

EFFECTS OF ACCLIMATION ON POSTSTOCKING DISPERSAL
OF AGE-1 PALLID STURGEON

by

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ABSTRACT

A propagation program for pallid sturgeon *Scaphirhynchus albus* in the upper Missouri River was implemented by the U. S. Fish and Wildlife Service in 1997. Preliminary research indicated that many hatchery-reared pallid sturgeon were experiencing significant downstream poststocking dispersal, negatively affecting their recruitment. Therefore, the objective of this study was to evaluate the effects of acclimation to flow and site-specific physicochemical water conditions on poststocking dispersal and physiological condition of age-1 pallid sturgeon. Fish from three acclimation treatments were radio-tagged, released at two locations (Missouri River and Marias River), and monitored using passive remote telemetry stations. Marias treatment was acclimated to flow and site-specific physicochemical conditions in tanks along the Marias River. Bozeman treatment was acclimated to flow in tanks at the Bozeman Fish Technology Center (BFTC), and traditional treatment had no acclimation (reared at the BFTC under traditional protocol). During both years fish released in the Missouri River dispersed less than fish released in the Marias River. In 2005, Marias treatment dispersed less and nearly twice as many fish remained in the Missouri River reach than traditional treatment. In 2006, pallid sturgeon dispersed similarly among treatments and fish remaining in the Missouri River reach were similar among all treatments. Differences in poststocking dispersal between years may be related to fin curl. Fin curl was present in all fish in 2005 and 27% of the fish in 2006. Pallid sturgeon from all treatments in both years had a greater affinity for the lower reaches of the Missouri River than the upper reaches. This area consists of more fines and sand substrate as opposed to gravel and cobble in the upper reaches. Thus, habitat at release site influenced poststocking dispersal more than acclimation treatment. No difference was observed in relative growth rate among treatments in 2006. However, acclimation to flow (i.e., exercise conditioning) may reduce liver fat content. Acclimation conditions used in this study may not benefit pallid sturgeon unless fin curl or fatty liver are present. It is evident from this study that natural resource agencies need to consider stocking location carefully to reduce poststocking dispersal.

INTRODUCTION

Pallid sturgeon *Scaphirhynchus albus* are native to the Missouri and Mississippi river drainages. Historically, these rivers provided a diverse array of environments (e.g., islands, alluvial bars, secondary channels, backwaters) that were in a constant state of fluctuation (Hesse and Sheets 1993; CMRES 2002). Anthropogenic alterations (e.g., impoundments and channelization) of the Missouri River in the past century have altered many natural river functions such as separation of the floodplain from the channel, loss of the natural hydrograph, reduced sediment and organic matter transport, altered temperature regimes, and removal of instream cover (Hesse and Sheets 1993). About 51% of the historical range of pallid sturgeon has been channelized, 28% has been impounded, and the remaining 21% is below dams where habitat variables (e.g., temperature, turbidity, discharge) have been altered (Keenlyne 1989). Additionally, contaminants that may negatively affect reproduction have been documented within the tissue of pallid sturgeon (Ruelle and Keenlyne 1993). These anthropogenic factors have likely caused the reduction in the range of pallid sturgeon. Pallid sturgeon were listed as an endangered species in 1990 (Dryer and Sandvol 1993).

No evidence of successful reproduction and recruitment of pallid sturgeon has been observed in the Missouri River above Fort Peck Reservoir (hereafter called the upper Missouri River) in the past 20 years (Gardner 1997). It was estimated that 50 wild (naturally produced in the river) pallid sturgeon remained in the upper Missouri River in 1997, and it was expected that these would be extirpated within 10-20 years (Gardner

1997). The exact mechanisms for the lack of recruitment in the upper Missouri River are unknown.

A propagation program to increase the number of pallid sturgeon in the upper Missouri River was implemented by the U. S. Fish and Wildlife Service in 1997 because of the lack of recruitment. In 1998, 732 age-1 hatchery-reared pallid sturgeon (1997 year class) from the Gavins Point National Fish Hatchery were released in the upper Missouri River. Subsequently, pallid sturgeon were reared at the U.S. Fish and Wildlife Service Bozeman Fish Technology Center (BFTC) in 2001 and 2003.

