American Shad Transport Feasibility Study Report
Cataract Dam – Saco River, Maine

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Whooshh Innovations

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1 Introduction

American shad (*Alosa sapidissima*) historically were an abundant anadromous fish in East Coast rivers. They are a vital component of the ecosystem and the commercial and recreational fisheries industries. Substantial declines in East Coast American shad populations have been attributed to barriers to successful reproduction such as dams, un-optimized fish passage mitigation efforts, pollutants, river and habitat erosion attributed to forestry and farming, and climate change (FWS 2017). As a species, shad are considered by many to be fragile, being sensitive to capture and handling (Olney et al. 2006), with upstream migration more energetically costly for shad than salmon (Leonard et al. 1999). Shad tend to travel in schools and they do not generally leap to ascend waterfalls or to pass through high velocity areas (Haro and Castro-Santo 2010) resulting in passage up typical fish ladders, through baffled fishways and attraction into lifts being only moderately successful (Bunt et al. 2012).

It has been reported that dams impact anadromous fish species in a number of ways from inundating shoreline and spawning areas to changing the flow patterns of rivers, creating reservoirs and still water which can result in a rise in water temperatures with a domino of ecosystem impacts (Harrison 2008, Haro and Castro-Santos 2010). The most obvious impediment, however, is that dams block anadromous fish passage between spawning, juvenile habitats and the ocean. The spawning migrations are fueled by energy stores acquired during ocean residency prior to entering freshwater rivers and streams (Nadeau et al. 2010, Castro-Santos and Letcher 2010). Because anadromous populations have genetic adaptations optimized for migration efficiency for specific river conditions, it is expected that the stresses encountered in travel through man-made passage facilities at dams could have negative implications regarding spawning success (Makrakis et al. 2012, Leonard et al. 1999, Haro and Castro-Santos 1999). Man-made passage impediments often require additional energy expenditures of the fish in comparison to run of the river swim.

The Whooshh Fish Transport System (WFTS) is an innovative fish passage system designed to decrease passage time with the benefit of requiring minimal fish energy expenditure during transport. The WFTS utilizes a novel differential pressure system that facilitates movement of individual fish through a soft, flexible, lightly-misted tube structure in a matter of seconds (Whooshh 2016). The WFTS has successfully demonstrated safe, timely, efficient and effective transport for a number of species including salmonids (Chinook, Atlantic, Pink, Sockeye, and Coho salmon) Steelhead, Rainbow trout, and Sturgeon (Amaral et al. 2016, Buckley 2016, Fryer 2017, Geist et al. 2016, Mesa et al. 2013, Kalama Falls 2014, Summerfelt et al. 2015, Whooshh 2016). Live salmon have been safely transported from 40 ft to 1700 ft with zero to 180 ft in vertical elevation change (Whooshh 2017). In controlled studies, WFTS transported species have exhibited no significant transport-associated concerns. Transport-associated concerns evaluated to date include: injury, descaling, increased stress, reduction in survival or egg viability, delayed migration or passage timing, aberrant homing, increased disease transmission or unusual behavior (Geist et al. 2016, Fryer 2017, Whooshh 2016 & 2016b).
Brookfield Renewable owns and operates one of the largest renewable energy portfolios including holdings in North American, Latin American and Europe. Hydroelectric facilities are a core component of their energy production portfolio (Brookfield Renewable 2017). As a balance must be achieved between hydroelectric energy production and sustaining the environment, the waterways and all that dwell and are supported by them, viable fish passage solutions are paramount to achieving a successful balance. American shad passage solutions to date have proven to have variable success rates (Bunt et al. 2012). The WFTS transport feasibility test provided the opportunity to evaluate safe transport of American shad. This was the first demonstration of the species to be transported through a WFTS tube. The goal of this study was to determine the applicability of the WFTS for American shad upstream fish passage by comparing a group of WFTS transported shad to a group of non-transport shad (control), and evaluating injury and short-term survival of the two groups. The results of this study provide useful insights into consideration of further implementation of the WFTS for American shad at dams on the East Coast and elsewhere.

