfs) to the system at full power (60 Hz on the VFD), output for this project was limited to $0.036 \text{ m}^3/\text{s}$ because there was insufficient static head between the collecting tank and the sump to return flows over $0.036 \text{ m}^3/\text{s}$ through the 0.20-

m diameter pipe (Figure 1-9). To resolve this issue, the size of the return line should be increased¹, or a second 0.20-m diameter line added to allow operation of the pump at full capacity when desired. During pilot trials with the pump, it became apparent that it was very important to maintain a constant net positive suction head (NPSH_A) in the sump by regulating the water level because slight changes in static head resulted in changes in pump output. The installation of an inline flow gauge on the pump's discharge pipe would allow future users to adjust the VFD output as necessary to compensate for slight changes in sump volume. The same effect could be achieved by adding a water level control system to the sump, combined with a secondary holding tank.

Fishway Assembly and Operation

The general fishway structure was relatively simple to assemble, but installing the PIT tag antennas proved challenging because of the EMI emitted by the VFD. As described above, a variety of methods and electronic noise suppression devices were tested, none of which had an appreciable impact, save for careful antenna design and orientation. The most efficacious approach was to design an effective pass-over antenna, as described above. Antenna read range for the 8-mm FDX tags was 6.4 cm, while read ranges for the 12- and 23-mm HDX tags were 12.7 cm and 30.5 cm, respectively. These detection ranges were sufficient to sample the entire water column for my study, but if higher flows and deeper water were used, detection probability of 8-mm FDX tags would be reduced. Of equal importance was that these read ranges were short enough to ensure that the fields from the antennas did not overlap, allowing for discrete fish tracking throughout the fishway.

¹ For example, increasing the size of the return line to 0.30 m in diameter more than doubles the cross-sectional area and would theoretically double the capacity.

Preliminary Fishway Hydraulic Measurements

Water surface elevation was relatively consistent between each slope treatment (Figures 1-6 and 1-8), with most differences between slopes being driven by changes in the tailwater effect in the lowest 1 – 2 m of the flume. Otherwise, water surface elevations were largely uniform throughout the fishway for a given slope. On average, observed water surface elevations were slightly greater at higher slope configurations due to fluctuations caused by turbulence amongst the roughness elements and because more flashboards were needed at the downstream end of the flume to maintain consistent fishway entrance conditions (Figure 1-6). A hydraulic jump developed approximately 6.4 m downstream from the top of the flume at slopes of 6 – 10% (Figure 1-5). As expected, mean water velocities were significantly different between 10, 8, 6, and 4% slope treatments, with greater velocities at higher slopes. There was no significant difference in mean water velocity between slopes 4 and 2% (Figure 1-7). Although mean velocities were higher for the higher slopes, the velocity field at each slope was heterogeneous, providing fish with a swathe of velocities to negotiate (Figures 1-5 and 1-7).

DISCUSSION

The design, construction, and operation of the adjustable, modular flume and fishway presented technical challenges, but the finished product proved useful for measuring fish performance in a full-scale rock-ramp fishway. The ability to change the configuration of both the flume and fishway should provide researchers with the flexibility needed to test various fishway innovations under controlled laboratory conditions prior to employing them in the field.

Evaluation of fishway design

The steel strut was a useful material for constructing the flume superstructure and the flume brace. Available in a range of sizes and strengths, it allowed for rapid assembly and reconfiguration of the support structures without requiring welding. Constructing the whole flume, fishway, and support structure apparatus as two separate “halves” made it easier to change configurations within limited laboratory space and would simplify transportation over a single-piece structure.

The chain hoists used to lift the flume performed acceptably for raising and lowering the flume, though careful coordination between two operators (one per hoist) was required for efficient operation and to avoid placing undue stress on the supporting brace and fiberglass flume. Alternatives to the chain hoists, particularly in situations where there was no overhead support structure for mounting the hoists include hydraulic jacks or motor-driven gear systems.

Although the fishway was made of smooth PVC sheet, Baker and Boubée (2006) showed that increasing roughness of an artificial ramp will greatly increase passage success of fishes. Future fishway studies should consider testing a rough substrate as opposed to smooth plastic or metal to evaluate passage success, as fishways in the field are often not smooth and may produce more realistic estimates of fish passage. The current fishway could be easily modified to test different levels of roughness by unbolting the roughness elements, installing a thin, flexible PVC sheet on top of the smooth PVC sheet, and covering the upper sheet with substrate of the desired size (e.g., sand or gravel) (Bestgen et al. 2010). The large roughness elements could then be reinstalled and the effect of substrate texture determined.

As mentioned before, the range of usable flows in the flume, as initially configured, were limited by the rate at which water drained from the collecting tank to the sump. In retrospect, it

is apparent that the 0.20-m diameter return line to the sump is undersized. Options for resolving this limitation include changing the size of the return line, increasing the head difference between the collecting tank and the sump, adding a booster pump to increase the return flow, or adding a second return line between the collecting tank and the sump. Of these options, the latter two are the most feasible. A booster pump would need to include a level control system to regulate its operation, but because one could use the higher pressure delivered by the pump to push water through a smaller-diameter pipe (e.g., 0.05-m diameter), the required modifications to the sump would be simple. Adding a second return line would obviate the need for a booster pump or active level control, but the line would need to be of similar size to the existing one, requiring more extensive modifications of the sump.

Collecting accurate hydraulic measurements in the fishway was challenging due to the shallow depth in the flume and the turbulent and aerated nature of the flow, especially at higher slopes. It may be possible to use the water surface elevations and known floor geometry to use develop a 2-dimensional model of the flume and fishway that could be used to predict flow fields. However, in order to collect more accurate velocity field data, higher flows and the resulting deeper water are needed. Alternately, a high-resolution ADV probe that could operate at depths of less than 50 mm would be ideal, if such a probe were developed.

It is important to point out that while the fishway was operated at shallow depths in this study, these depths are not out of character with those seen in operational fishways, particularly during low flow periods or when a large proportion of the water in a stream or river is being diverted. By evaluating fish passage under low flow conditions, we can provide fisheries biologists with fish passage estimates even if available flows are less than optimal.

Future modifications

The electronic interference emitted from the VFD was not eliminated. Adding a drive reactor, EMI filter, steel enclosure, and steel conduit around the power lines did not prevent interference with the PIT tag antennas in the flume when the VFD and pump were operational. Electrical engineers raised the possibility that the interference was traveling back through the power lines and into the stainless-steel breaker box and being emitted throughout the building. The line reactor and EMI filter were designed to filter out high frequency interference (150 kHz – 30 MHz), however, the PIT tag arrays operate on a low frequency (134.2 kHz), and low frequency interference was likely the issue for my arrays. Most electronic filters tend to eliminate high frequency interference, while filters that eliminate low frequency interference are more expensive. Future efforts to address electronic interference should investigate the source of the interference using a gaussmeter. If the source of the interference happens to be the breaker box, then replacing the stainless-steel enclosures with steel should help reduce EMI because of the latter material's resistance to EMI. Investment in a filter that reduces low frequency interference may also help with this issue, although the abilities of the filter should be thoroughly researched before purchase.

Increasing the number of PIT tag antennas in the fishway would provide greater resolution in fish movements throughout the fishway, provided that the antenna spacing could be optimized to prevent overlap and inter-antenna interference. Under higher flow conditions, the read ranges of smaller PIT tags may not exceed the depth of the water column. Changing the antenna configuration and orientation might address this issue. If such changes are ineffective, then careful measurements of detection ranges and detection probability should be made. Program MARK uses capture histories of organisms to determine survival, or in this case,

estimates of fish passage (White and Burnham 1999). Even if a fish is not detected at an individual PIT tag antenna, but is at a subsequent antenna, passage estimates can still be calculated for that fish. Data analysis software such as Program MARK can provide estimates of fish passage, even when detection probability is not 1.0, therefore it is not always necessary for PIT tag antenna read range to sample the entire water column depending on the objectives of the study.

Conclusions

While construction of a hydraulic research flume was challenging and expensive (approximately \$130,000 USD), the resulting apparatus provided a valuable tool for evaluating instream structures under controlled conditions and at a realistic scale. When combined with a well-designed research or holding facility that can hold or raise a variety of fish species, a research flume and modular fishway make it possible to test fishway designs for a variety of species relatively rapidly (approximately two months to test slopes of 2 - 10% for a single species, with nine replicate trials per slope, and 20-h passage trials).

There are other fish passage research flumes in existence including those at the USGS LSC Conte Anadromous Fish Center (Castro-Santos et al. 1996; Haro et al. 2004) and at the U.S. Fish and Wildlife Service Bozeman Fish Technology Center (Dockery et al. 2017). Each fishway has its own unique capabilities based on the species it was designed to test and on the characteristics of its location. Being located in the Great Plains and southern Rocky Mountain region makes the flume and fishway described here ideal for developing passage guidelines of small- and medium-sized fishes, and coldwater fish found in these areas. Overall, the Colorado State University Foothills Fisheries Laboratory (FFL) flume is a flexible device that can be used

for a variety of fisheries and hydraulic research projects and is a valuable addition to the research community of the intermountain west.

FIGURES

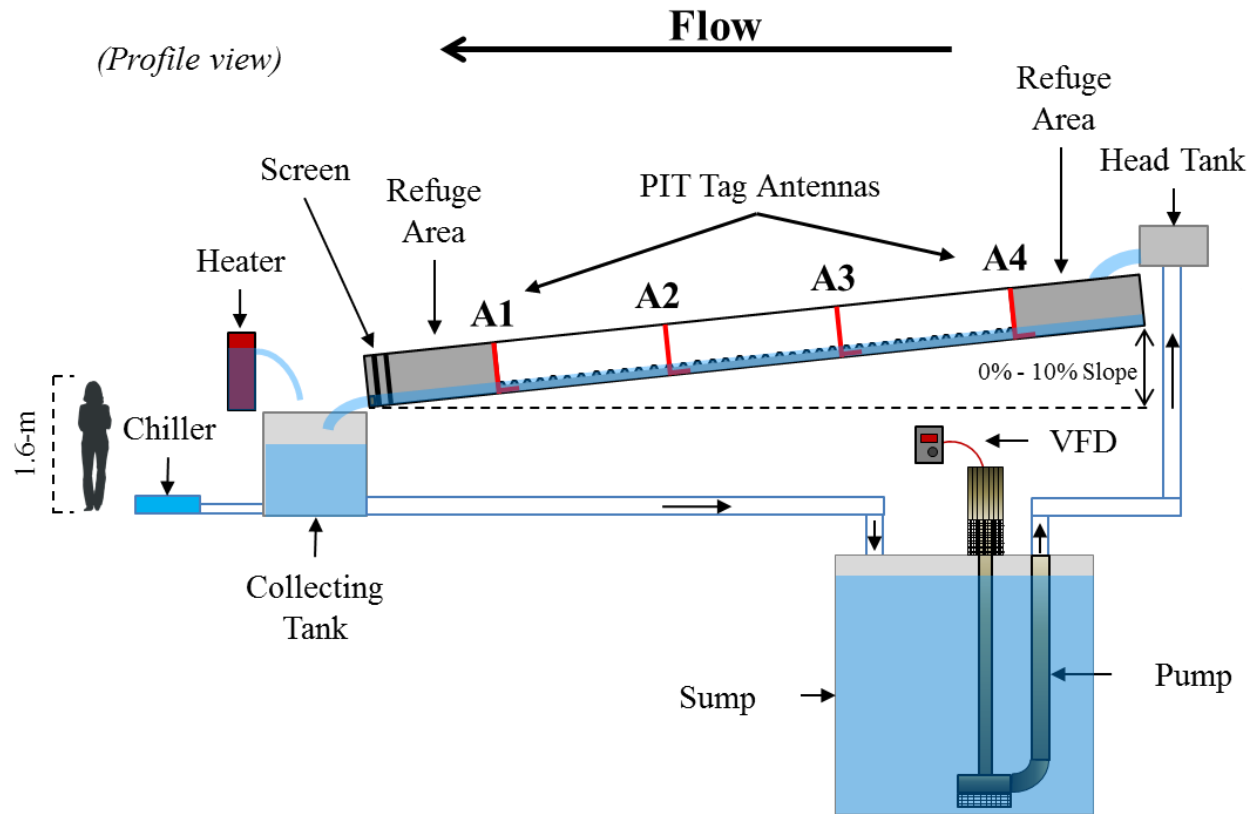


Figure 1-1.— Diagram of the research flume built at the CSU Foothills Fisheries Laboratory. The flume is a closed recirculating system equipped with a 4,800-watt heater and large chiller for temperature control. The flume can be adjusted from 0 – 10% slope by using two overhead chain hoists. A variable frequency drive (VFD) can adjust the output of the 15-hp (11.19 kW) pump to deliver flows of up to 0.082 m³/s (3 cfs) to the flume from a 22,700-l underground sump. Four evenly spaced swim-over PIT tag antennas (A1 – A4) were installed in the flume to monitor fish progress as they navigated the fish passage structure. This diagram is to scale for what a person who is 1.6-m tall would look like standing next to the structure.

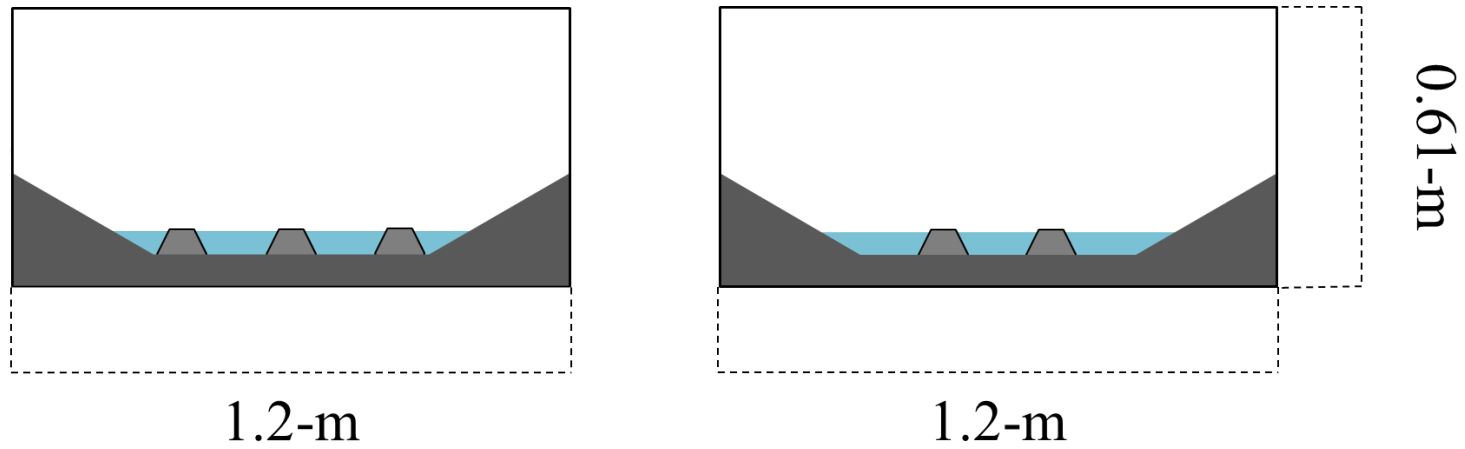


Figure 1-2.— Cross-sectional view of the roughness elements in the trapezoidal rock ramp fishway. A cross-section of the fishway with three roughness elements is shown on the left and a cross-section of the fishway with two roughness elements on the right. Water depth is shown at the target depth of 51 mm throughout the fishway at each slope treatment. The fishway was made of epoxy coated wooden studs covered with PVC sheet. Fishway side slopes were 30° to allow the formation of a wetted margin. Diagrams are to scale.