Age-1 pallid sturgeon were released at multiple locations in the upper Missouri River system from the Fred Robinson Bridge on the Missouri River [river kilometer (rkm) 3,092] upstream to the confluence of, and within, the Marias River (rkm 3,307). Pallid sturgeon were released in the upper reaches of the upper Missouri River (i.e., in or near the Marias River) based on the hypothesis that if those fish remained and spawned within the upper reaches the resulting larval pallid sturgeon may have enough distance (about 300 km) to develop and settle out of the drift before reaching the headwaters of Fort Peck Reservoir (rkm 3,001). It is hypothesized that the lack of recruitment in the Missouri River may be a function of larval pallid sturgeon drifting into the headwaters of Fort Peck Reservoir and succumbing to mortality. After hatching, larval pallid sturgeon drift downstream for a period of 11-12 days until they have developed enough to maintain position within the river (Kynard et al. 2002, 2007; Braaten et al., in review) which may require between 245 and 530 km of river based on mean water velocities of 0.3 and 0.6 m/s, respectively (Braaten et al., in review). Although pallid sturgeon

spawning habitat has not been defined in the upper Missouri River, it is clear that Fort Peck Dam and resulting Fort Peck Reservoir have been a migration barrier and inundate approximately 200 km of natural lotic habitat.

Of the 732 hatchery-reared pallid sturgeon released from the 1997 year class, 29 were recaptured by age-5 (Gardner 2004). However, only 11 of the 2,221 fish stocked from the 2001 year class were recaptured by age-5 (Gardner 2007). It is likely the 2001 year class dispersed downstream into Fort Peck Reservoir, experienced high mortality, or both. Therefore, a preliminary study was conducted in 2004 to evaluate poststocking dispersal of age-1 pallid sturgeon. Twenty-nine radio-tagged pallid sturgeon were stocked in the Marias River at rkm 93 and their movements were recorded using passive remote telemetry stations. Twenty-six of the twenty-nine fish had dispersed downstream 93 km within the Marias River and entered the Missouri River in 33 d. Additionally, 19 of those fish had passed the lowest remote station at Judith Landing (210 km downstream of release) by the same time. These data indicated rapid downstream poststocking dispersal by hatchery-reared age-1 pallid sturgeon; thus, these fish may be dispersing into Fort Peck Reservoir increasing mortality and subsequently reducing recruitment.

Little information exists regarding poststocking dispersal of hatchery-reared age-1 pallid sturgeon throughout their range. However, juvenile (age-3) pallid sturgeon released in the Missouri River below Fort Randall Dam generally remained near the stocking location or moved upstream during the first year after stocking and showed a general trend of moving upstream in the spring and downstream during the summer of the two subsequent years (Jordan et al. 2006). Moreover, hatchery-reared age-6 and age-7

pallid sturgeon released in the lower Platte River generally dispersed downstream after release; however, these fish did not disperse greater than 65 km downstream from the release site throughout the following year (Snook et al. 2002). These studies demonstrate that the rapid downstream poststocking dispersal observed in the upper Missouri River may not be natural and that hatchery rearing conditions may affect poststocking dispersal. Although some downstream dispersal may be natural in juvenile pallid sturgeon, the rate and magnitude of dispersal may be related to hatchery rearing conditions.

Many differences exist between the hatchery environment and the river environment (e.g., flow, physicochemical conditions, food, habitat), and poststocking dispersal of hatchery-reared pallid sturgeon is likely a function of physical condition and chronic stress related to the transition between these environments. Hatchery-reared pallid sturgeon may not become acclimated to the river environment during their first year post-release (Jordan et al. 2006). Further, survival, growth rate, and condition factors of hatchery-reared white sturgeon *Acipenser transmontanus* suggest that most are not acclimated to natural conditions until 1 – 3 years after release (Ireleand et al. 2002). Hatchery-reared age-1 lake sturgeon *Acipenser fulvescens* (Lake Winnebago origin; mean length = 30 cm) stocked in the Menominee River, Wisconsin, dispersed rapidly downstream as far as 32 km in 24 h after stocking and out of the stretch of river where the investigators wanted the fish to reside (Thuemler 1988). However, most wild caught lake sturgeon (Menominee River origin; mean length = 48 cm) transplanted from lower reaches to an upper reach of the Menominee River remained in the upper reach of the

river and most did not move downstream (Thuemler 1988). These studies further suggest that the hatchery environment influences poststocking dispersal.