The primary study objectives include:

- Compare American shad treatment (WFTS transport) and non-treatment (no transport control) for survival within one hour of transport and latent mortality 24 hour post transport.
- Compare American shad treatment (WFTS transport) and non-treatment (no transport control) for fluorescein dye evidence of epithelial injury.

2 Methods

2.1 Experimental Design

The study was conducted on a pilot scale, with a sample size sufficient to provide evidence of feasibility. The study evaluated survival within the first hour post-transport and after 24 hours relative to a non-treatment (no transport) control group. In addition, a small subset of shad were evaluated for epithelial injury. Fluorescein dye has been used as a rapid, sensitive method of detecting epithelial damage (Noga and Udomkusonsri 2002, Colotelo and Cooke 2011). There has been no evidence of WFTS-associated epithelial damage or signs of descaling occurring in the species tested to date (Geist et al. 2016, Whooshh 2016).

The experimental design is shown in Table 1. Briefly summarized, 120 shad were planned to be used in the study, 60 in the WFTS treatment group and 60 in the no transport control group. However, due to a slower than expected American shad run on the Saco River this year, and MDMR permit (Appendix A) restrictions limiting sampling to only 5% of the run at the time of the study, the total sample size was 83 (41 WFTS treatment, 42 control). The study consisted of two days on which shad were transported through the WFTS. On the first day (June 21), 12 shad were transported through the WFTS and 12 controls were collected from the holding tanks and subjected to fluorescein dye treatment.
to assess for evidence of epithelial injury. On the second day (June 22), the remaining shad were transported via the WFTS (29 shad) or held as controls (30 shad).

Control – Non-transported fish. Fish were netted from the fish lift hopper and hand transferred into a circular holding tank, with recirculating pumps and oxygenation, located a short distance from the lower fish lift hopper.

WFTS – Fish transported approximately 175 ft through the WFTS. Fish were netted and hand loaded directly into the WFTS and transported approximately 175 ft to a circular holding tank with recirculating pumps and oxygenation. The entrance to the WFTS was located equidistant from the fish lift hopper as the control holding tank.

**Table 1. Assessment of American shad transport via WFTS Experimental Design Groups**

<table>
<thead>
<tr>
<th>Group</th>
<th>Total Shad (n)</th>
<th>Transport Method</th>
<th>Shad (n) in Safe Transport Survival Assessment</th>
<th>Shad (n) in Epithelial Fluorescein Dye Sub-study</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>42</td>
<td>No Transport</td>
<td>32</td>
<td>10</td>
</tr>
<tr>
<td>WFTS</td>
<td>41</td>
<td>Whooshh tube</td>
<td>31</td>
<td>10</td>
</tr>
</tbody>
</table>

The study shad were sampled (netted and hand transferred) randomly, alternating group assignment (Control vs WFTS) until the total study shad were divided between the control holding tank or WFTS transported to the WFTS holding tank. Once in the holding tanks, fish from both groups were monitored for survival. Survival assessments were made within 1 hour of sampling, and at 24 hours to assess for latent mortality. It is known that shad do not fare well when held in captivity and thus the study time selected was tailored to allow enough time for survival assessment relative to transport with the desire to minimize the potential compounding effect of the additional, new “holding in captivity” variable. Circular tanks were utilized, as they are known to help prevent damage due to collisions with the tank walls and limit stress. Any mortalities were netted out of the holding tanks. Each dead shad were measured (fork length, weight and circumference just anterior to the dorsal fin) and condition assessed (percent descaling and visible injuries noted) and recorded on the study assessment worksheets.

To facilitate establishing the appropriate anesthetizing condition for the epithelial injury fluorescein dye sub-study, a small sample of shad (5 to 10) outside of the study groups were collected from the fish lift hopper and subjected to MS-222 treatment at a 60 mg/L low-end dose to determine the anesthetic induction and recovery times. This assessment was conducted prior collecting the ten shad per group from the holding tanks for the epithelial injury fluorescein dye sub-study. Establishing the induction and recovery parameters for the American shad sampled informed the researchers as to time windows for the various steps in the fluorescein dye treatment procedure.