Figure 1-3.— Photograph showing the shape of the roughness elements used throughout the fishway. Roughness elements were identical molded polyethylene rocks and could be rearranged by bolting the roughness elements to the floor of the PVC fishway. The sloped side of the roughness elements were oriented upstream to mimic the orientation of cobbles in streams.

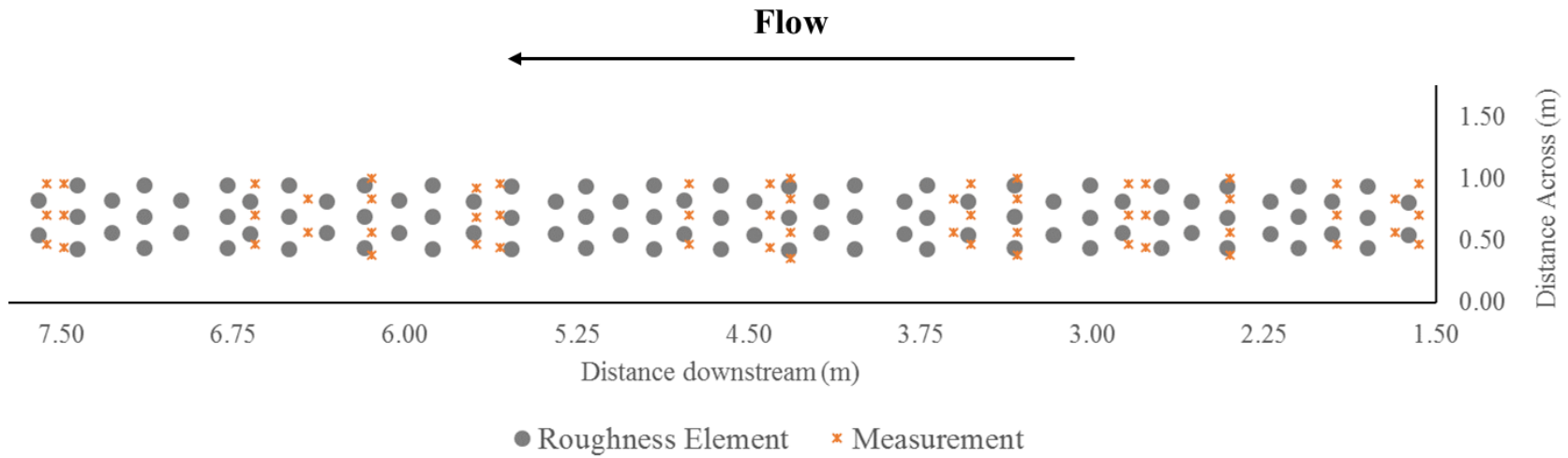


Figure 1-4.— Location of roughness elements in the rock ramp fishway in relation to the location of where water velocity and water surface elevation measurements were taken at each slope treatment (2 – 10% slope). Water velocity measurements were taken at approximately 60% depth. X-axis represents the distance downstream from the top of the flume. This diagram is not to scale to show needed detail.

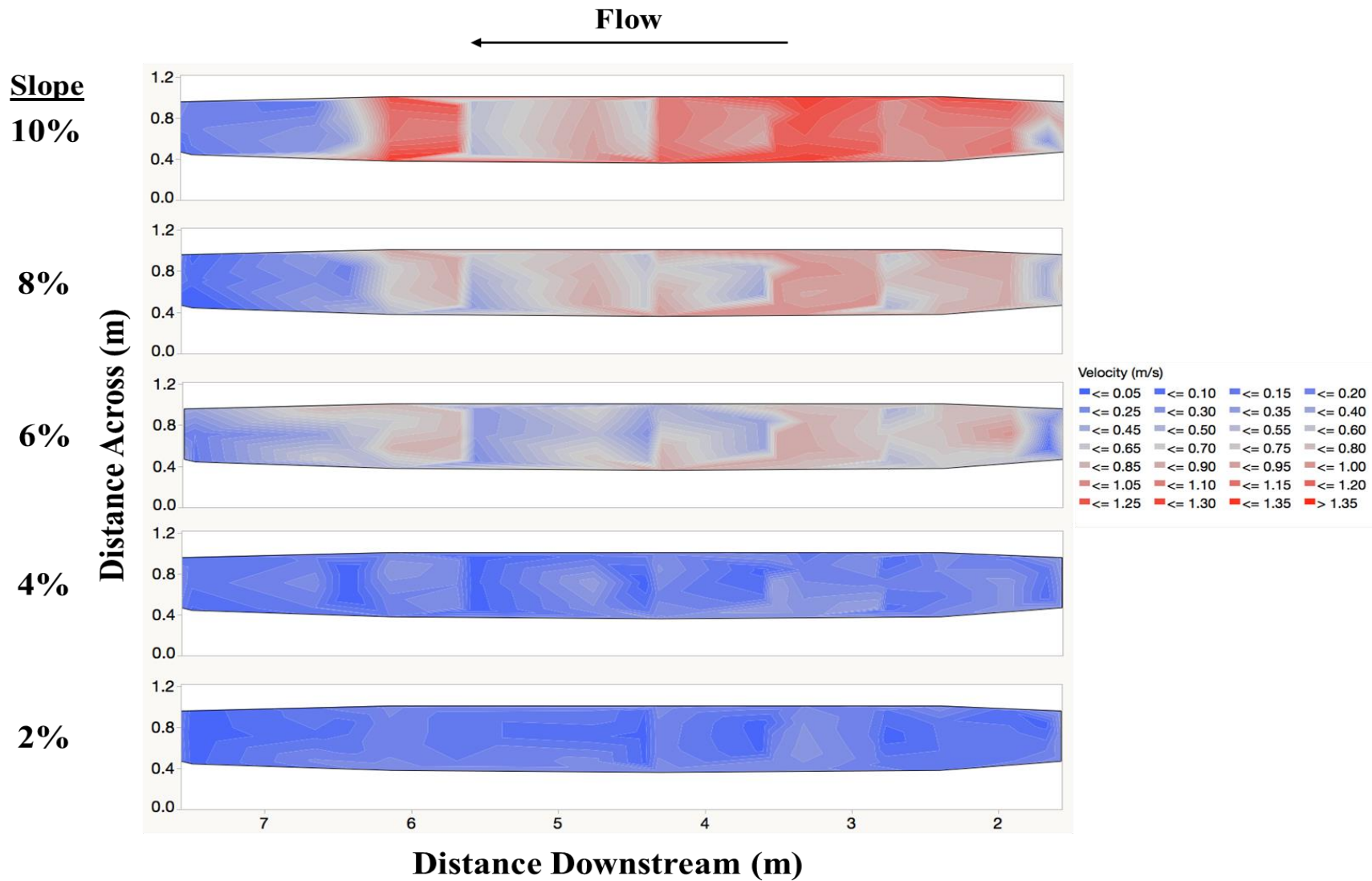


Figure 1-5.— Contour plots of the velocity measurements taken throughout the rock ramp fishway at five slopes. Depth-averaged velocity measurements were taken using a Marsh McBirney flowmeter at 58 points in 19 cross-sections.

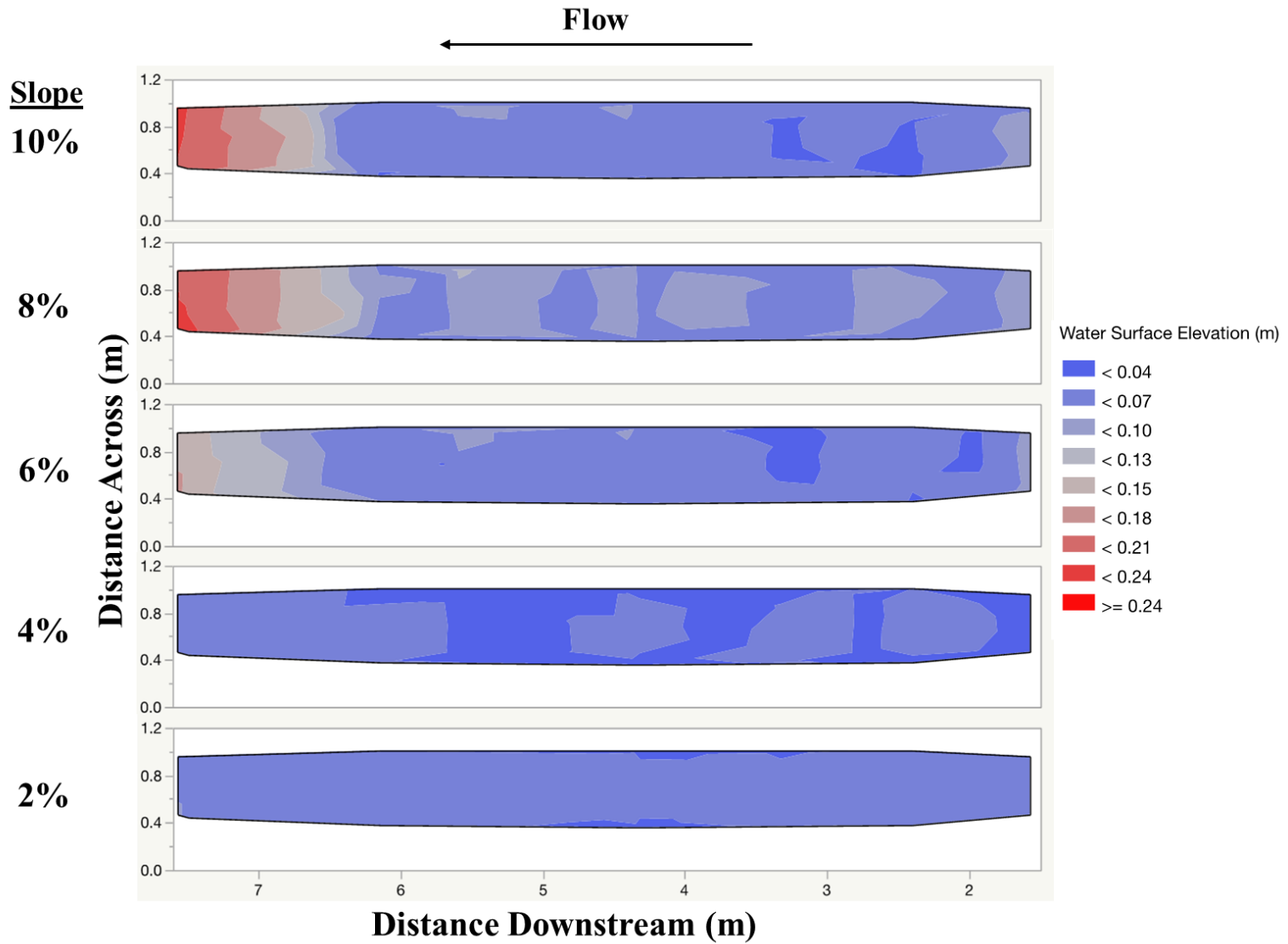


Figure 1-6.— Contour plots of water surface elevations throughout the fishway at five slopes. Water surface elevations were taken using a point gauge at 58 points in 19 cross-sections.

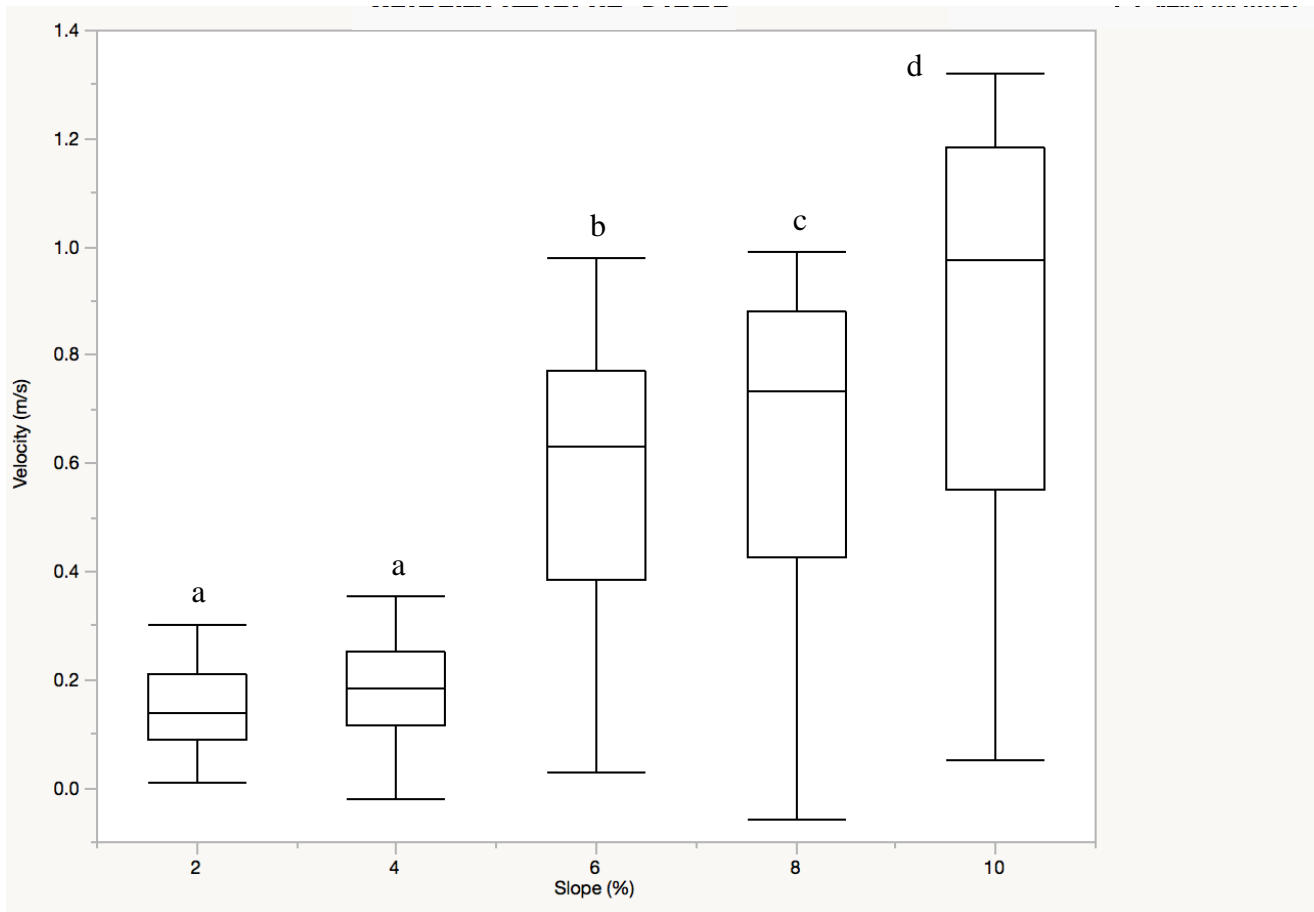


Figure 1-7.— Box and whisker plot of water velocity in the flume (m/s) at five different slopes (%) in a rock ramp fishway. Letters indicate significance between slope treatments.