Although acclimation experiments have had mixed results (Cresswell and Williams 1983; Minckley et al. 1991; Vidergar et al. 2003), acclimation has positively affected some large river fishes. Exercising fish that were reared in tanks without flow may improve swimming performance and increase survival (Ward and Hilwig 2004). Poststocking dispersal decreased when razorback suckers *Xyrauchen texanus* were preconditioned to flow (Mueller et al. 2003). Further, Swimming performance increased for flannelmouth sucker by 10%, bonytail *Gila elegans* by 15%, razorback sucker by 26%, and spikedace *Meda fulgida* by 40% after exercise conditioning in flowing water (Ward and Hilwig 2004).

Acclimation to site-specific physicochemical conditions may also reduce poststocking dispersal in hatchery-reared pallid sturgeon. Adriatic sturgeon *Acipenser naccarii* that were acclimated to brackish water had greater critical swimming speeds than non-acclimated fish in trials conducted in brackish water (McKenzie et al. 2001). Further, hatchery-reared brown trout *Salmo trutta* acclimated to in-stream water conditions exhibited a more limited poststocking dispersal than non-acclimated fish (Cresswell and Williams 1983).

In addition to increasing swimming performance and reducing poststocking dispersal, acclimation to flow (exercise conditioning) may also increase the physiological condition of pallid sturgeon. The hatchery environment can negatively influence the normal physiological activities of pallid sturgeon and shovelnose sturgeon

Scaphirhynchus platyrhynchus which reduces metabolism and promotes accumulation of fat in the liver (USFWS 2003). Fatty liver is pathologic in fish and accumulation of lipids in the liver causes metabolic disorder of the liver (Zhang et al. 2004). However, lipids are an important source of potential chemical energy and their presence or absence reflects the metabolic capacity of fish (Busacker et al. 1990). Acclimation to flow may reduce liver fat content in hatchery-reared pallid sturgeon while maintaining enough potential chemical energy to aid in the transition from the hatchery to the river environment.

Objective three in the Pallid Sturgeon Recovery Plan states that a need exists for research on methods to improve spawning, culture, and rearing of pallid sturgeon in hatcheries (Dryer and Sandvol 1993). Pre-release environment (e.g., hatcheries) affects poststocking dispersal of some large river fishes (Thuemler 1988; Mueller et al. 2003). Although it has been demonstrated in other fish species that exercise conditioning can increase physiological fitness and reduce stress in subsequent exercise bouts (Barrett and McKeown 1988), increase swimming performance (Ward and Hilwig 2004), and increase survival (Davidson 1997), most reintroduction programs do not test whether acclimation or physical conditioning of stocked fish influences their performance after stocking (Mueller et al. 2003). A meta-analysis on the effects of semi-natural culture techniques on post-release survival of anadromous salmonids recommends that more natural culture techniques should be used in hatcheries (Maynard et al. 1995). Further, it has been suggested that semi-natural rearing methods have value for rearing small lots of fish, such as those cultured in recovery programs (Fuss and Byrne 2002).

The successful use of hatcheries for recovery of pallid sturgeon will depend on how well hatchery-reared pallid sturgeon can acclimate to natural conditions. Fish reared in static water conditions may experience increased stress, downstream displacement, or increased mortality from predation when released into lotic environments (Ward and Hilwig 2004). Acclimation to flow and site-specific physicochemical water conditions may aid pallid sturgeon in adapting more quickly to the river environment. Understanding conditions that better acclimate hatchery-reared pallid sturgeon to the river environment and the duration of exposure needed will be an important tool for managers to reduce downstream poststocking dispersal of pallid sturgeon.