For the epithelial injury fluorescein dye assessment, ten fish from each group were be gently netted and transferred to a tub containing MS-222 for the mean induction time.
determined in the MS-222 assessment. Once handleable, the shad were gently hand transferred and submerged, using cotton gloved-hands, in a tub containing 0.20 mg/mL fluorescein dye: fluorescein disodium salt in water (Noga and Udomkusonsri 2002, Colotelo and Cooke 2011) for about 2 minutes, as determined from the MS-222 recovery time, followed by 2 rinse steps (60-120 seconds) in fresh water. The dyed shad were euthanized and transferred to a cooler, top closed to prevent photobleaching. The cooler was then transported to the dark room tent and dyed shad photographed under UV and white light, right side view, followed by left side view. The photographed, fluorescein dyed, euthanized shad were subsequently disposed of.

2.2 Experimental Set-up

American shad migrating up the Saco River volitionally enter the east channel below Cataract dam and swim into the hopper of the lowered fish lift. The hopper was elevated a short distance by Brookfield personnel to enable sampling via netting. The control fish holding tank was positioned on the flat ground adjacent to the hopper lift (red circle on right, Figure 1). The circular holding tank had a 10 ft diameter and contained about 800 gallons of water with a recirculation pump and hose to provide fresh water at a rate of ~40 gal/min and aeration. The WFTS accelerator and blower units and control box (orange box, Figure 1) were positioned equidistant from the hopper to the control holding tank. The WFTS accelerator unit was the fish entry point (Figure 2), connected to a 175 ft Whoosh tube that transported fish into an identical circular holding tank. The WFTS holding tank was located on a platform installed on the upper flume that provided a flat area for the tank (red circle on left, Figure 1). Covers were placed over the holding tanks to shade the tanks and ensure no fish loss due to jumping. The smallest Whoosh tube, the T123, was used. The T123 has been used to transport Sockeye salmon with an average weight of 1.39 kg and range 1.16-2.24 kg (average ~3 lb, range ~2.5-5 lbs). The American shad sampled at Cataract Dam typically fell within the lower end of this size range.

![Figure 1. Location of the control tank (right red circle), WFTS tank (left red circle), WFTS (orange box), and the Whooshh transport tube (long orange arrow) at the Cataract Dam, Saco Maine.](image-url)
Smaller tubs (15-40 gallons) were used for the fluorescein dye substudy. Initially, two tubs were used to establish the American shad response to MS-222 treatment. One tub contained 60 mg/L MS-222 and the other ~40 gallons of water. These tubs were subsequently used in the fluorescein dye treatment along with three additional tubs, one containing 0.20 mg/ml fluorescein dye, one used as a second water rinse and the last containing a lethal dose of MS-222 for euthanasia post fluorescein dye treatment prior to photo-documentation. These five tubs were repurposed immediately after completion of the fluorescein dye treatment of the 10-control shad for the fluorescein dye treatment of the WFTS transported shad.

The set-up minimized handling in terms of the distance the netted fish were walked either to the holding tank (controls) or the WFTS accelerator unit and those two paths were designed, as much as possible, to be of equal distance. Once at the holding tank or accelerator, the netted fish were hand transferred out of the net via cotton-gloved hands to maintain comparable handling for each group and to minimize the potential for handling-induced damage. Control fish were hand transferred into the holding tank and WFTS transport fish were hand loaded head first into the WFTS accelerator.

The principle behind the WFTS transport is illustrated in Figure 3. Differential pressure is generated in front of and behind the fish, with less than a 1 PSI differential across the fish. This pressure differential enables gentle, rapid, continuous passage of the fish through the near fiction-less tube.
A dark room tent was set up onsite and consisted of a prep table, UV florescent light, white light, digital camera with appropriate filters and transport coolers for the fluorescent and light photography of the shad epithelial injury sub-study.

2.2.1 The Fish

Migrating wild American shad captured at the Cataract Dam on the Saco River in Maine were used in this study. The WFTS T123 requires a minimum fish size to ensure an effective tube/fish seal is created enabling differential pressure propulsion, so shad that appeared by visual inspection, to be smaller than normal were excluded from the study. The water temperature of the Saco River were recorded in addition to the water in the holding tanks and tubs. Through exchanges of water in the tubs and pumped and circulating water flow in the tanks, the temperature in the tubs and tanks was maintained at a temperature similar to the Saco River, therefore providing a comfortable environment and minimizing the potential of stress induced by thermal shock. As shad are considered to be sensitive to capture and handling (Olney et al. 2006), all efforts to minimize handling were attempted. The shad were directly handled with cotton-gloved hands to ensure good hold and to minimize epithelial damage. The fluorescein dye treated shad were euthanized prior to photographing. All other surviving shad were returned to the river.