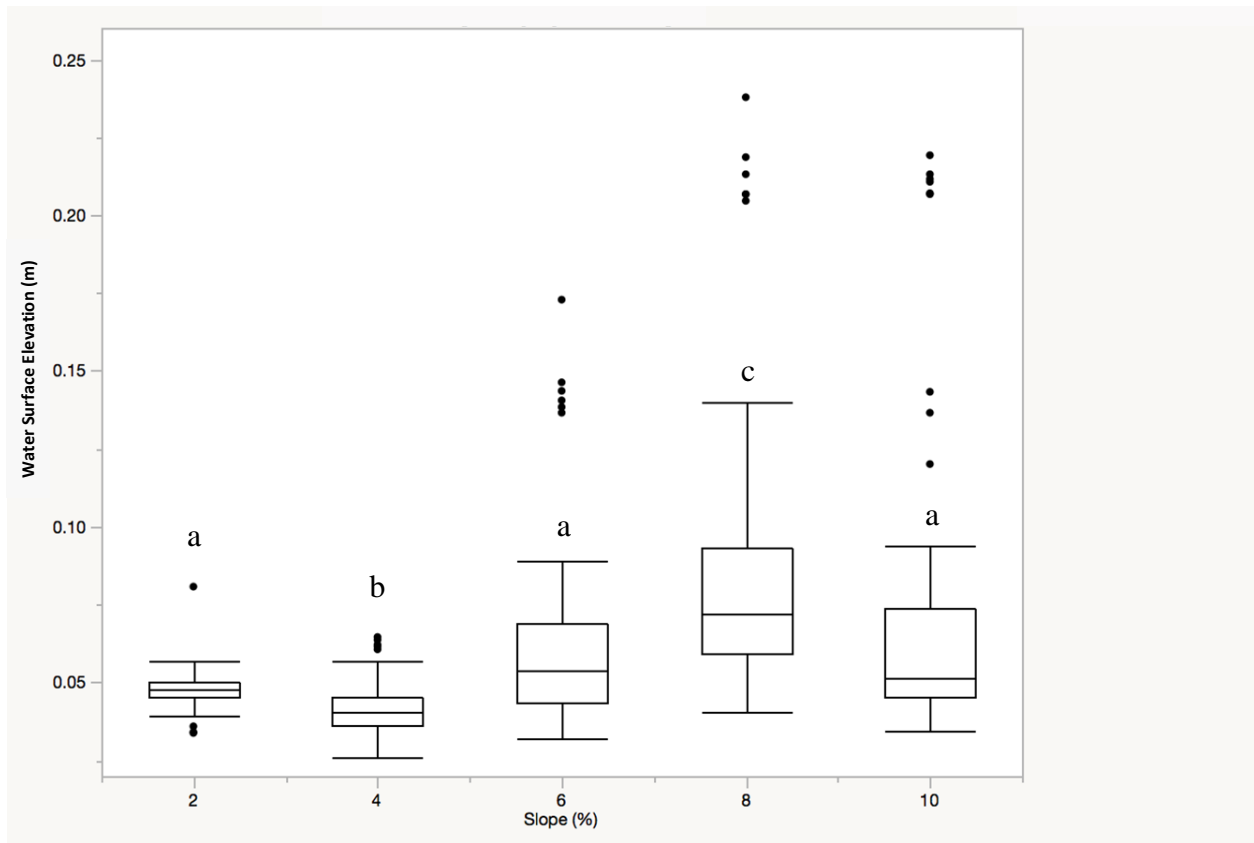


Figure 1-8.— Box and whisker plot of water depth (m) at five different slopes (%) in an artificial rock ramp fishway. Letters indicate significance between slope treatments.

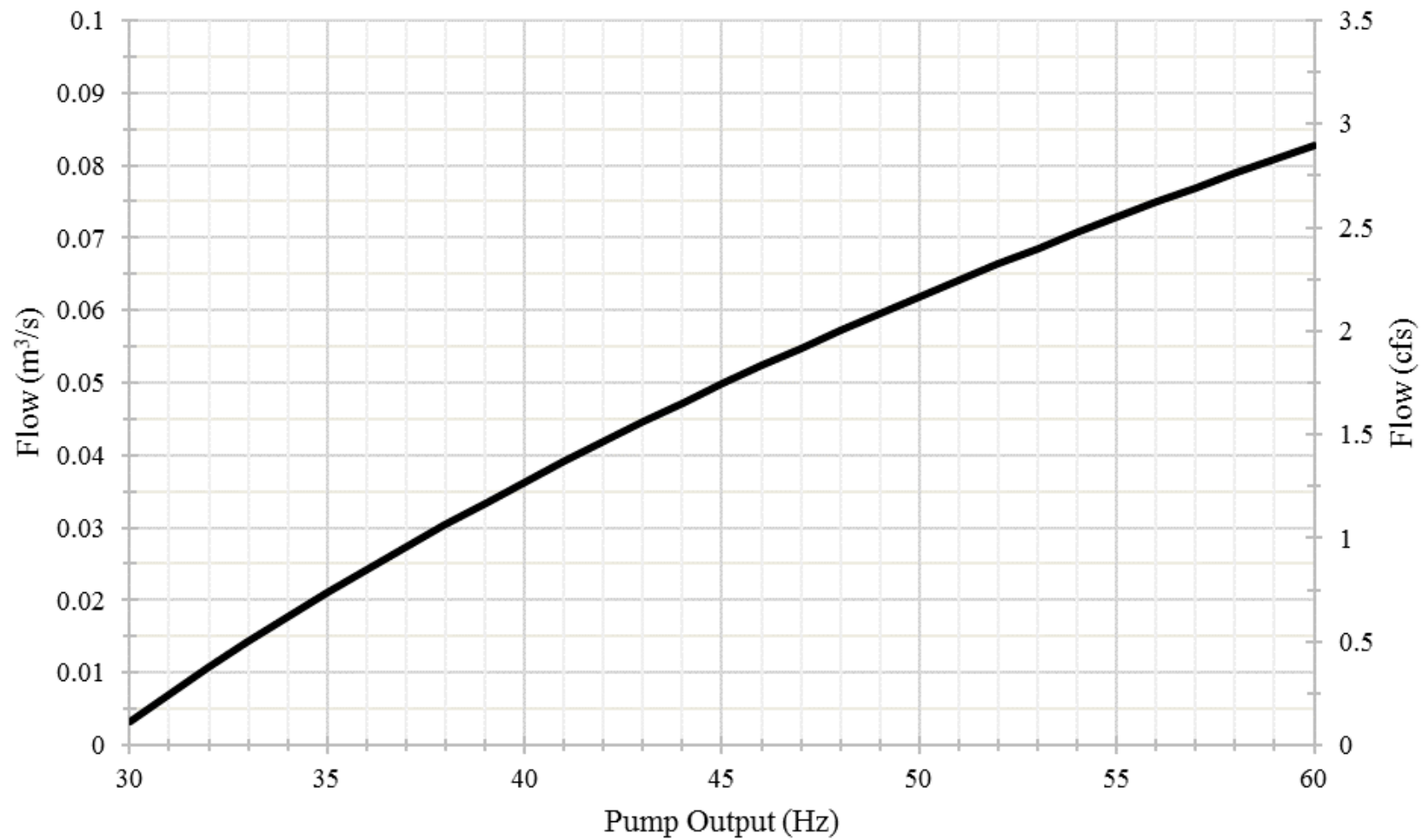


Figure 1-9.— Pump output curve for the 15-hp Vertiflo model 832 vertical column pump used in this study.

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CHAPTER 2: PIT TAG RETENTION, SURVIVAL, AND EFFECTS ON SWIMMING
PERFORMANCE IN A SMALL-BODIED FISH, THE ARKANSAS DARTER *Etheostoma
cragini*

INTRODUCTION

Conservation and management of native fishes requires an understanding of their life history, ecology, and habitat preferences, along with active monitoring of population status (Fausch et al. 2002; Bestgen et al. 2007). Much of this information can be gleaned through well-designed mark-recapture or mark-resight studies. However, such studies are challenging for species whose body size or morphology precludes them from consideration for most mark or tag types, such as darters (Percidae: *Etheostomatinae*). This subfamily of fishes is disproportionately imperiled compared to most other groups of North American fishes (Helfman 2007). Of the 203 recognized species of darters, 54 (26%) are designated as critically Endangered, Endangered, or Vulnerable to extinction (IUCN 2018). One species, the Maryland Darter *Etheostoma sellare*, was declared extinct in 1996. Many darters have traits that increase their vulnerability, including small adult size (< 200 mm total length), small geographic range, preference for headwater habitats, and benthic breeding and feeding habits (Helfman 2007). Because the group is of growing conservation concern, development of effective monitoring methods is crucial.

Passive integrated transponder (PIT) tags are commonly used in fisheries biology to individually identify and discretely monitor the movement of fishes. When coupled with fixed antennas or mobile antenna arrays (Fetherman et al. 2014; Ficke 2015), fisheries biologists can identify individual fish and collect spatiotemporal and movement information. Large fishes (>

200 mm) can be given an external PIT tag (Castro-Santos et al. 1996; Haro et al. 2004), or tagged internally, commonly in the intraperitoneal cavity or in dorsal musculature (Lee et al. 2009). However, smaller fishes are generally tagged intraperitoneally to minimize negative effects on swimming ability caused by external tags or tags in the axial swimming muscles.

There are currently two published studies that describe attempts to PIT tag Darters. Baxter (2015) tagged Kentucky Arrow Darters *E. spilotum* using 8.4 x 1.4 mm PIT tags in a field setting. The Kentucky Arrow Darter is a relatively large species of Darter with adults ranging from 60 – 125 mm TL. Baxter (2015) did not report fish survival or tag retention rates due to the nature of the study. Musselman (2007) PIT tagged limited numbers of Orangethroat Darter *E. spectabile* (n = 9) and Greenside Darter *E. blennioides* (n = 3) using 12-mm PIT tags and reported survival exceeding 80% 60 days post-tagging. Survival of Orangethroat Darters drastically declined after 60 days to 56%, which was attributed to a poor diet.

Methodology used to insert tags into fish can drastically affect survival. Baxter (2015) and Musselman (2007) both used hand held tagging guns to inject tags into the body cavity of the fish, a technique commonly used for larger species. Archdeacon et al. (2009) reported that 64 – 67 mm TL Rio Grande Silvery Minnow *Hybognathus amarus* had significantly higher survival if 12-mm PIT tags were surgically implanted (>87%) rather than injected (50%). Similarly, Baras et al. (1999) reported higher survival (71 -100%) in small (1.9 – 13.7 g) Nile Tilapia *Oreochromis niloticus* with surgically implanted tags versus those with injected tags (10 – 50%). Moreover, Ficke (2015) noted that Iowa Darters (*E. exile*) and Johnny Darters (*E. nigrum*) have a relatively small peritoneal cavity; my pilot dissections of preserved Johnny Darters, Iowa Darters, and Arkansas Darters confirmed that this holds true for these three darter species. Thus, a surgical approach to tagging is likely warranted for this group of fishes.

Determining whether PIT tags impact darter health and behavior is an important step in their adoption as a monitoring method—key assumptions of any tagging effort are that the tags do not affect the survival or behavior of the tagged organisms (Guy et al. 1996). I used this study to develop a surgical technique to successfully PIT tag Arkansas Darters and evaluated tag retention, fish survival post-tagging, and the effects of PIT tags on swimming performance. Additionally, I evaluated survival and tag retention for tagged fish whose incisions were sutured following tag insertion when compared to fish that were not sutured.

METHODS

Source of Fish and Fish Care

Hatchery-reared Arkansas Darters (mean \pm SE TL: 51 ± 3 mm; mean wet weight: 1.40 ± 0.28 g) from the Colorado Parks and Wildlife Native Aquatic Species Restoration Facility (Alamosa, CO) were held in a 340-L round polyethylene tank receiving 5 - 10 L/min of air-saturated water. A spray bar produced a gentle current of 0.05 - 0.10 m/s along the periphery of the tank. After one week of acclimation to laboratory conditions, the water temperature was increased to $20 \pm 0.5^\circ\text{C}$ at $1^\circ\text{C}/\text{d}$. Darters were held for \geq two weeks at this temperature to allow sufficient time for physiological acclimation (Lyytikäinen et al. 1997; MacNutt et al. 2004). The laboratory was kept under a natural photoperiod for Fort Collins, Colorado, USA (40.581°N , 105.138°W). Cover was provided in the form of PVC pipe, PVC sheets, and artificial aquatic plants. Fish were fed daily satiation rations of thawed bloodworms.

Individual fish were taken from the holding tank and placed in a 0.9-L tank for 24 h prior to the first measurement of swimming performance and treatment application. Fish were

randomly assigned to one of three treatments used in the study: control (handled, but no surgery or tag); sham (surgery and suture without a PIT tag), and; PIT tagged (8-mm PIT tag surgically implanted into the fish's body cavity and sutured closed) with a sample size of 15 fish per treatment.

Swimming Performance Measurement

The constant acceleration test (CAT) swimming methodology was used to measure the maximum swimming velocities (V_{\max}) of Arkansas Darters (Leavy and Bonner 2009). The constant acceleration approach was used because it requires fish to use a full range of swimming gaits and should indicate whether one particular gait was more susceptible to the effects of PIT tag insertion than the others. Secondly, the high relative velocities reached in a CAT trial forces fish to use their fastest swimming gait, similar to what they would use to escape predators or negotiate a fishway.

Individual fish V_{\max} was measured at three time points to evaluate the short-term effects of PIT tagging on fish swimming ability.

T₀: Immediately prior to treatment application (control, sham surgery, surgical implantation of tag) to determine baseline swimming ability;

T₁: One day following the surgical treatment, and;

T₇₋₈: Seven to eight days after the treatment.

Fish were swum in a Loligo Model 32 swim flume (32-L volume, 55 cm × 14 cm × 14 cm test section; velocity range of 3-110 cm·s⁻¹; Loligo Systems, Denmark). Fish were given 1 h to become familiar with the flume while an 11-cm·s⁻¹ current was provided for rheotaxis. At the beginning of each constant acceleration test, water velocity was increased from the starting velocity by 5 cm·s⁻¹ every 5 s until exhaustion, defined as partial or full-body impingement for

more than 5 s on the rear screen of the swimming chamber. The velocity at exhaustion was defined as the maximum exposure velocity (V_{\max}), and was recorded in $\text{cm}\cdot\text{s}^{-1}$ and converted into body lengths per second ($\text{BL}\cdot\text{s}^{-1}$). If fish were found “cheating”, defined as resting on the rear screen of the flume or resting in a low velocity area of the flume, the current was momentarily reversed to encourage swimming behavior. Non-performing fish, or fish that refused to swim in one of the trials were removed from the study (Table 2-1).

Tagging Procedure

After the initial swimming trial, all darters were anesthetized using $40 \text{ mg}\cdot\text{L}^{-1}$ of tricaine methanesulfonate, weighed (g), and measured (TL, in mm). Control fish were placed in a recovery tank for approximately five minutes until equilibrium was regained. The recovery tank was supplied with air-saturated fresh water. After regaining equilibrium, fish were then returned to their individual holding tank and Kordon® Pond Fish Protector™ was added at a concentration of $0.4 \text{ mL}\cdot\text{L}^{-1}$ as a prophylactic (Swanson et al. 1996). Fish in the sham treatment were weighed and measured, and then a #12 scalpel blade was used to make a 2-mm long ventral incision slightly offset from the ventral mid-line approximately three-fourths the length of the body cavity, just anterior of the vent (Figure 2-1). Incisions were offset from the ventral mid-line to reduce the likelihood of the incision becoming irritated while the darters rested on the tank substrate. The incision was closed using Braunamid DS24 polyamide pseudo monofilament suture attached to a 24-mm curved suture needle. Once fish recovered from the anesthesia, they were placed in their individual AHAB tank with the same post-handling treatment as the control fish. Darters in the PIT tag treatment underwent the same procedures as the fish in the sham surgery treatment, with the additional step of having an $8.0 \times 1.4\text{-mm}$ PIT tag (FDX-B, 134.2

kHz ISO, 0.027 g; Oregon RFID, Portland, OR, USA) inserted anteriorly into the incision prior to suturing.