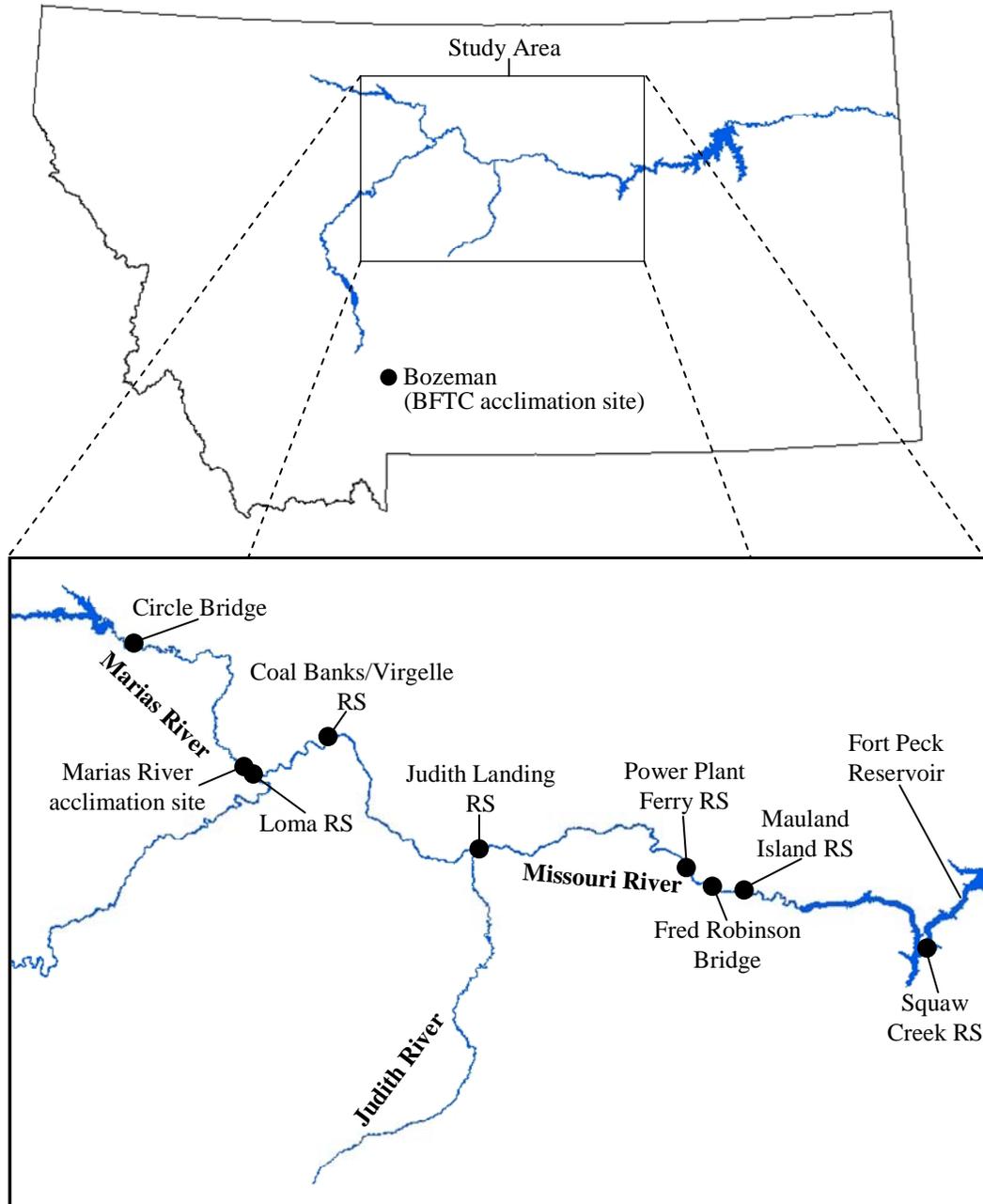
The objectives of this study were: 1) to evaluate the effects of acclimation to flow and site-specific physicochemical conditions on poststocking dispersal, growth (length and weight), energy reserves (crude fat content), and liver fat score of age-1 pallid sturgeon, and 2) to evaluate the effects of stocking location on poststocking dispersal of age-1 pallid sturgeon. I predicted hatchery-reared pallid sturgeon acclimated to flow and physicochemical conditions would have a lower dispersal rate and less downstream dispersal than non-acclimated fish; pallid sturgeon acclimated to flow would have decreased growth rates, fat content, and liver fat scores compared to non-acclimated fish; and that pallid sturgeon released in the Missouri River would have a lower dispersal rate and less downstream dispersal than fish released in the Marias River.