2.2.2 Sampling and Assessment Recording

Sampling was random, dictated by arbitrary netting of shad trapped in the fish lift hopper. Efforts were made to minimize harm to the shad in the netting and transfer process. The netted shad were walked alternating to either the control holding tank or the WFTS accelerator, and counted at the transfer. The expectation was 60 shad would be allotted to the control holding tank and 60 shad would be WFTS transported; however, numbers
were lower as described above based on the slower than expected shad run this year on the Saco River.

After 24 hours, all surviving shad were netted, hand held with cotton-gloved hands, quickly measured (forklength) and condition (descale and visible injury) recorded on the Mortality, Measurement and Condition Worksheets. Shad that did not survive were netted out of the holding tanks and measured (forklength, circumference and weight) and condition assessment (descaling, visible injury) recorded on the Mortality, Measurement and Condition Worksheet along with date and hour of mortality observation.

2.2.3 Anesthesia and Fluorescein dye

The recommended dose for the moderate anesthesia of salmonids by the manufacturer of MS-222 (Western Chemical Inc.) is 50-60 mg/L MS-222. It was recommended that the low 50 mg/L dose be tried first before increasing the dose in the dose timing assessment and sub-study for the sedation of the fluorescent dye treatment fish. The MS-222 treatment time to sedation induction, which is analogous to the stage at which the shad are handleable, was recorded on the worksheet. Shad were deemed handleable when they reach a state equivalent to stage 3-4 as previously described (Summerfelt 1990, Bowker 2015). Essentially, handleable relates to loss of equilibrium loss of responsiveness to external stimuli, in which fish are easily caught by hand and do not struggle when held. Once the pre-study shad reach sedation induction they were hand transferred to a fresh water tub and time to recovery from sedation was recorded. Recovery relates to regained equilibrium, resumed normal swim behavior, evading attempts of capture and when fish swim to avoid obstacles. Ideally, MS-222 treatment should achieve induction in ~2 minutes and recovery in ~5 minutes. The expected range for induction and recovery are 0-5 minutes and 4-12 minutes, respectively.

Fluorescein dye is effective at highlighting epithelial damage through green light emission when placed under ultraviolet (UV) light. Fluorescein penetrates the damaged epithelium. In a portable dark room tent setting with UV light excitation, fluorescein emits epithelial injury-associated green light, and this was photographed. The damaged area was subsequently measured using computer software as described by Noga and Udomkusonsri (2002), and Colotelo and Cooke (2011). The image software, ImageJ (http://rsb.info.nih.gov/ij/; National Institute of Health, Bethesda, MD), traces the areas of green and measures the number of pixels which can then be related to the total number of pixels that encompass the image of the fish providing a normalized outcome measure, a proportion of epithelial damage relative to the entire fish.

The results of the sedation induction and recovery tests informed the fluorescein dye treatment sub-study design, which determined that a dose of 60 mg/L of MS-222 sedated shad in about 3 minutes, and took them about 3 minutes to recover. The anesthetized shad were placed in a tub containing 0.20 mg/ml Fluorescein disodium salt in water for ~2 minutes. The dyed fish were rinsed via hand assisted “swim” in a tub with fresh water for ~60 seconds and repeated in a second rinse tub for an additional ~60-120 seconds. At this point, the shad began to show signs of recovery. Due to the site location set-up and desire to minimize handling, the dye and rinse procedure was set up and conducted next to the holding tanks. After rinsing, the shad were euthanized (200 mg/L of MS-222) and placed in a cooler with the top closed to reduce light preventing potential photobleaching. The cooler was hand carried to the darkroom tent setup where the shad
were photographed right followed by left side under UV and right followed by left side under white light. Image analysis was subsequently conducted at an offsite laboratory.