Fish were swum at T_1 and T_{7-8} using the same procedures as before. Survival of all groups and tag retention of fish in the tagged treatment were monitored daily for up to 199 days. Repeated-measures ANOVAs were used to determine if there were significant differences in V_{\max} in body lengths per second (BL/s) between treatments (Control, Sham, or Tag) and swimming trial (Pre-treatment, 1-day post-tagging, and 7-8 days post-tagging).

Suture vs. non-suture comparison

During the study, it became apparent that suturing posed three challenges: 1) it was a difficult technique to use on such small fish; 2) the probability of causing harm was high if the body wall was improperly pierced with the suture needle, and; 3) the technique added an additional 30 - 45 seconds to each tagging procedure. To address these challenges, I conducted a follow-up study comparing sutured vs. un-sutured Arkansas darters to determine if suturing improved tag retention and survival.

Randomly selected individual darters were placed in 9-L holding tanks and fasted for 24 h prior to surgery. Fish were randomly assigned to either the sutured or un-sutured group. A PIT tag was inserted into the peritoneal cavity using the technique described above, with fish receiving a suture ($n = 16$) or no suture ($n = 29$). Fish were inspected daily for the presence of a suture and closure of the incision. Fish remained in the individual tanks for 21 days post-tagging and were then moved to a communal 340-L tank identical to the holding tank described above. Tag retention and survival continued to be monitored daily in the communal holding tank for up to 221 additional days.

RESULTS

Differences in total lengths and wet weights of Arkansas Darters used in the various treatments were not statistically significant (Tables 2-1 & 2-2). Neither the full PIT tag insertion procedure nor the sham surgery affected Arkansas Darter swimming performance, nor were there differences between pre-surgery and either post-surgery swimming performance measurements (RMANOVA; $p > 0.05$; Table 2-2). PIT tag retention and survival rates were 100% in the swimming portion of the study. Two tagged fish that were removed from the study because of non-performance died 7 and 12 days post-tagging. These were the only mortalities during the study, and if included in the results, survival of PIT tagged fish would drop to 88%. The mortalities were thought to have resulted from injuries incurred during suturing.

Tag retention and survival were 100% for both sutured and un-sutured fish in the second study (Table 2-3). The sutures were expelled 7 – 14 days post-surgery for sutured fish; incisions of non-sutured fish closed 3 – 5 days after surgery. The process of weighing, measuring, and surgically inserting a PIT tag took 40 – 60 seconds depending on the experience of the tagger; suturing the incision added an additional 30 – 45 seconds of handling time.

DISCUSSION

My results show that it is possible to tag Arkansas Darters ≥ 48 mm TL with 8-mm PIT tags without significantly affecting their swimming ability or survival. Despite the relatively large size of the PIT tags (up to 16% of the fish's TL), the surgical approach allowed me to successfully tag fish. Importantly, two of the key assumptions of any marking or tagging

operation were not violated—there was no significant difference in survival between tagged and untagged individuals and the tags did not affect the physical performance of the fish. Indeed, the continued growth of some of the Arkansas Darters (up to 12 mm during the 199-d post-tagging period) and the sexual maturation of male and female darters were further evidence that the tags had little impact on the fish. Before starting this study, the author measured the body cavity length of a four Darters and observed the body cavity of 52-mm Arkansas Darters was only 10 mm. This suggests that 12-mm PIT tags should not be used on small darter species to reduce the possibility of injuring the organs or causing mortality.

Review of small-bodied fish PIT tagging

Surgical technique and tag size can affect the survival of multiple species of small-bodied fishes post-surgery. In four PIT tagging studies using small-bodied fish, surgical implantation was superior in terms of survival when compared to injection (Archdeacon et al. 2009; Baras et al. 1999; Baras et al. 2000; Kano et al. 2013; Table 2-4). Tag size relative to fish body size also impacts survival. Acolas et al. (2007) tagged Brown Trout *Salmo trutta* of varying sizes with 11.5 x 2.1 mm PIT tags and found that larger fish had higher survival. Baras et al. (1999) tagged Nile Tilapia using a variety of surgical techniques and at different sizes with 10.3 x 2.1 mm PIT tags and found that survival was almost always higher for larger fish for any given surgical procedure (surgery, surgery and suture, and injection) (Table 2-4). Ficke et al. (2012) also noted that survival decreased (68.4% to 62.5%) for Creek Chub *Semotilus atromaculatus* of similar size to one another, when a larger tag (12-mm versus 23-mm) was used (Table 2-4). Because larger fish have a larger peritoneal cavity, it is intuitive that they may be able to accommodate a larger tag, thus improving survival. Future attempts to PIT tag small-bodied fish should consider peritoneal cavity size and select a size-appropriate PIT tag before proceeding, remembering

functional limitation differences of smaller tags with regards to tag type (full duplex vs. half duplex) and tag detection range.

Additionally, suturing small-bodied fish is not recommended due to the increased likelihood of accidental injury during the suturing process. Baras et al. (1999) noted lower survival rates 49 days after surgery on two size (weight) classes of Nile Tilapia (< 3 g and 4 – 7 g) for sutured fish (75%; 91.7%) compared to non-sutured fish (83.3%; 100%) showing the potential for mortality when suturing small-bodied fish. The results from my sutured versus non-sutured comparison indicate that tag retention and survival were comparable to that of sutured fish, therefore it does not appear necessary to suture small darters to enhance tag retention or fish survival.

The results of this study show that Arkansas Darters ≥ 48 mm TL can be successfully tagged with 8-mm PIT tags without impairing their swimming ability and long-term survival. This may improve monitoring and conservation efforts for this imperiled group of fishes by allowing fisheries biologists to monitor their movements with passive or mobile antenna arrays placed in stream networks, or allow rapid broodstock identification in conservation hatcheries. I successfully applied the same tagging technique to other small-bodied fishes including Flathead Chub *Platygobio gracilis* and Stonecat *Noturus flavus* with success, supporting the view that the technique is applicable to other taxa. This study, and other literature on small-bodied fish PIT tagging studies have shown that using a surgical tagging approach as opposed to using hypodermic injectors should improve chances of survival. Further, it is important to match tag size to peritoneal cavity capacity, and suturing tag incisions should be avoided because it does not gain much benefit in terms of retention, and, in some cases may reduce survival.

TABLES

Table 2-1.— Effects of tagging procedure on the survival, tag retention rate, and baseline swimming ability (T_0), and 1- and 7-day post-tagging swimming performance (T_1 and T_{7-8}) of Arkansas Darters. Tagged fish had an 8.0×1.4 -mm PIT tag surgically implanted in their peritoneal cavity. Survival and tag retention were monitored for 199 days post treatment application. Values are means with SD in parentheses. Incisions of PIT tagged fish were sutured for this portion of the study. There were no statistically significant differences within or between treatments (RMANOVA; $p > 0.05$).

Treatment	<i>n</i>	TL (mm)	Wt (g)	199-d Survival Rate (%)	199-d Tag Retention Rate (%)	Maximum Swimming Velocity (BL/s)			Non-performers
						T_0 (cm/s)	T_1 (cm/s)	T_{7-8} (cm/s)	
Control	15	51 (3)	1.40 (0.26)	100	-	13.1 (1.8)	12.2 (1.4)	12.1 (1.4)	1
Sham	15	52 (3)	1.41 (0.28)	100	-	11.9 (1.8)	11.6 (1.1)	11.6 (0.9)	4
Tag	15	52 (3)	1.41 (0.26)	100	100	12.8 (1.6)	12.1 (1.2)	12.3 (2.1)	1

Table 2-2.— Results of repeated measures ANOVA comparing swimming performance of Arkansas Darters between treatments ($\alpha = 0.05$; $n = 15$). Individual fish were swum three times, T₀: baseline swimming ability immediately before treatment application, T₁: one day post treatment application, and T₇₋₈: seven to eight days post treatment application. Control fish were handled, sham fish were given an incision in the peritoneal cavity and had the wound sutured closed, and tag fish had an 8.0 x 1.4-mm PIT tag surgically implanted in their peritoneal cavity and the wound sutured closed.

a)		T ₀			
Treatments		DF	t-value	p-value	
Control	vs. Sham	41.72	1.73	0.0906	
Control	vs. Tag	41.72	0.44	0.6630	
Sham	vs. Tag	41.72	-1.29	0.2054	

b)		T ₁			
Treatments		DF	t-value	p-value	
Control	vs. Sham	42.94	1.01	0.3181	
Control	vs. Tag	42.94	0.31	0.7552	
Sham	vs. Tag	42.94	-0.69	0.4931	

c)		T ₇₋₈			
Treatments		DF	t-value	p-value	
Control	vs. Sham	44.9	1.08	0.2842	
Control	vs. Tag	44.9	-0.42	0.6785	
Sham	vs. Tag	44.9	-1.51	0.1386	

Table 2-3.— Comparison of the 243-day survival and tag retention rates of Arkansas Darters tagged with 8-mm PIT tags where the incisions were sutured closed or left open. Values are means (SD). There were no statistically significant differences among treatments (X^2 ; $P > 0.05$).

Treatment	<i>n</i>	TL (mm)	Wt (g)	Tag Retention Rate (%)	Survival Rate (%)
Sutured	16	51 (3)	1.42 (0.31)	100	100
Not Sutured	29	53 (3)	1.50 (0.20)	100	100

Table 2-4.— Summary of studies reporting the effects of PIT tagging on the survival and retention data for various small-bodied fishes. Fish sizes are presented in the units provided in the reference. Key: TL = total length; FL = fork length; SL = standard length; n.r. = not reported; P = polymer.

Family	Species	Common Name	Fish Size (various units)	Tag Size (mm)	Tagging Method	n	Survival (%)	Retention (%)	Study Duration (Days)	Reference
Catostomidae	<i>Catostomus commersoni</i>	White Sucker	117 mm TL	–	Control	19	95	–	30	Ficke et al. 2012
	<i>Catostomus commersoni</i>	White Sucker	113 mm TL	–	Sham Surgery	18	83	–	30	Ficke et al. 2012
	<i>Catostomus commersoni</i>	White Sucker	118 mm TL	12.5	Surgery	19	32	100	30	Ficke et al. 2012
	<i>Catostomus commersoni</i>	White Sucker	139 mm TL	23	Surgery	18	44	100	30	Ficke et al. 2012
Centrarchidae	<i>Micropterus dolomieu</i>	Smallmouth Bass	148 mm TL	23	Injection	25	96	100	60	Musselman 2007
Cichlidae	<i>Oreochromis niloticus</i>	Nile Tilapia	< 3 g	10.3 x 2.1	Surgery	12	83	90	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	< 3 g	10.3 x 2.1	Surgery + Suture	8	75	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	< 3 g	10.3 x 2.1	Injection	10	0	N/A	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	3 - 4 g	10.3 x 2.1	Surgery	14	71	90	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	3 - 4 g	10.3 x 2.1	Surgery + Suture	11	73	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	3 - 4 g	10.3 x 2.1	Injection	10	20	50	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	4 - 7 g	10.3 x 2.1	Surgery	7	100	86	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	4 - 7 g	10.3 x 2.1	Surgery + Suture	12	92	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	4 - 7 g	10.3 x 2.1	Injection	10	30	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	7 - 15 g	10.3 x 2.1	Surgery	7	100	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	7 - 15 g	10.3 x 2.1	Surgery + Suture	24	100	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	7 - 15 g	10.3 x 2.1	Injection	10	50	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	15 - 20 g	10.3 x 2.1	Injection	10	90	100	49	Baras et al. 1999
	<i>Oreochromis niloticus</i>	Nile Tilapia	20 - 25 g	10.3 x 2.1	Injection	10	100	100	49	Baras et al. 1999

Table 2-4.(continued)—

Family	Species	Common Name	Fish Size (various units)	Tag Size (mm)	Tagging Method	n	Survival (%)	Retention (%)	Study Duration (Days)	Reference
Cobitidae	<i>Misgurnus anguillicaudatus</i>	Oriental Weather Loach	84 mm TL	12.5 x 2.07	Control	32	100	N/A	30	Kano et al. 2013
	<i>Misgurnus anguillicaudatus</i>	Oriental Weather Loach	84 mm TL	12.5 x 2.07	Sham Surgery	32	100	N/A	30	Kano et al. 2013
	<i>Misgurnus anguillicaudatus</i>	Oriental Weather Loach	84 mm TL	12.5 x 2.07	Sham Injection	32	91	N/A	30	Kano et al. 2013
	<i>Misgurnus anguillicaudatus</i>	Oriental Weather Loach	84 mm TL	12.5 x 2.07	Surgery	32	97	97	30	Kano et al. 2013
	<i>Misgurnus anguillicaudatus</i>	Oriental Weather Loach	84 mm TL	12.5 x 2.07	Injection	32	93	97	30	Kano et al. 2013
Cottidae	<i>Cottus gobio</i>	Bullhead	70 mm TL	12 x 2.1	Surgery	6	100	100	28	Bruyndoncx et al. 2002
	<i>Cottus gobio</i>	Bullhead	50 - 60 mm	–	Control	12	92	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	50 - 60 mm	–	Sham Surgery	12	100	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	50 - 60 mm	12 x 2.1	Surgery	13	100	100	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	65 - 79 mm	–	Control	22	95	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	65 - 79 mm	–	Sham Surgery	20	100	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	65 - 79 mm	12 x 2.1	Surgery	21	100	90	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	80 - 94 mm	–	Control	9	100	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	80 - 94 mm	–	Sham Surgery	9	100	–	49	Knaepkens et al. 2007
	<i>Cottus gobio</i>	Bullhead	80 - 94 mm	12 x 2.1	Surgery	10	90	100	49	Knaepkens et al. 2007
Cyprinidae	<i>Camptostoma anomalum</i>	Central Stoneroller	94 mm TL	12	Injection	8	88	100	90	Musselman 2007
	<i>Hybognathus amarus</i>	Rio Grande Silvery Minnow	65 mm SL	–	Control	80	99	–	32	Archdeacon et al. 2009
	<i>Hybognathus amarus</i>	Rio Grande Silvery Minnow	65 mm SL	12.5 x 2.07	Surgery	80	87	90	32	Archdeacon et al. 2009
	<i>Hybognathus amarus</i>	Rio Grande Silvery Minnow	67 mm SL	12.5 x 2.07	Injection	80	50	89	32	Archdeacon et al. 2009
	<i>Luxilus cardinalis</i>	Cardinal Shiner	91 mm TL	12	Injection	12	100	100	90	Musselman 2007
	<i>Platygobio gracilis</i>	Flathead Chub	124 mm TL	–	Control	17	100	–	30	Ficke et al. 2012
	<i>Platygobio gracilis</i>	Flathead Chub	115 mm TL	–	Sham Surgery	18	100	–	30	Ficke et al. 2012
	<i>Platygobio gracilis</i>	Flathead Chub	119 mm TL	12.5	Surgery	17	100	100	30	Ficke et al. 2012
	<i>Platygobio gracilis</i>	Flathead Chub	115 mm TL	23	Surgery	17	100	100	30	Ficke et al. 2012
	<i>Semotilus atromaculatus</i>	Creek Chub	123 mm TL	–	Control	17	77	–	30	Ficke et al. 2012