### 3 Results

#### 3.1 American Shad Transport Results

As described above, a total of 83 American shad were used for this study, which was approximately 5 percent of the total population captured at the Cataract Dam at the time of the study. Forty-two of these were held as controls for at least 24 hours, and 41 were transported through the WFTS and held in a separate tank for at least 24 hours.

Water temperatures in the Saco River and in the holding tanks ranged between 22°C and 23.5°C during the study conducted between June 21 and June 23, 2017.

On June 21, 24 shad were taken from the fish lift hopper and used for the study. Twelve were transported through the WFTS and 12 were held as controls. All 12 WFTS survived immediate transport through the WFTS. In addition, all 12 WFTS shad and all 12 control shad survived in the holding tanks overnight. On the following morning (June 22), 10 WFTS shad and 10 control shad were used for the fluorescein dye study (results in subsection below). On June 22, an additional 29 shad were transported through the WFTS, and an additional 30 shad were placed in the control holding tanks.

The WFTS used in the study was a temporary demo system installation put in place just for the June 2017 test days. As such there was no appreciable opportunity to pre-run the system to “de-bug” any installation issues and/or provide the opportunity to optimize the system for shad specific transport. Fish enter the WFTS and slide into the T123 accelerator unit where the differential pressure across the fish is established and the fish is pushed through the tube. The system is lubricated with water, misting at the entry point and at portions through the system to ensure smooth passage. During study operation on day two, there was an intermittent issue in the WFTS, where a few shad did not slide completely through the accelerator unit, the component of the WFTS system located just before the shad would enter the WFTS tube. It was determined that there was an issue with the misting spray nozzles which provide water to the system. They were not providing an adequate amount of water. Due to this issue, two shad did not slide through the accelerator into the tube in the typical manner and were injured. The injuries plus possible associated stress were such that upon reaching the WFTS holding tanks after transport, those two fish were found to be dead. One other shad, which was smaller than most used in the study but not excluded from the study, entered the WFTS tube but then failed normal transport. It stopped in the WFTS tube for several minutes before it completed transport of the entire length of the tube and exited into the WFTS holding tank. It is assumed that this shad experienced substantial stress during this delayed transport as once it entered the WFTS holding tank it died several minutes later.

All other WFTS transported shad survived immediate transport. Although there were covers for the tanks, during transport the tanks were not covered completely as there was a need to provide a fish access/entry point in to the tank. The result of this temporary partial cover was that one WFTS transported shad did manage to jump out of the tank just after transport. It landed on the ground and was hand lifted back into the tank.
tank. On the following day, one WFTS shad was found dead within the 24 hour holding time. It is suspected that this shad was the one observed to have skipped out of the tank the day before when transported. All other 37 WFTS shad survived at least 24 hours after transport before being transferred alive to a stocking truck and released in the river upstream of the dam. In addition, all 42 control shad survived in the control holding tanks for at least 24 hours before being released alive into the river downstream of the dam. Release locations were dictated by the proximity of the shad in the holding tanks to the standard Saco fish passage process equipment (i.e., haul truck and fish lift).

Measurements were taken of fish before they were released. The average fork length for the control group was 47.1 cm, and the average fork length for the WFTS group was 47.7 cm. Circumferences and weights were not taken for all shad, as it was anticipated that this would have caused additional stress to the fish before being released back to the river.

Overall, there were no mortalities in the control, non-transported, group and four mortalities considered to be operationally-related mortalities in the WFTS group verses true WFTS transport-related mortalities. Without excluding these operationally-related mortalities, 90.2 percent survival was observed for the WFTS transported shad group after 24 hours, and 100 percent for the control shad group after 24 hours (Table 2). However, the four WFTS group shad mortalities were associated with correctable mechanical/operational difficulties that would be quickly correctable in a permanent installation, 100 percent of the WFTS transport shad that did not succumb to technical difficulties survived immediate transport as well. In a permanent installation, the shad would not be held post-transport in a tank. They would exit the WFTS into the river upstream of a barrier or into a haul truck. There would not be a risk associated with fish being contained in a tank. In this study however, holding the fish for 24 hr assessment was required and one mortality was observed. Therefore, although the one case of delayed mortality was likely due to study set-up associated issues, tank containment and containment stress, 97.4% of the Whooshh group survived for at least 24 hours after successful transport.

General observations on the condition of the shad once captured from the fish lift and before used in the study were made. It was noted that most shad appeared in good condition, with no signs of major descaling. The shad also appeared in good condition in the tanks after transport and in the holding tanks. On the last day (June 23) researchers observed some scales on the bottom of both the control tank and the WFTS tank, along with shad eggs. This indicates that both groups had some stress associated with being held in tanks overnight and manifest via shedding some scales and releasing some eggs. However, since this was observed in both tanks, this stress is not associated with WFTS transport, and would not be expected to be observed in a permanent installation where shad would be released directly upstream of a dam or into a stocking truck for release upstream soon after transport.
Table 2. Survival Results of American Shad Transport via WFTS and Control

<table>
<thead>
<tr>
<th>Group</th>
<th>Total Shad</th>
<th>Day 1 Total</th>
<th>Day 1 - Immediate Survival</th>
<th>Day 1 - 24hr Survival</th>
<th>Day 2 Total</th>
<th>Day 2 - Immediate Survival</th>
<th>Day 2 - 24hr Survival</th>
<th>Total Shad Immediate Survival</th>
<th>Total Shad 24hr Survival</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>42</td>
<td>12</td>
<td>12 (100%)</td>
<td>12 (100%)</td>
<td>30</td>
<td>30 (100%)</td>
<td>30 (100%)</td>
<td>42 (100%)</td>
<td>42 (100%)</td>
</tr>
<tr>
<td>WFTS</td>
<td>41</td>
<td>12</td>
<td>12 (100%)</td>
<td>12 (100%)</td>
<td>29</td>
<td>26* (89.7%)</td>
<td>25** (86.2%)</td>
<td>38* (92.7%)</td>
<td>37** (90.2%)</td>
</tr>
</tbody>
</table>

* Operational issues identified that prevented typical transport, correctable in permanent installation.

** One shad skipped out of tank after exiting the WFTS. Shad containment required of the study but would not be an issue as not a part of a permanent installation.

3.2 Fluorescein Dye Injury Assessment Results

Overall, the fluorescein dye study revealed that there was little difference in epithelial damage between the WFTS and control groups and the level detected relative to the entire area of the fish was low. In both groups, the observed epithelial damage was consistent with injuries due to normal handling, no WFTS transport-associated epithelial injury was noted.

On day 2 of the study 10 shad from the control group were mildly sedated, treated with fluorescein, rinsed in water to remove excess dye and euthanized prior to photo-documentation at a location adjacent to the control holding tank. The equipment for this procedure was then moved to be adjacent to the WFTS holding tank and 10 shad of the WFTS transported group were subjected to the same process. The euthanized shad were held in a covered cooler to prevent photobleaching while the fish were individually photographed. The overall process took approximately five hrs with the first control shad being photographed within 54 minutes of fluorescein treatment and the first WFTS shad being photographed within approximately 2.5 hours of fluorescein treatment. Complete photo documentation of an individual fish took approximately 20 minutes.

The fluorescent images were evaluated using the Image J software. After initial analysis wherein the same color analysis parameters were used for each group (control and WFTS) in Image J, it was determined that the fluorescent intensity of the shad in the two groups was substantially different. The control group were more intense and thus using those parameters, the total area of the WFTS shad, was not being accurately detected and evaluated. Thus, it was determined that different parameters would be needed in order to completely assess the entire surface area of the shad and to accurately represent the dyed areas in each group. This approach was necessary for two reasons:

1.) To account for the less intense fluorescence of the WFTS group; presumed to be caused by a greater amount of time between dye treatment and photographs relative to the control group.

2.) To account for the areas of non-specific dying observed in the control group and not the WFTS group.
When the same color analysis parameters for each group were used in Image J it appeared that the WFTS group had much less dyed area compared to the control group. However, upon further review it appeared that these parameters did not capture all areas of visible dye on the WFTS group causing the amount of epithelial damage to be underestimated. There was a longer amount of time between dye treatment and photography for the WFTS group and the control group; during this period of time the intensity of the dye fluorescence is presumed to have declined relative to the control group, since the same process was used for dyeing both groups. The only procedural difference between the two groups was the time between being dyed and photographed, verifying that the dye intensity declined over time.