Table 2-4. (continued)—

Family	Species	Common Name	Fish Size (various units)	Tag Size (mm)	Tagging Method	n	Survival (%)	Retention (%)	Study Duration (Days)	Reference
Cyprinidae (cont.)	<i>Semotilus atromaculatus</i>	Creek Chub	126 mm TL	–	Sham Surgery	17	82	–	30	Ficke et al. 2012
	<i>Semotilus atromaculatus</i>	Creek Chub	128 mm TL	12.5	Surgery	19	68	95	30	Ficke et al. 2012
	<i>Semotilus atromaculatus</i>	Creek Chub	127 mm TL	23	Surgery	16	63	100	30	Ficke et al. 2012
Ictaluridae	<i>Noturus exilis</i>	Slender Madtom	77 mm TL	12	Injection	13	100	100	60	Musselman 2007
	<i>Noturus flavus</i>	Stonecat	143 mm TL	12 x 2.15 P	Surgery + Suture	71	99	89	751	D'Amico et al. (In preparation)
	<i>Noturus flavus</i>	Stonecat	146 mm TL	12.1 x 2.12	Surgery + Suture	35	89	83	840	D'Amico et al. (In preparation)
	<i>Noturus flavus</i>	Stonecat	175 mm TL	23	Surgery + Suture	45	93	98	840	D'Amico et al. (In preparation)
Percidae	<i>Etheostoma blennioides</i>	Greenside Darter	95 mm TL	12	Injection	3	100	100	60	Musselman 2007
	<i>Etheostoma cragini</i>	Arkansas Darter	51 mm TL	–	Control	15	100	–	199	This study
	<i>Etheostoma cragini</i>	Arkansas Darter	52 mm TL	–	Sham Surgery	15	100	–	199	This study
	<i>Etheostoma cragini</i>	Arkansas Darter	52 mm TL	8.0 x 1.4	Surgery + Suture	15	100	100	199	This study
	<i>Etheostoma cragini</i>	Arkansas Darter	53 mm TL	8.0 x 1.4	Surgery	29	100	100	243	This study
	<i>Etheostoma cragini</i>	Arkansas Darter	51 mm TL	8.0 x 1.4	Surgery + Suture	16	100	100	243	This study
	<i>Etheostoma specabile</i>	Orangethroat Darter	60 mm TL	12	Injection	9	56	88	60	Musselman 2007
	<i>Etheostoma spilotum</i>	Kentucky Arrow Darter	> 50 mm TL	8.4 x 1.4	Injection	121	N/A	N/A	N/A	Baxter 2015
	<i>Perca fluviatilis</i>	Eurasian Perch	65 - 90 mm FL	–	Control	30	90	–	28	Baras et al. 2000
	<i>Perca fluviatilis</i>	Eurasian Perch	65 - 90 mm FL	11 x 2.2	Surgery	30	77	80	28	Baras et al. 2000
	<i>Perca fluviatilis</i>	Eurasian Perch	65 - 90 mm FL	11 x 2.2	Surgery + Suture	30	77	100	28	Baras et al. 2000
	Salmonidae	<i>Salmo trutta</i>	Brown Trout	49 mm FL	–	Control	136	96	–	27
<i>Salmo trutta</i>		Brown Trout	49 mm FL	11.5 x 2.1	Injection	145	79	n.r.	27	Acolas et al. 2007
<i>Salmo trutta</i>		Brown Trout	> 52 mm FL	11.5 x 2.1	Injection	n.r.	95	70	27	Acolas et al. 2007
<i>Salmo trutta</i>		Brown Trout	> 57 mm FL	11.5 x 2.1	Injection	n.r.	99	80	27	Acolas et al. 2007

FIGURES



Figure 2-1.— Incisions were made at approximately three fourths the length of the peritoneal cavity slightly offset from the ventral midline just anterior of the vent. The surgeon created a small flap of skin by gently pushing against the body cavity with their thumb, then pierced the raised section of skin at an approximately zero-degree angle perpendicular to the long axis of the fish using a #12 scalpel blade. The PIT tag is then inserted anteriorly to avoid impacting the gut.

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CHAPTER 3: EFFECTS OF SLOPE ON SMALL-BODIED FISH PASSAGE SUCCESS IN AN EXPERIMENTAL ROCK RAMP FISHWAY

INTRODUCTION

Streams and rivers of the Great Plains of North America are highly fragmented due to the construction of instream barriers, including those used to capture and divert water for municipal and agricultural uses (Perkin et al. 2015). Maintaining the longitudinal connectivity of stream systems is imperative to ensure the continued persistence of stream fishes (Winston et al. 1991; Schlosser and Angermeier 1995; Fausch and Bestgen 1997; Platania and Altenbach 1998; Perkin and Gido 2011; Walters et al. 2014; Perkin et al. 2015). Some stream fishes require uninterrupted stream segment lengths in excess of 100 km to access habitats to complete their life history and avoid extirpation (Perkin and Gido 2011). Currently, the Great Plains is a highly fragmented ecosystem with over 19,000 known physical barriers to fish movements in the central region alone (Perkin et al. 2015). The sheer number of physical structures, and their utility in delivering societal and economic benefits makes it unlikely that most of these structures will be removed. However, the barrier effect of such structures, such as low-head diversions, can be mitigated by building fish passage structures (Ficke et al. 2011; Ficke 2015).

There are a variety of fish passage structure or fishway designs used on streams and rivers. Pool and weir fishways were optimized for large-bodied coastal migratory species such as Atlantic Salmon *Salmo salar* and Pacific salmon *Oncorhynchus spp.* (Katopodis and Williams 2012; Noonan et al. 2012). A key assumption of these fishways is that the target species are willing and capable of jumping over a series of weirs to ascend vertical barriers. This design has

been successfully used to pass salmonids and other species that are behaviorally and physiologically adapted for leaping instream obstacles. Pool-weir-and-orifice, Denil and vertical slot fishways were designed for species capable of making short sprints through high velocity segments to ascend vertical barriers (Katopodis and Williams 2012).

Unfortunately, species that lack the behavioral motivation or physiological ability to jump or navigate high velocities can be excluded by pool-and-weir, Denil, and vertical slot fishways (Billman and Pyron 2005; Leavy and Bonner 2009; Ficke et al. 2011; Ficke 2015; Prenosil et al. 2016). This is especially true for fishes native to the Great Plains of North America, many of which are small-bodied as adults (< 230 mm TL) and have evolved in streams lacking abrupt vertical drops and frequent high velocity areas (Fausch and Bestgen 1997). Because of this, an alternate fishway design is needed to allow effective passage of these fishes—the design that appears most suitable is the rock ramp fishway.

Rock ramp fishways (sometimes referred to as nature-like fishways) are comprised of a sloped section of channel with roughness elements installed on the bed to provide velocity refuges and decrease water velocity for fishes as they ascend the fishway. This design is widely recognized as a good choice to allow passage of small-bodied fishes because they can be built without vertical drops, high velocity sections, and provide heterogeneous hydraulic conditions to accommodate a diversity of swimming behaviors. The successful passage of some Great Plains fishes (e.g., Rio Grande Silvery Minnow *Hybognathus amarus*, Flathead Chub *Platygobio gracilis*, and Longnose Dace *Rhinichthys cataractae*), has been recorded at rock ramp fishways (Archdeacon and Remshardt 2012; Ficke 2015; M. C. Kondratieff, and R. M. Fitzpatrick, Colorado Parks and Wildlife, personal communication) but refinement of the design may lead to

greater passage success, especially for fishes with relatively poor swimming performance such as darters (*Etheostoma spp.*).

This study was designed to improve the design of rock ramp fishways by identifying the ideal slope and length combinations for successful passage of small-bodied Great Plains fishes. A custom-made adjustable full-scale fishway (described in Chapter 1) was used to test fish passage success at slopes of 2 to 10%. This range of slopes, and associated water velocities, encompasses the range of slopes of existing or proposed fishways used along Colorado's Front Range.

METHODS

Three representative Great Plains fish species were chosen for this project, based on their availability and on the existence of prior swimming performance data. The swimming performance data allowed me to select species with excellent, moderate, and poor swimming abilities, relative to each other. Flathead Chub was chosen as the excellent swimming species, based upon work by Ficke et al. (2012) and Ficke (2015). Stonecat *Noturus flavus* was selected as the species with moderate swimming ability, based upon the work of Ficke (2015), and Arkansas Darter *Etheostoma cragini* was designated as the species with poor swimming ability, as described by Ficke et al. (2011).

When possible, wild-caught fish were used in the study. Flathead Chubs (62 – 149 mm TL; 1.73 – 37.32 g) were collected from Fountain Creek, CO, USA, a turbid, sand-bed stream, using backpack electrofishers (Smith-Root, Vancouver, WA, USA). Stonecats (94 – 213 mm TL; 7.54 – 121.22 g) were collected from Horse Creek, WY, USA, a small, clear stream, using backpack

electrofishers and a barge electrofisher (Smith-Root, Vancouver, WA, USA). Wild-caught Arkansas Darters were not available in sufficient numbers, so hatchery reared Arkansas Darters (47 – 67 mm TL; 0.94 – 3.23 g) were obtained from the Colorado Parks and Wildlife Native Aquatic Species Restoration Facility in Alamosa, CO. All fish were transported to the Colorado State University Foothills Fisheries Laboratory where they were separated by species and held in round 300-l tanks receiving continuous flows of air-saturated water. Temperatures were regulated by solenoid valves controlled by programmable temperature controllers. Water was delivered through spray bars creating 0.05 m/s - 0.10 m/s currents to provide cues for rheotactic orientation and opportunity for exercise. Current direction was changed weekly.

After one week of post-transport recovery and acclimation to laboratory conditions, tank temperatures were increased by 1°C/day to the $20 \pm 1^\circ\text{C}$ test temperature. Fish were held at 20°C for at least 3 weeks prior to testing to allow sufficient time for physiological acclimation. Artificial habitat (PVC pipes and plastic vegetation) provided fish with cover. Fish were fed daily satiation rations of a mixture of natural and prepared feeds (e.g., frozen bloodworms and pelleted feed).

Prior to use in fishway experiments, each fish was tagged with a size-appropriate PIT tag (8 x 1.4-mm [Arkansas Darters and Flathead Chubs], 12 x 2.15-mm [Stonecats and Flathead Chubs], or 23-mm [Stonecats and Flathead Chubs]; Oregon RFID, Portland, OR, USA). A small incision was made in the peritoneal cavity approximately three fourths the length of the body cavity just anterior of the vent using a #12 scalpel blade. A PIT tag was inserted anteriorly into the incision. Arkansas Darter and Flathead Chub incisions were not sutured and a drop of Kordon® Pond Fish Protector™ was massaged onto the incision as a prophylactic to expedite the healing process (Swanson et al. 1996). Stonecat incisions were sutured closed using a

Braunamid DS24 polyamide pseudo-monofilament suture attached to a 24-mm curved suture needle. Stonecats were sutured because they were being observed for a tag retention study (D'Amico, unpublished data). Fish recovered from tagging for at least three weeks to ensure the incision was fully healed and the tag was retained before being tested in the fishway.

Fish passage was tested at slopes of 10, 8, 6, 4, and 2%, starting with the steepest slope. These slopes were chosen because they encompass the steepest slopes seen in fishways that have been installed on streams in the transition zone between the plains and mountain streams in Colorado and the gentlest slope recommended for a Great Plains fishway (White and Mefford 2002; Ficke 2015). Fish species were tested separately in groups of five (Stonecats) or ten individuals per trial (Flathead Chub and Arkansas Darters). There were nine replicate trials per species at each slope; prior experience with a similar flume study suggested that this was enough replicates to detect difference among treatments. Individual fish were reused for a maximum of three trials at the same slope to reduce the number of fish required for the study. Individual fish were given at least two days of rest between fish passage trials. Water temperatures in the fishway were adjusted to match those of the fish holding tanks (e.g., of $20 \pm 1^\circ\text{C}$). Flows to the fishway at each slope treatment (10, 8, 6, 4, and 2%) were set to 0.0363, 0.0316, 0.0229, 0.00963, and 0.00743 m^3/s (1.282, 1.116, 0.808, 0.343, and 0.262 cfs), respectively, to maintain an average water depth of approximately 51-mm at each slope treatment as described in Chapter One.

Two concrete cinder blocks (39.7 x 19.4 x 19.4 cm) were placed in the downstream refuge area to provide cover. The blocks were placed side-by-side, approximately 20 cm apart, and oriented so the openings faced outward, perpendicular to the flow, for the Flathead Chub and Stonecat trials. For the Arkansas Darter trials, the blocks were placed next to each other, oriented

so they formed a wall with the openings facing vertically to provide additional velocity refuge (Figure 3-1). An upstream refuge area was created by the combination of the uppermost PIT tag antenna (A4), four 76-mm long sections of 152-mm diameter PVC pipe oriented perpendicular to flow, and three rock slabs (approximately 40 x 25 x 2.5 cm) leaned against the PIT tag antenna to prevent the flow from dislodging the antenna (Figure 3-1).

At 15:00 h on a trial day, fish were transferred from their tanks to the fishway in a 50:50 mixture of tank and flume water. All PIT tag antennas were tested before every trial using a dummy 8-mm PIT tag. After testing the antennas, fish were placed in the downstream refuge area of the fishway and given 20 hours to ascend the fishway, with all trials ending at 11:00 h. A 14L:10D photoperiod (lights on at 06:00, lights off at 20:00) was used to provide the fish with equal amounts of light and dark while in the fish passage structure. After each trial, the fish were captured, scanned and identified with a handheld PIT tag reader, and returned to holding tanks. Data were downloaded from the PIT tag readers after each trial.

Data Analyses

Fish movement data were analyzed with Program MARK (White and Burnham 1999) to calculate maximum likelihood estimates of movement probabilities. Estimates were derived from tag detection data that describe individual detection histories at the four antennas used in the study. Data gathered were the number of tagged fish that were placed in the flume, encountered the first, second, third, fourth antennas, and if fish were found above the fourth antenna at the end of each trial. Thus, a capture history matrix of 111000 would indicate a fish was placed in the flume, detected at antennas A1 and A2, but did not ascend to antenna A3. A capture history of 111111 indicated that the fish was placed in the flume, detected at all four PIT tag antennas, and then found in the upstream refuge area at the end of the trial. Full passage

success was defined as ascending the entire 6.1-m fishway by being detected at the uppermost antenna (A4) or being found in the upper refuge area at the end of the trial (Figures 3-1 and 3-2). Partial passage success was defined as successfully traversing a portion of the fishway determined by detection on any of the lower PIT tag antennas (A1 – A3). The Cormack-Jolly-Seber (CJS) model was used to analyze the movement data based on a simple linear array to determine the probability of full or partial passage success (Burnham et al. 1987). Full and partial passage success data were used to identify the ideal length and slope combinations to build future fish passage structures by observing the probability of a fish reaching one of the PIT tag antennas placed every 2.03 m in the fishway at a given slope.

Model covariates for apparent probability of movement between antennas (ϕ) included: antenna (t), slope, number of trials completed, fish TL (mm), and all two-way interactions. Model covariates for detection probability of an antenna (p) included: antenna (t), slope, tag size (8-mm [Flathead Chub and Arkansas Darters], 12-mm [Flathead Chub and Stonecats] or 23-mm [Flathead Chub and Stonecats] coded as dummy variables of 0, 1, or 2, respectively), tag duplex (half or full; coded as 0 or 1), and the two-way interactions between slope and all other p covariates. AIC_c model selection was used to identify the model of best fit for each species. Model selection for each species was carried out by building competing sets of models for p and setting ϕ covariates to the global model. Once the model of best fit for p was identified using AIC_c model selection, the process was repeated using the model of best fit for p and all competing models for ϕ and until the model of best fit for ϕ was identified (Tables 3-1, 3-2, and 3-3).