The parameters used on the control fish were set to select dye areas consistent with injuries while excluding non-specific dying; areas of residual dye that are not consistent with an injury and would cause the amount of epithelial damage to be overestimated. The parameters that exclude the non-specific areas of dye on the control group also excluded actual dyed areas on the less intensely dyed WFTS group.

By setting the parameters such that the total area assessment represented approximately the total surface area of the fish, it allowed for a relative means of normalizing the parameter settings for the groups and enabling evaluation of potential injury across the whole fish. When the different color analysis parameters were used, the location and extent of dye areas were consistent for both groups. Both groups showed the prominent areas of epithelial damage occurred around the mouth, eyes, fin edges, and at the base of the tail. These areas were also observed as the common areas of scales loss for both groups of fish in the photographs taken under white light (no dye analysis).

The control fish had an average of 8.6% dyed. The WFTS group had an average dyed area of 11.4%; however, the removal of one potential outlier\(^1\) (W1) brings the average dyed area for the WFTS group to 8.3%, which shows no difference from the control group. Given the setting used for the WFTS group, W1 results fell within true outlier criteria for both dye area sides. Tables 3 and 4 show the results from the fluorescein dye assessment for the control group and WFTS group, respectively. Figures 4 and 5 show examples of the photographs taken for the fluorescein dye study for a control and a WFTS shad, respectively.

These results suggest that there is not a clear difference in surficial damage between the WFTS group and the control group. Areas of dye outlined in these images are consistent with injuries from normal handling; as there were no obvious injuries specifically attributable to Whoosh transfer methods.

It should be noted that although there was handling of the shad required in the sampling and evaluation of the fish in this study, an optimized permanent WFTS installation would potentially support volitional entry with limited or no handling required. This pilot feasibility test set a high bar comparing WFTS transport survival to no transport survival with limited handling in each case.

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\(^1\) Calculated IQR values for this dataset identify any value above 20.6% as an outlier
### Table 3. Fluorescein Dye Result for the Control Group

<table>
<thead>
<tr>
<th>Fish</th>
<th>Area Total</th>
<th>Dye Area Right</th>
<th>Dye Area Left</th>
<th>Total Dye Area</th>
<th>% Dye</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>680</td>
<td>20.1</td>
<td>32.4</td>
<td>52.5</td>
<td>7.7</td>
</tr>
<tr>
<td>C2</td>
<td>574</td>
<td>41.3</td>
<td>18.1</td>
<td>59.5</td>
<td>10.4</td>
</tr>
<tr>
<td>C3</td>
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<td>92.1</td>
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</tr>
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<td>49.3</td>
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<td>11.9</td>
<td>21.6</td>
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</table>

### Table 4. Fluorescein Dye Result for the WFTS Group

<table>
<thead>
<tr>
<th>Fish</th>
<th>Area Total</th>
<th>Dye Area Right</th>
<th>Dye Area Left</th>
<th>Total Dye Area</th>
<th>% Dye</th>
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<tbody>
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<td>W1</td>
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<tr>
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</table>
Summary

The results of this pilot study are encouraging as they readily exemplify the feasibility of use of the WFTS for transporting American shad. Excluding the few shad that were injured due to correctable operational/mechanical difficulties that occurred during this study, immediate survival post-WFTS transport was 100% and 24hr survival of shad was 97.4%. Given that shad are known to be highly sensitive to handling and stress, and that traditional transport methods such as trap and haul or fish ladders have notable associated mortality rates and fish passage delay challenges, the survival rates achieved
in this small WFTS pilot feasibility test are quite positive. The results of the fluorescein dye injury assessment conducted on a subset of shad in this study are also highly encouraging as they indicate that WFTS transport does not physically injure American shad.

These results indicate that the WFTS can safely transport American shad, a species traditionally known as sensitive to handling and difficult to pass upstream. Although not included as components in this study, Whooshh has applied mechanisms for attracting fish and has developed a means of scanning and sorting fish prior to entry into the WFTS system that can be applied to attract shad to the device, to sorting them for appropriate size for transport, to distinguish them from other species and to transport them upstream or to a holding facility. Whooshh plans to pursue American shad passage via the WFTS system and further optimize the use of this technology at sites on the East Coast and other potential locations to help accomplish safe, timely, and effective American shad passage.