Model selection was run independently by species because of the known or assumed differences in swimming performance (Ficke et al. 2011, 2012; Ficke 2015; Swarr and Myrick,

unpublished data). For the analyses, $\varphi_1 \neq \varphi_2 \neq \varphi_3 \neq \varphi_4$, $\varphi_5 = 1$, and $p_2 = p_3 = p_4 \neq p_5$ (Figure 3-2). To avoid confounding the φ_4 condition, I recorded separately fish that were found on or above A4 in the upstream refuge area at the end of a trial; this was designated as φ_5 . Additionally, φ_5 gave me more accurate passage probability estimates because fish were likely to rest on or near A4 due to its proximity to the upstream refuge area, causing tag collisions that were not read by the PIT tag reader. Because I assumed that if a fish made it to A4 it had successfully entered the upstream refuge area, φ_5 was set to 1 (Figure 3-2). Due to the likelihood of tag collisions occurring mostly at A4 once fish made it into the upstream refuge area, p_5 was set as not equal to p_2 , p_3 , and p_4 . For this study I was only concerned with the probability of upstream passage success and did not evaluate downstream movements or the number of attempts. The sheer volume of tag detections per trial (up to 40,000 detections at one antenna for one trial) made more comprehensive analyses prohibitive without developing analytical code to process the data, which was beyond the scope of this study.

The daily activity patterns of each species were determined from time stamp data for each initial PIT tag detection. Daily activity data were analyzed to determine the proportion of day versus night movements for each species. Data from 2% slope were used to examine activity patterns because all species were likely to have the highest probability of fully ascending the fishway. The proportion of detections occurring by day and night were compared with bootstrap estimates in Program R (version 3.3.1) that were created by resampling the observed data from the 2% slope trials 10,000 times and measuring detections occurring during each time period (Table 3-4).

RESULTS

Fish Passage

Passage success in the fishway varied by species, with strong-swimming Flathead Chub having the highest probability of passage success at higher gradient treatments, followed by Stonecats, and then Arkansas Darters (Figures 3-3, 3-4, and 3-5). Detection probability at each antenna was approximately 1.0. The CJS model determined that the true probabilities of Flathead Chub successfully ascending a 6.1-m rock ramp fishway at slopes of 2, 4, and 6% were 1.0. Probability of Flathead Chub passage success was very high (0.96) for a 4.06-m, 8% slope fishway (Figure 3-3). Stonecats had a successful passage probability of 1.0 for a 6.1-m fishway at 2 and 4% slopes, and a successful passage probability of 0.83 for a 4.06-m, 6% slope fishway (Figure 3-4). Arkansas Darters never achieved a probability of 1.0 for ascending a 6.1-m fishway. However, their probability of partial passage success was moderate for a 2.03-m, 4% slope fishway with a probability of 0.43, and for a 4.06-m, 2% slope fishway with a probability of 0.54 (Figure 3-5).

Daily Activity Patterns

Both Stonecats and Flathead Chub were more active during darkness and substantially reduced movement during the day. Arkansas Darters moved more often during daylight and reduced their activity levels at night (Figure 3-6). The frequency of fish detections during the 2% slope trials was highest for Flathead Chub, peaking at over 12,000 detections per hour from 20:00 – 21:00, immediately after the end of the simulated daylight period. Stonecat detections also peaked between 20:00 – 21:00, reaching 8,800 detections per hour. Arkansas Darter

detections were highest in the morning once simulated daylight resumed, peaking at approximately 2,300 per hour.

Bootstrap estimates of the proportion of daytime movement were similar for Flathead Chub and Stonecat (Table 3-4) and provided evidence that these species preferentially moved during darkness. Bootstrap estimates for Arkansas Darters indicated that they were more active during the day (Table 3-4).

DISCUSSION

Fish Passage

My results show that rock ramp fish passage structures can be designed to effectively pass a variety of small-bodied fish species including those with relatively poor swimming ability. Fish passage success of all three species was maximized with shorter fishway segments (~ 2 m long) at a 4% slope (Figures 3-3, 3-4, and 3-5); other combinations (greater lengths and/or steeper slopes) led to reductions in the passage success of the three species tested. I identified clear differences in passage success between species presented with fishways of identical configuration.

Flathead Chub were the most successful at ascending the fishway, as was expected based on available swimming performance data. Mean sprint velocity for Flathead Chub in a swim tunnel was 114 cm/s (Ficke et al. 2012; Ficke 2015), and this velocity was only exceeded in the fishway when the slope was 10% (Chapter 1). The mean and maximum water velocities at 10% slope were 85 cm/s and 132 cm/s respectively, which may help explain why the Flathead Chub were not capable of ascending the full 6.1-m fishway, but could ascend to the 4.06-m mark (Figure 3-

3). At all other slope configurations, mean and maximum water velocities were lower than their published sprinting velocity, suggesting that Flathead Chub should be able to successfully ascend the fishway barring limitations due to endurance, which was what I observed.

Ficke (2015) reported a mean sprinting velocity for Stonecat of approximately 62 cm/s. Stonecats had a relatively high probability of partially ascending the fish passage structure at 6% slope when mean and maximum water velocities were 58 cm/s and 98 cm/s respectively (Figure 3-4). When the fishway slope resulted in mean water velocities that exceeded 62 cm/s, as was the case with the 8% and 10% slope treatments, the probability of successful Stonecat passage success declined sharply.

Arkansas Darter passage success was low compared to Flathead Chub and Stonecat, even though the maximum sprinting velocity for this species was estimated to be approximately 66 cm/s (Swarr and Myrick, unpublished data). Swimming endurance data available for Arkansas Darter showed endurance was poor (0.2 min) at water velocities of 32 cm/s in a swim tunnel (Ficke et al. 2011). Although Arkansas Darter sprint velocity is similar to that of Stonecats, their endurance appears to be lower, in light of this fish passage data (Figures 3-4 and 3-5). Ficke (2015) observed physiological traits of 15 species of Great Plains fishes by taking cross sections of each species at approximately 50% of their total length and measuring the proportion of red muscle, which is aerobically fueled, and white muscle, which is anaerobically fueled (Gordon 1968). Both Johnny Darter *E. nigrum* and Iowa Darter *E. exile* had no red muscle at 50% of their total length, whereas cross sections of Black Bullhead and Flathead Chub had a mean of $3.0 \pm 0.5\%$, and $4.0 \pm 1.0\%$ respectively (Ficke 2015). The lack of red muscle may indicate why some Great Plains darters have relatively poor aerobic swimming abilities. Arkansas Darter passage probability was comparatively low at all slope treatments except for 2%, when mean and

maximum water velocities were 14 cm/s and 30 cm/s respectively. This indicates that fish swimming performance data collected using swim tunnels provides an approximation of fish passage ability for small-bodied fishes because they accurately depict physiological swimming ability. However, they are not perfect approximations of passage performance because of the known effects that swimming in a confined swim tunnel has on swimming ability and behavior (Peake and Farrell 2006). Literature values for swimming ability and fish passage rates observed in this study were similar, especially when both swimming endurance and sprinting velocity data are available. Trends in fish passage success seen in this study for all three species conform with available swimming performance data (Ficke et al. 2011, 2012; Ficke 2015).

Based on prior swimming performance data, the results of this study can be used to design fishways for species of similar swimming ability. For example, Stonecat sprinting speed is similar to that of another ictalurid species, Black Bullhead *Ameiurus melas*, suggesting similar probabilities of fish passage in a rock ramp fishway (Ficke 2015). The measured sprinting ability for Arkansas Darter was very similar to that of Johnny Darter and Iowa Darter, indicating that passage success would likely be comparable for those darter species in a rock ramp fishway (Ficke 2015). This may provide managers some flexibility in choosing a ramp type for a given assemblage of fish, in the absence of detailed swimming speed information.

Although the average fishway depth used for this study was shallow (51 mm) and flows in the fishway were low, I achieved high passage rates for adult Flathead Chub and Stonecat. In a field situation, this would be beneficial because the amount of water allocated for fishway function at an irrigation diversion can be quite low. If water depth was increased for each slope treatment it is likely that passage success would have increased as a result of the lower Froude number throughout the fishway, the formation of a boundary layer around the roughness

elements, and the increased potential pathways for fish to choose from as they ascended the fishway (Cheong et al. 2006). Given these constraints, this study could be construed as providing conservative estimates for fish passage success; nevertheless, using these conservative estimates may lead to higher field passage rates. To calculate the Froude number (Fr) of a system one can use the following equation (Lamouroux et al. 2002):

$$i. \quad Fr = \frac{v}{\sqrt{g \times d}}$$

Where v is the water velocity, g is acceleration due to gravity and d is water depth. $Fr < 1$ is subcritical flow, $Fr = 1$ is critical flow, and when $Fr > 1$ this indicates super-critical flow leading to the formation of hydraulic jumps.

Caution should be taken to not use unnecessarily shallow depths in future fishways. In a natural setting predation may be increased in areas of shallow depth where fewer routes of escape are available for prey species. Deeper water will benefit larger-bodied fishes and allow a wider assemblage of fishes to use the structure (Harvey and Stewart 1991). Unnecessarily shallow water may also lead to avoidance behaviors and alter fish daily activity patterns by allowing increased light penetration into the water column (Mallen-Cooper 1996; Jones 2017).

Swimming Behaviors

Observation of the Flathead Chubs, Stonecats, and Arkansas Darters during passage indicated that each species used a variety of approaches to negotiate the fishway. At higher gradients, Flathead Chubs often held position below the hydraulic jump at the downstream end of the fishway and would individually sprint up the fishway until they became exhausted or successfully ascended the entire structure. At lower gradients they would stay in schools and methodically work their way up the fishway, using the interstices between roughness elements. Occasionally, Flathead Chubs held position behind roughness elements by angling their pectoral

fins downwards like ailerons and constantly beating their caudal fin with tailbeat frequency changing based on water velocity in the fishway.

Unlike Flathead Chubs, which schooled at lower slopes, Stonecats were never observed using schooling behaviors and almost always ascended the fishway individually. Stonecats negotiated the fishway at higher gradients, although their larger average size (compared to the Flathead Chubs) and anguilliform swimming mode caused them to contact roughness elements as they went. Providing heterogeneous roughness element spacing could possibly accommodate sprinting behaviors in larger-bodied species such as Stonecats. Using heterogeneous roughness element sizes would also increase hydraulic diversity in the fishway and may accommodate swimming behaviors of a larger variety of fish species if hydraulic conditions were carefully designed not to exceed the swimming ability of target species. At lower gradients Stonecats also used their pectoral fins to hold position downstream of roughness elements, but tailbeat frequencies were lower than those used by Flathead Chubs to hold and correct position.

Of the three species, Arkansas Darters used the roughness elements the most during their fishway ascents. Like Stonecats, the darters showed no schooling or group ascent behaviors. In high velocities they would sometimes dart from roughness element to roughness element and then hold position by tucking their body completely against the downstream side of a roughness element in the area of lowest velocity. They also used both pectoral and pelvic fins to anchor themselves between the roughness elements and the side slope of the trapezoidal channel or in seams between fishway substrate plates as they ascended the fishway. Because the floor of the fishway was smooth PVC, I presume that it was difficult for the darters to hold position using their fins. Baker and Boubée (2006) found that by increasing the roughness of an artificial ramp set to 26.8% slope, the proportion of fish successfully ascending the ramp was significantly

higher than using a smooth plastic ramp for adult Redfin Bully *Gobiomorphus huttoni* (0.6 – 1.0 versus 0.3) and Inanga *Galaxias maculatus* (0.6 – 0.9 versus 0.1). Further exploration of the relationship between fishway surface texture and passage success for small-bodied Great Plains fishes is warranted, particularly for benthic-oriented species like darters and suckers. Future fishway designs could also incorporate crevasses for small-bodied benthic fishes to hide in or cling to as they ascend the fishway, provided there was a way to keep them from filling with sediment. Based on the behaviors observed in this study and the results of Baker and Boubée (2006), if a future fishway were constructed of concrete, I recommend roughening the concrete surface to provide adhesion surfaces for benthic fish passage.

When designing future rock ramp fishways I would recommend using a trapezoidal channel cross-section to allow for a diversity of depth and velocity conditions at any given cross-section. The trapezoidal shape has been used successfully in multiple of fishway projects in the state of Colorado (Bestgen et al. 2010; Ficke 2015; M. C. Kondratieff, and R. M. Fitzpatrick, Colorado Parks and Wildlife, personal communication). Providing a wetted margin in an artificial ramp with a side slope of 10° was essential for the passage success of Redfin Bully, a small-bodied benthic fish species (Baker and Boubée 2006). Side slopes of the fishway in this study were 30°, and provided a larger wetted margin for fish passage compared to the 42° side slopes in the fishway used by Ficke (2015). A. D. Ficke (personal communication) hypothesized that Johnny Darters were using the wetted margin of a trapezoidal rock ramp fishway (side slopes of 42°) to ascend the structure, although it was never observed. Arkansas Darters in this study were observed occasionally using the wetted margin of the trapezoidal channel to ascend the fishway when little turbulence was present. This confirms Ficke's (2015) hypothesis on darter ascent behavior and underscores the importance of providing diverse velocity and depth conditions

throughout fishways. Further investigations into the effects of side slope angle on fish passage success rates would be beneficial.

Daily Activity Patterns

Flathead Chub were more active at night, which is possibly explained by an adaptation to turbid waters across their range (Olund and Cross 1961; Davis and Miller 1967; Bonner and Wilde 2002). The nocturnal increase in activity could have been caused by the very low turbidity of my fishway. Fountain Creek, CO, where the fish were captured, is a sand-bed stream that is generally turbid (Haworth 2015), while the water in my fishway was clear. The fish could have altered their behavior to preferentially move during darkness because it offered more concealment from perceived predation threats. During the eight and ten percent slope trials, Flathead Chub sought refuge beneath the bubble curtain that formed below the hydraulic jump at the bottom of the fishway during daylight. At lower slopes the hydraulic jump and bubble curtain were not present, and Flathead Chub sheltered in the openings in the cinder blocks in the downstream refuge area, suggesting that the fish were actively seeking areas of lower light levels or with less exposure to potential overhead predators. Field observations of movement patterns and timing may reveal the true daily activity patterns of this species in the wild. Stonecat activity was strongly associated with darkness, not an unexpected behavior pattern for an ictalurid catfish. The Stonecats generally sought refuge in the cinder blocks at the bottom of the fishway during daylight with only occasional attempts to ascend the fishway. Arkansas Darters were most active during daylight and reduced their activity during the night. The darters were often observed seeking low velocity areas, even while in the downstream refuge area. Bootstrap estimates of the proportion of daytime movements support the behaviors observed in the fishway and the trends seen in movement patterns (Figure 3-6; Table 3-4).

My data suggest that fish movements are affected by light level and have implications for wild fish related to their diel activity patterns (Mallen-Cooper 1996; Ono and Simenstad 2014; Jones et al. 2017). For example, if nighttime ambient light levels near a stream were artificially increased (e.g., street lamps near streams), my results suggest that some fish species, such as Stonecats or Flathead Chubs might reduce their activity or actively avoid the light. Jones et al. (2017) documented a reduction in passage of Freshwater Shrimp *Macrobrachium australiense* and Freshwater Prawn *Paratya australiensis* in a vertical slot fishway during daylight or when artificial light was placed over the fishway, indicating that this behavior may be shared among other aquatic taxa. Additionally, for diurnal species such as Arkansas Darters, if perpetual darkness is present in a fishway or other instream structure (e.g. bridge, culvert, etc.), the fish may actively avoid areas of lower light level, even if the hydraulic conditions allow passage. Similar aversion to darkness was documented in five Australian freshwater fish species in a vertical slot fishway (Jones et al. 2017). Movements of migratory fishes in the Murray River through the fishway decreased under low-light conditions and increased when the fishway was exposed to daylight or artificial light of similar intensity. Mallen-Cooper (1996) was able to reduce and exclude the passage of Silver Perch *Bidyanus bidyanus* and Bony Herring *Nematalosa erebi* respectively, by installing a tunnel to block sunlight over a portion of a different vertical slot fishway on the Murray River. The movement of Silver Perch, Bony Herring, Australian Smelt *Retropinna semoni*, and Common Carp *Cyprinus carpio* through the fishway were also strongly associated with daylight (Mallen-Cooper 1996).

Light levels in streams are also affected by naturally and anthropogenically induced levels of turbidity. Light levels in streams with unnaturally high levels of sediment inputs are reduced, while those in clear, sediment-starved systems are increased, potentially impacting fish behavior

if they are not adapted to those levels of turbidity. It is known that altered turbidity levels can change feeding success and aggressive behaviors between individuals (Sweka and Hartman 2001; Hazelton and Grossman 2009). Bonner and Wilde (2002) documented a reduction in prey consumption for a variety of Great Plains minnows under increased turbidity conditions. Based on my fish movement data for Flathead Chub, I hypothesize that altered turbidity levels also affect fish movement patterns.

Conclusions

Designing a fishway using criteria for strong swimming species such as Flathead Chub may impose artificial selection against species with weaker swimming abilities, such as Arkansas Darters, and exclude them from upstream passage. Therefore, when designing a fishway for a community of fishes, it is ideal to use passage data from a species with the lowest swimming ability to ensure that a majority of the fish assemblage can successfully use the structure. In the case of this study, a fishway designed with Arkansas Darters in mind would have a slope of 4% and fishway segments of around 2 m in length. These are passable by a majority of the darters and by all Stonecats and Flathead Chubs I tested. My study did not test the effects of fish size or age on passage performance, but given that younger/smaller fish generally do not swim as well as their adult conspecifics, fishways designed for the slower-swimming species should also increase the passage probability of juveniles of some of the fish assemblage.

Although this study focused on specific gradients as treatment effects, in reality the design of fishways should be thought of in terms of velocity conditions rather than purely gradient alone. As slope increases, velocity will also increase, however, hydraulic conditions inside of the fishway can be manipulated by altering water depth, fishway material, and roughness element size and spacing. It is of utmost importance to design fishways that do not exceed the

swimming ability of target species by understanding the physiological limitations of said species. By using the design guidelines and principles suggested here for rock ramp fishways, the rates of passage success of future plains fish passage structures can be increased for a variety of small-bodied fishes.

TABLES

Table 3-1.— Cormack-Jolly-Seber models used to estimate apparent passage success (ϕ) and detection probability (p) for Flathead Chub. The top eight models selected by AIC_c are shown. Effects included slope, trial, fish total length (TL), antenna (t), tag type (HDX or FDX), tag size, and all two way interactions.

Model	AIC_c	ΔAIC_c	AIC_c Weights	Model Likelihood	K	Deviance
{ $\phi(t + \text{slope} + \text{Trial} + t*\text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	770.86	0.00	0.33	1.00	21	728.34
{ $\phi(t + \text{slope} + \text{Trial} + t*\text{Trial} + \text{slope}*\text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	772.04	1.17	0.18	0.56	22	727.47
{ $\phi(t + \text{slope} + \text{Trial} + t*\text{Trial} + \text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	772.43	1.57	0.15	0.46	18	736.05
{ $\phi(t + \text{slope} + \text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	774.33	3.47	0.06	0.18	18	737.95
{ $\phi(t + \text{slope} + \text{Trial} + t*\text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL}) p(t3+ \text{slope} + t3*\text{slope})$ }	774.74	3.88	0.05	0.14	20	734.26
{ $\phi(t + \text{slope} + \text{Trial} + \text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	775.01	4.15	0.04	0.13	15	744.74
{ $\phi(t + \text{slope} + \text{Trial} + \text{slope}*\text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	775.05	4.18	0.04	0.12	19	736.62
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + t*\text{Trial} + \text{slope}*\text{Trial} + \text{TL} + t*\text{TL} + \text{slope}*\text{TL} + \text{TL}*\text{Trial}) p(t3+ \text{slope} + t3*\text{slope})$ }	775.92	5.06	0.03	0.08	25	725.19

Table 3-2.— Cormack-Jolly-Seber models used to estimate apparent passage success (ϕ) and detection probability (p) for Stonecat. The top eight models selected by AIC_c are shown. Effects included slope, trial, fish total length (TL), antenna (t), tag type (HDX or FDX), tag size, and all two way interactions.

Model	AIC_c	ΔAIC_c	AIC_c Weights	Model Likelihood	K	Deviance
{ $\phi(t + \text{slope} + t*\text{slope} + \text{TL}) p(t3+ \text{slope})$ }	280.34	0.00	0.24	1.00	13	253.91
{ $\phi(t + \text{slope} + t*\text{slope}) p(t3+ \text{slope})$ }	280.40	0.06	0.23	0.97	12	256.03
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{TL}) p(t3+ \text{slope})$ }	282.13	1.79	0.10	0.41	14	253.63
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial} + \text{TL} + \text{slope}*\text{TL}) p(t3+ \text{slope})$ }	283.01	2.67	0.06	0.26	16	250.36
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + t*\text{Trial} + \text{slope}*\text{Trial} + \text{TL} + \text{slope}*\text{TL}) p(t3+ \text{slope})$ }	283.18	2.84	0.06	0.24	19	244.27
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{TL} + \text{Trial}*\text{TL}) p(t3+ \text{slope})$ }	283.77	3.43	0.04	0.18	15	253.20
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial}) p(t3+ \text{slope})$ }	284.08	3.74	0.04	0.15	14	255.58
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + t*\text{Trial} + \text{slope}*\text{Trial} + \text{TL}) p(t3+ \text{slope})$ }	284.15	3.80	0.04	0.15	18	247.33

Table 3-3.— Cormack-Jolly-Seber models used to estimate apparent passage success (ϕ) and detection probability (p) for Arkansas Darter. The top eight models selected by AIC_c are shown. Effects included slope, trial, fish total length (TL), antenna (t), and all two way interactions.

Model	AIC_c	ΔAIC_c	AIC_c Weights	Model Likelihood	K	Deviance
{ $\phi(t + \text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL} + t*\text{TL}) p(t3+ \text{slope})$ }	767.01	0.00	0.23	1.00	18	730.35
{ $\phi(t + \text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial}) p(t3+ \text{slope})$ }	767.83	0.82	0.15	0.66	14	739.43
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial}) p(t3+ \text{slope})$ }	768.17	1.15	0.13	0.56	17	733.58
{ $\phi(t + \text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL}) p(t3+ \text{slope})$ }	768.23	1.21	0.13	0.55	15	737.77
{ $\phi(t + \text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL} + \text{Trial}*\text{TL}) p(t3+ \text{slope})$ }	768.87	1.86	0.09	0.39	16	736.35
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL} + t*\text{TL}) p(t3+ \text{slope})$ }	769.68	2.66	0.06	0.26	21	726.78
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL} + \text{slope}*\text{TL} + t*\text{TL}) p(t3+ \text{slope})$ }	770.39	3.37	0.04	0.19	22	725.41
{ $\phi(t + \text{slope} + t*\text{slope} + \text{Trial} + \text{slope}*\text{Trial} + t*\text{Trial} + \text{TL} + \text{Trial}*\text{TL} + t*\text{TL}) p(t3+ \text{slope})$ }	770.75	3.73	0.04	0.15	22	725.77

Table 3-4.— Bootstrap estimates of the proportion of fish movements occurring during daylight as determined by detection on any of four PIT tag antennas in the rock ramp fish passage structure at 2% slope during 20 hour passage trials. Estimates for proportion of daytime movements are shown for Flathead Chub (FHC), Stonecat (STP), and Arkansas Darter (ARD).

Species	Proportion of Daytime Movement	Standard Error	Confidence Interval
FHC	0.244	0.019	(0.21, 0.28)
STP	0.234	0.032	(0.17, 0.30)
ARD	0.903	0.019	(0.87, 0.94)

FIGURES



Figure 3-1.— Photographs of the orientations of the cinderblocks in the downstream refuge area for the Flathead Chub and Stonecat trials (left) and for the Arkansas Darter trials (center). Photo of the upstream refuge area with the uppermost PIT tag antenna (A4; right).

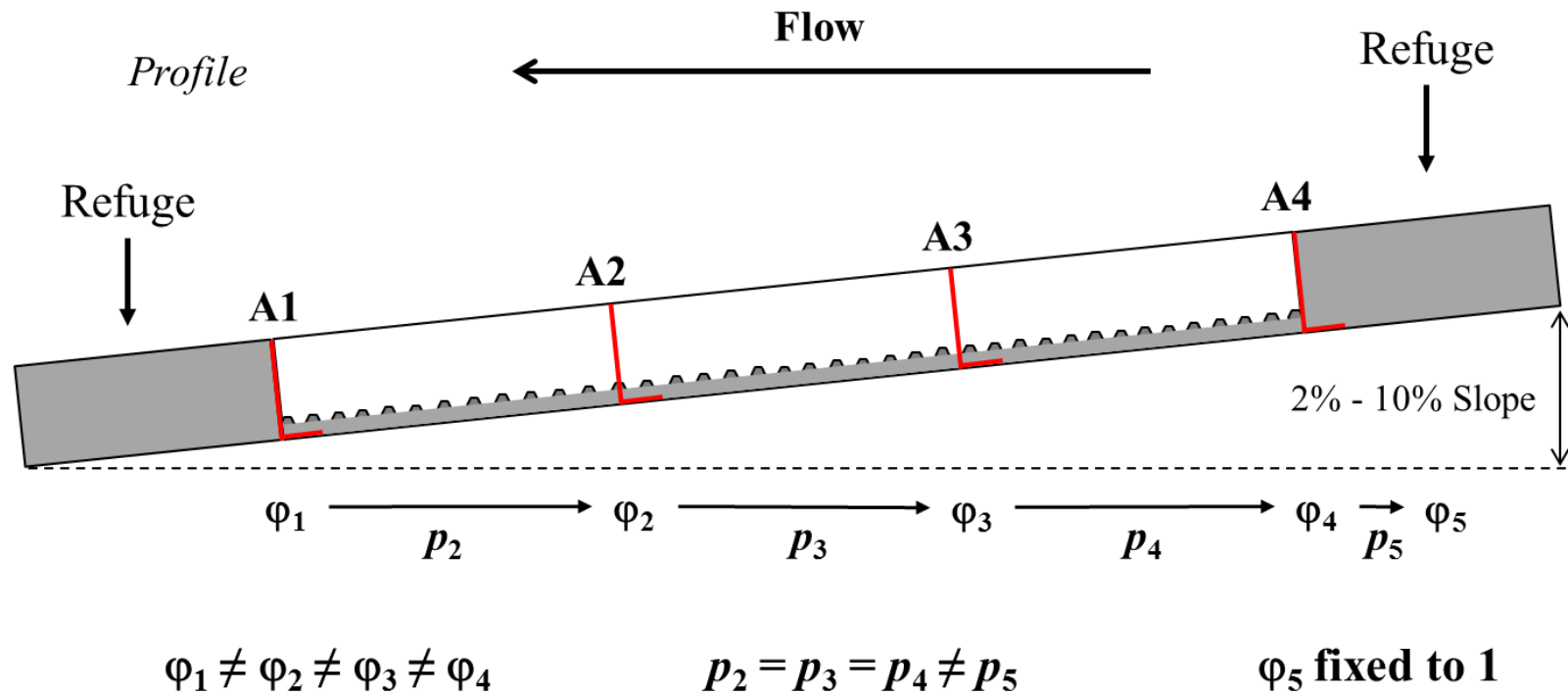


Figure 3-2.— Conceptual model for the analysis of passage data in program MARK using the Cormack-Jolly-Seber model. A1 – A4 indicate the locations of the four swim-over PIT tag antennas used to monitor fish movements. Fish were placed in the refuge downstream of A1 at the start of each trial and given 20 hours to attempt to ascend the flume. Diagram is to scale for the 9.1 x 1.2 x 0.6-m flume used in the study. Apparent probability of fish movement between antennas is shown as φ . Detection probability of an antenna is shown as p .

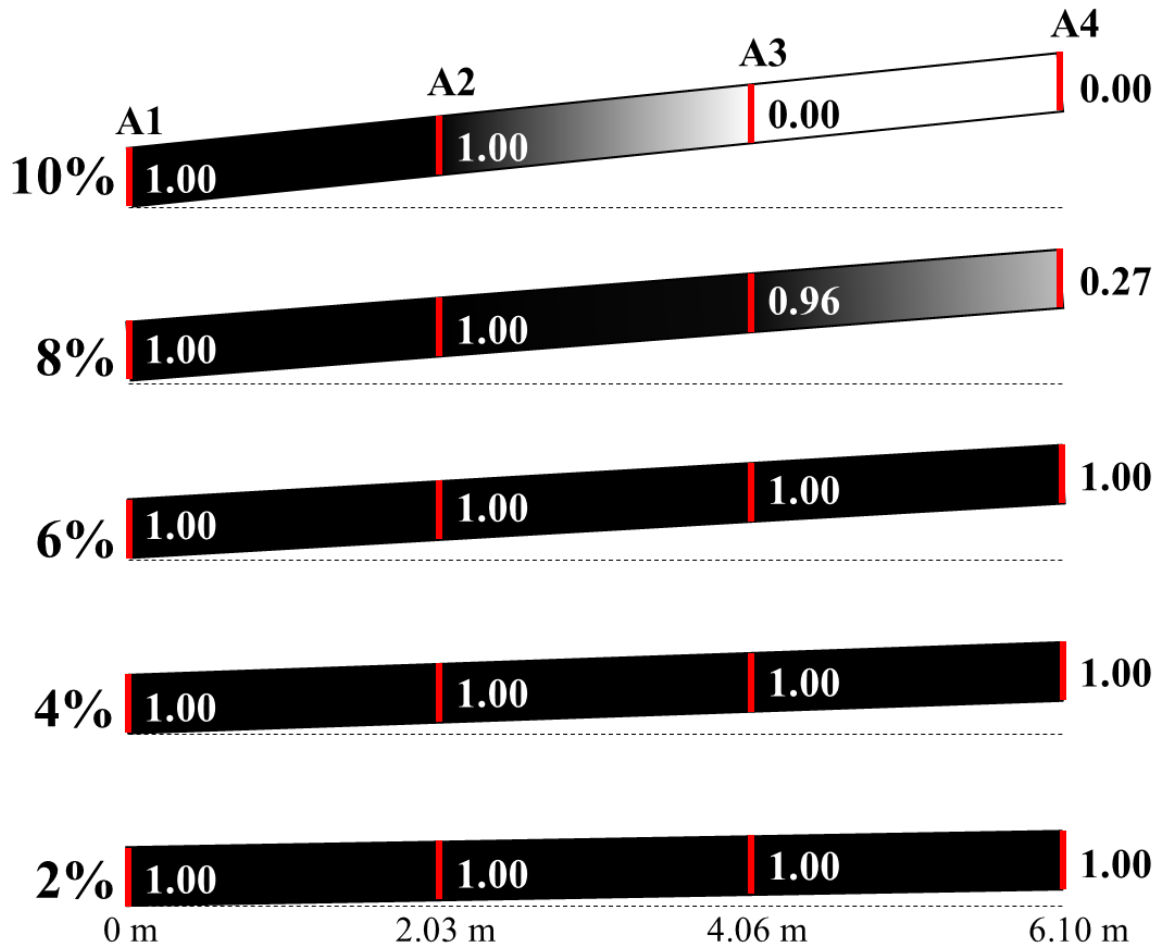


Figure 3-3.— True estimates of the probability that an adult Flathead Chub can successfully ascend to one of the PIT tag antennas (A1 – A4) in the rock ramp fish passage structure at slopes of 2 to 10%. PIT tag antennas were evenly spaced (every 2.03 m) in the fishway to detect fish movements. Dashed lines indicate 0% slope for reference. Each flume diagram is tilted to scale for its respective slope treatment. Darker fill indicates higher probability of passage success. Estimates were determined by multiplying the apparent rates of fish movement between antennas (Appendix 3-1).