5 Literature Cited


Geist, DR, Colotelo, AH, Linley, TJ, Wagner, KA, and Miracle, AL. (2016) Physical, physiological, and reproductive effects on adult fall Chinook Salmon due to passage through a novel fish transport system. Journal of Fish and Wildlife Management In-Press. doi: http://dx.doi.org/10.3996/102015-JFWM-108


Appendix A. MDMR Study Permit
May 30, 2017
SPECIAL LICENSE NUMBER ME 2017-74-04

Acting under the authority vested in the Commissioner of Marine Resources by virtue of 12 M.R.S. §6074(8)(A), I hereby issue subject to renewal a Special License to GAIL WIPPELHAUSER representing the Maine Department of Marine Resources and MICHAEL SEARS representing HDR, Inc. This Special License exempts said for the purpose of evaluating American shad passage through the Whooshsh Fish Transport System (WFTS). Exemptions provided are from those portions of Department of Marine Resources (DMR) laws 12 M.R.S §6502-A relating to commercial pelagic and anadromous fishing license, and regulations Chapter 44.02 pertaining to limits on American shad, Chapter 8.20(Q) relating to harvest landings of pelagic and anadromous fish, Chapter 55.03 pertaining to special area fishing closures, Chapter 55.69 relating to fishing in the Saco River, and Chapter 80 pertaining to the commercial pelagic and anadromous fishing license. This Special License is issued subject to the following conditions:

1. **Who:** DMR employee: Gail Wippehauzer HDR, Inc: Michael Sears
2. **What/How:** A new system for gentle transport of fish, the Whooshsh Fish Transport System (WFTS), will be evaluated in a feasibility study at Cataract Dam, located on the Saco River in Maine. American shad will voluntarily enter the Cataract East Dam fish passage channel to the lift system. Once in the lift hopper, shad will be netted out and hand fed directly into the WFTS. Transport of American shad (Alosa sapidissima) 200ft through a WFTS tube to a holding tank will be compared to non-transported shad (control) held in a comparable holding tank. The state of the fish will be evaluated in terms of survival approximately one hour post sampling +/- transport, and latent mortality 24 and 48 hours post sampling. A subset of each group (treatment vs. control) will be treated with fluorescein dye to enable detection and photographic evidence of epithelial damage. The ease of use and evidence of successful, safe, timely, efficient and effective transport will be noted.
3. **Where:** Cataract Dam on the Saco River, Maine
4. **When:** date of issuance-June 30, 2017
5. **Vessels:** no fishing vessels will be used.
6. **Conditions:** Marine Patrol Division I office (west) of Port Clyde, Tel: 633-6595 shall be contacted prior to the start up of collecting activities to make arrangements as to the necessary frequency when to contact Marine Patrol to provide the Special License (SL) number, dates, location(s) of activities, name of special license holder, name(s) of person(s) collecting in the field, and if transporting of specimens will occur, etc. A log of your contact with Marine Patrol is advised. Failure to contact Marine Patrol shall be grounds for the immediate revocation of this Special License.
   - **Quantities** of regulated organisms limited to 60 in the experimental treatment and 60 in the control group, not to exceed 200 fish.
   - **Specimens that expire** must be properly disposed of by means other than overboard discharge into Maine marine waters.
   - **Reporting:** A report (electronic format) on the specimens collected/released/held (numbers per species/collection location/dates) and any accomplishments/results shall be provided to the Department prior to renewal. The report shall be sent to dmr.rulemaking@maine.gov.
   - **Penalty:** Any infraction of these conditions or any violation of any Marine Resources laws shall be grounds for the immediate revocation of this Special License. Pursuant to 12 M.R.S.A. §6074(9) an individual who fails to comply with the conditions or limitations on the licensed activity under this section commits a civil violation for which a fine of not less than $100 nor more than $500 may be adjudged.
   - **Additional** conditions may be added at the discretion of the Commissioner.

This Special License expires December 31, 2017 and has four renewals.

Deirdre Gilbert
For Commissioner Patrick C. Keliher

cc: Marine Patrol Division I