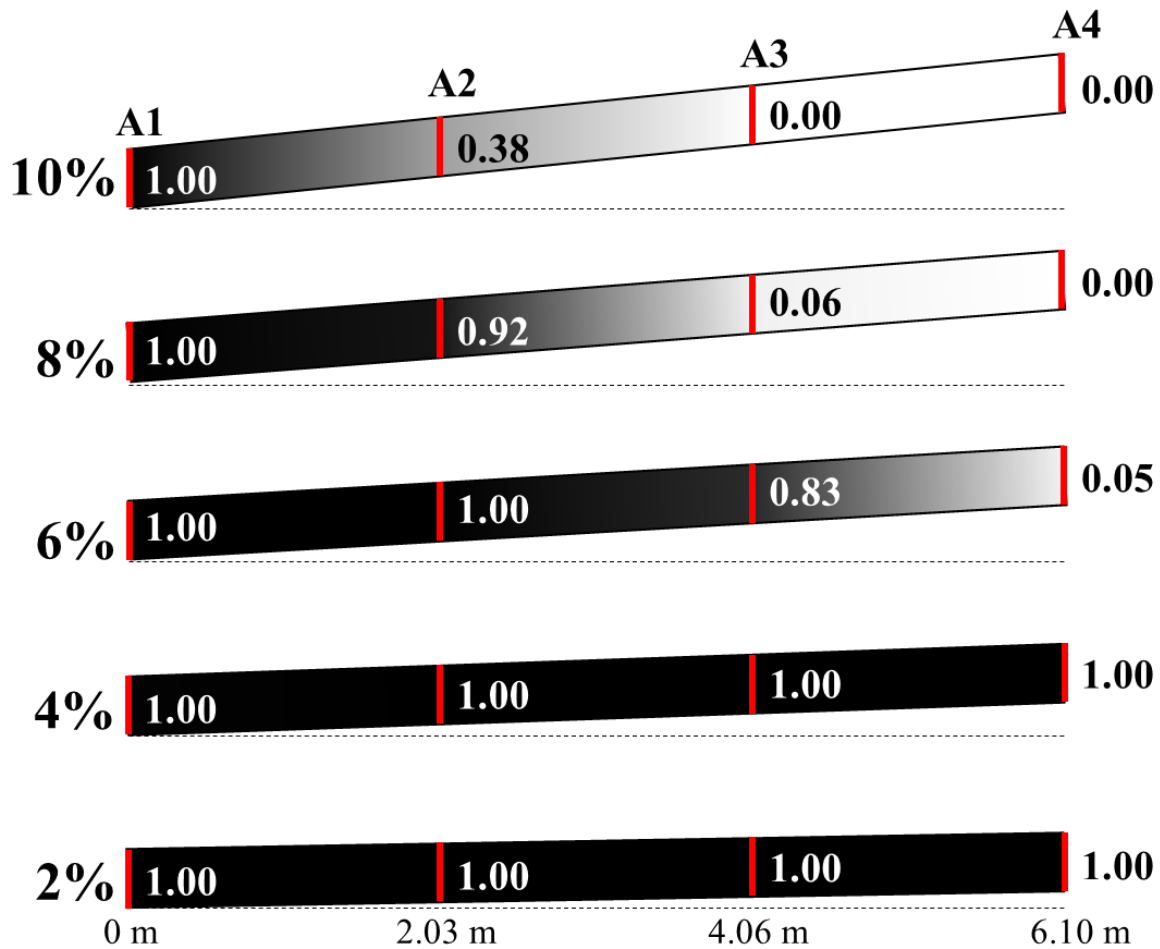


Figure 3-4.— True estimates of the probability that an adult Stonecat can successfully ascend to one of the PIT tag antennas (A1 – A4) in the rock ramp fish passage structure at slopes of 2 to 10%. PIT tag antennas were evenly spaced (every 2.03 m) in the fishway to detect fish movements. Dashed lines indicate 0% slope for reference. Each flume diagram is tilted to scale for its respective slope treatment. Darker fill indicates higher probability of passage success. Estimates were determined by multiplying the apparent rates of fish movement between antennas (Appendix 3-1).

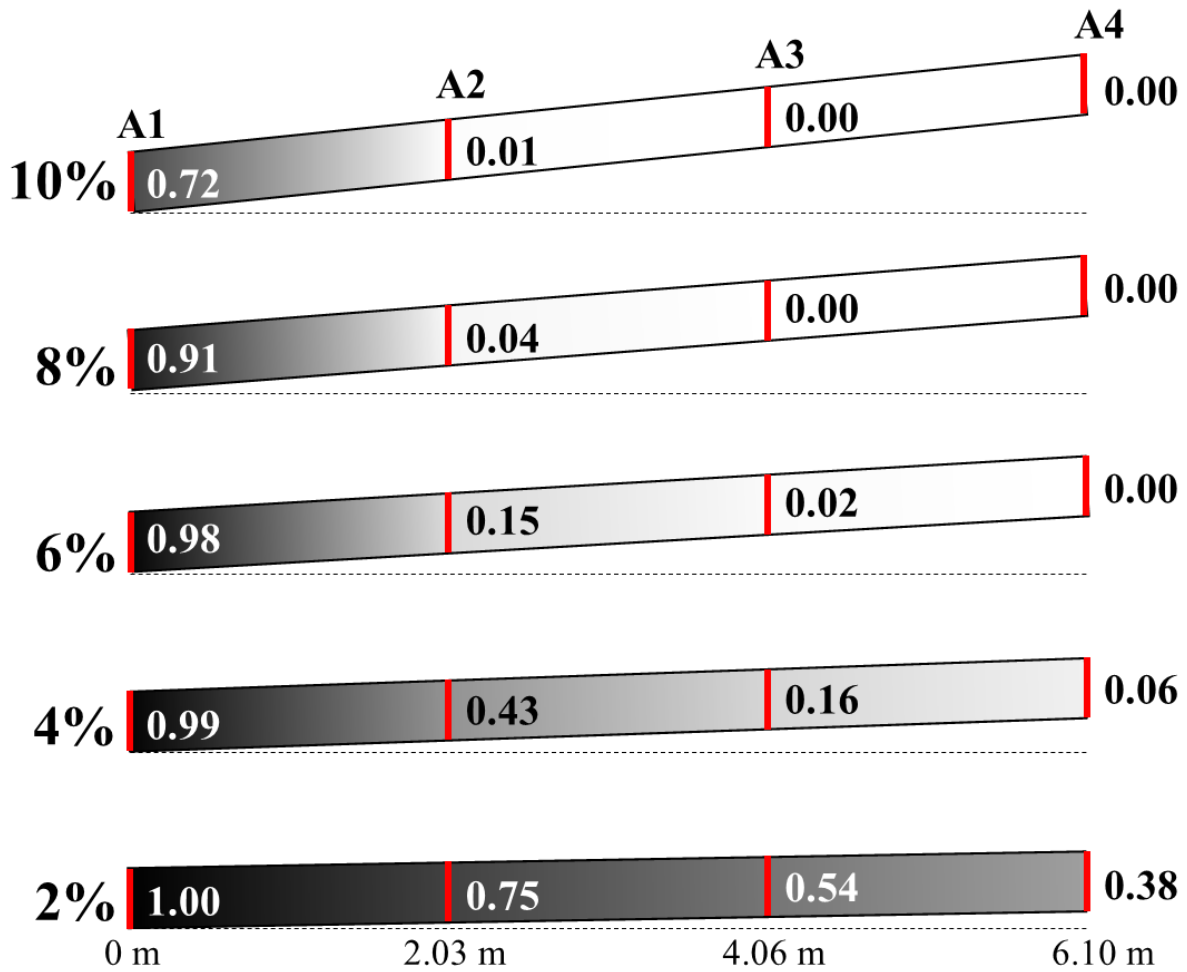


Figure 3-5.— True estimates of the probability that an adult Arkansas Darter can successfully ascend to one of the PIT tag antennas (A1 – A4) in the rock ramp fish passage structure at slopes of 2 to 10%. PIT tag antennas were evenly spaced (every 2.03 m) in the fishway to detect fish movements. Dashed lines indicate 0% slope for reference. Each flume diagram is tilted to scale for its respective slope treatment. Darker fill indicates higher probability of passage success. Estimates were determined by multiplying the apparent rates of fish movement between antennas (Appendix 3-1).

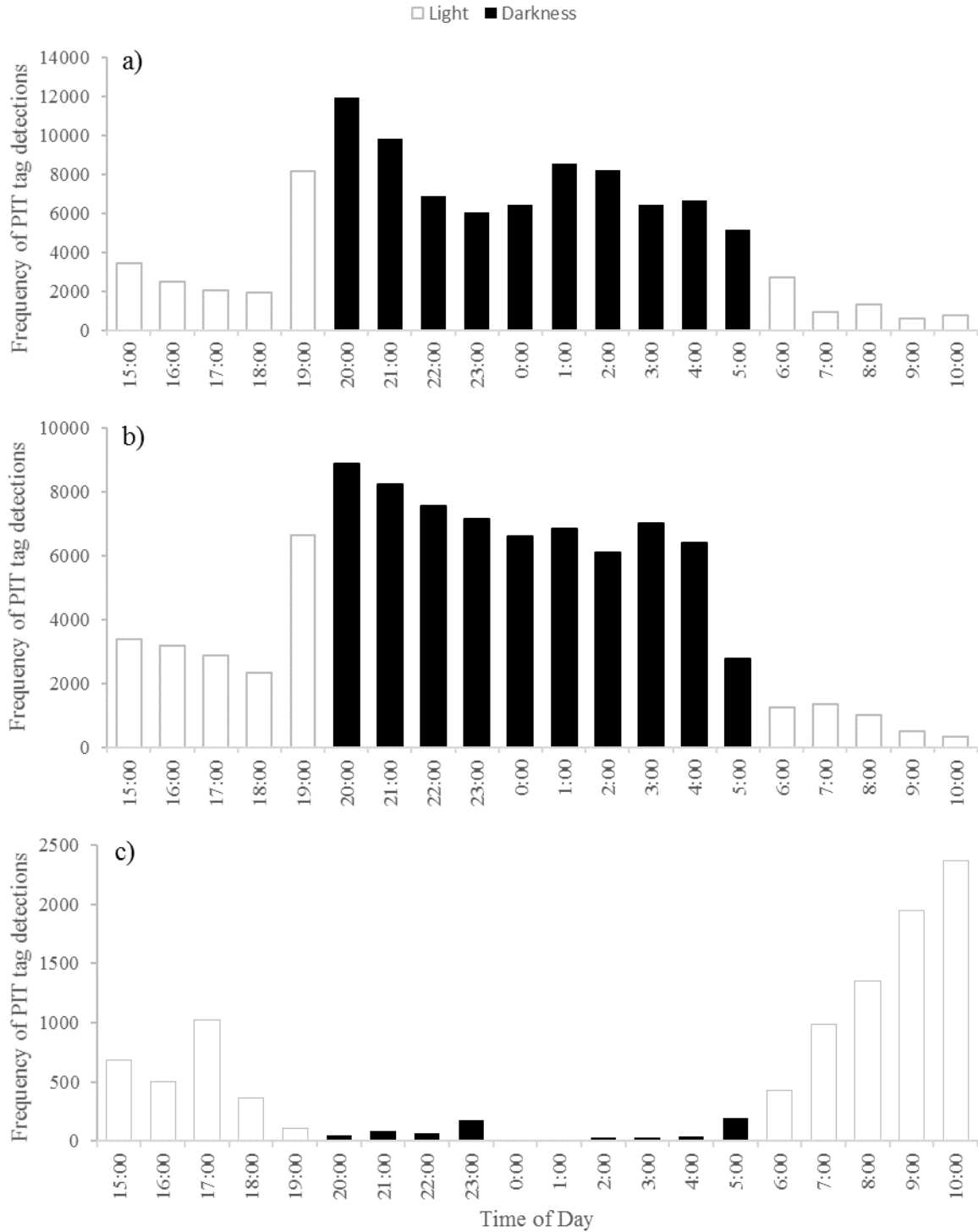


Figure 3-6.— Effects of photoperiod on PIT tag detection frequency (pooled across all antennas; surrogate for fish activity) for trials at 2% slope for Flathead Chub (a), Stonecat (b), and Arkansas Darter (c). Trials were run on a 14L:10D photoperiod, with lights on from 0600 to 2000 h. Trials began at 15:00 and ended at 11:00 the following day.

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APPENDIX

CHAPTER 3 APPENDIX

Appendix 3-1.— Apparent probability of fish movement (ϕ) between two antennas and 95% confidence intervals for Flathead Chub (a), Stonecat (b), and Arkansas Darter (c) when placed in an artificial rock ramp fishway. Movement probabilities are shown at five slope treatments between four evenly spaced PIT tag antennas (A1-A4) in a 6.10-m long fishway.

(a)

Slope	A1		A2		A3		A4	
	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI
10	1.000	1.000-1.000	1.000	1.000-1.000	0.003	0.000-0.029	0.000	0.000-0.002
8	1.000	1.000-1.000	1.000	1.000-1.000	0.958	0.780-0.993	0.283	0.063-0.699
6	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000	1.000	0.983-1.000
4	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000
2	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000

(b)

Slope	A1		A2		A3		A4	
	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI
10	1.000	1.000-1.000	0.376	0.248-0.524	0.001	0.000-0.009	0.000	0.000-0.000
8	1.000	1.000-1.000	0.921	0.818-0.968	0.060	0.019-0.174	0.000	0.000-0.000
6	1.000	1.000-1.000	0.996	0.970-1.000	0.836	0.704-0.917	0.064	0.016-0.222
4	1.000	1.000-1.000	1.000	1.000-1.000	0.998	0.984-1.000	1.000	1.000-1.000
2	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000	1.000	1.000-1.000

Appendix 3-1. (continued)—

(c)

Slope	A1		A2		A3		A4	
	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI	ϕ	95% CI
10	0.718	0.533-0.850	0.012	0.003-0.042	0.010	0.002-0.049	0.009	0.002-0.052
8	0.911	0.818-0.959	0.045	0.018-0.107	0.037	0.011-0.124	0.036	0.009-0.134
6	0.976	0.936-0.991	0.158	0.093-0.256	0.135	0.057-0.289	0.132	0.048-0.315
4	0.994	0.977-0.998	0.430	0.322-0.545	0.386	0.231-0.566	0.379	0.196-0.604
2	0.998	0.992-1.000	0.752	0.611-0.854	0.716	0.549-0.839	0.710	0.498-0.858