STUDY SITE

The study area on the upper Missouri River was the 306-km reach from the headwaters of Fort Peck Reservoir (rkm 3,001) upstream to the confluence with the Marias River (Figure 1). This reach of river maintains many of the characteristics of a free-flowing river such as islands, alluvial bars, secondary channels, and backwaters (Gerrity 2005). Discharge measurements for the Missouri River were obtained from a U.S. Geological Survey gauging station located at rkm 3,093. Mean monthly discharge from 1935 to 2004 varied from 164 m³/s in September to 501 m³/s in June. Mean daily discharge (\pm SE) in the Missouri River during the study (September through October) was 158.63 ± 0.71 m³/s in 2005, and 151.78 ± 0.79 m³/s in 2006 (USGS 2005).

The Marias River originates south of Cut Bank, Montana, at the confluence of Bank Creek and the Two Medicine River and runs southeast to the confluence with the Missouri River. The Marias River is impounded by Tiber Dam (rkm 127). The study area on the Marias River is the reach from the confluence with the Missouri River upstream to the Circle Bridge (rkm 93). Discharge measurements for the Marias River were obtained from a U.S. Geological Survey gauging station located at rkm 65. Mean monthly discharge of the Marias River from 1959 to 2004 varied from 8 m³/s in January to 58 m³/s in June. Mean daily discharge in the Marias River during the study (September through October) was 11.28 ± 0.09 m³/s in 2005, and 14.75 ± 0.03 m³/s in 2006 (USGS 2005).

Figure 1. Map of the study area with remote stations (RS), release sites (Marias River release: Circle Bridge in 2005 and Marias River acclimation site in 2006, Missouri River release: Fred Robinson Bridge for both years), and the Marias River and Bozeman Fish Technology Center (BFTC) acclimation sites denoted.



Rearing tanks for the treatments used in this study were located along the bank of the Marias River (3 km upstream from the confluence with the Missouri River) and in the containment building at the BFTC (Figure 1). Tanks at the Marias River site were supplied with water from the river. Mean daily temperature of the Marias River water at the tank site varied from 18°C - 25°C throughout the study. Mean daily temperature of the BFTC tank water varied from 18°C - 24°C throughout the study.

METHODS

The 2004 year class of pallid sturgeon from the BFTC was used to evaluate the effects of acclimation treatments (see below) on poststocking dispersal of age-1 pallid sturgeon in 2005. Sixty of the largest pallid sturgeon from the BFTC were selected, measured [mean (\pm SE) length = 330 ± 3 mm; weight = 121 ± 4 g], and tagged with passive integrated transponder (PIT) tags and elastomer tags. Passive integrated transponder tags were inserted below the dorsal fin and elastomer tags were injected in the ventral surface of the rostrum in accordance with the Upper Basin Pallid Sturgeon Propagation Plan (UBPSPC 2005). The PIT tags were used to identify individuals for the remainder of the study. Elastomer tags were not used as an identification method in this study, but will assist Montana Fish, Wildlife and Parks in identifying the year class and stocking location for recaptured individuals that may expel their PIT tags. Pallid sturgeon were randomly assigned to two treatments (see below). Tanks were the experimental unit and each treatment was assigned two tanks containing 15 fish (observational unit) per tank. Pallid sturgeon were reared for 36 d (acclimation period). All individuals were implanted with a radio-transmitter on day 33 of the acclimation period. After implantation, all fish were placed back in tanks and held for 3 d until release. Pallid sturgeon from each tank were randomly assigned one of two stocking locations. Stocking locations were in the Missouri River at the Fred Robinson Bridge and in the Marias River at the Circle Bridge (Figure 1).

The 2005 year class of pallid sturgeon from the BFTC was used to evaluate the effects of acclimation treatments (see below) on poststocking dispersal and physiological

condition of age-1 pallid sturgeon in 2006. Two hundred seventy of the largest pallid sturgeon (length = 302 ± 2 mm; weight = 96 ± 2 g) from the BFTC were selected and randomly assigned to three treatments (see below). Two of the treatments were identical to those in 2005. Tanks were the experimental unit with two tanks per treatment and each tank contained 45 individuals. Acclimation period was 74 d in 2006. Fifteen randomly selected fish from each tank were implanted with a radio-transmitter on day 63 of the acclimation period. After implantation, all fish were placed back in their tanks and held for 11 d until release. Stocking locations were in the Missouri River at the Fred Robinson Bridge and in the Marias River at the acclimation site (rkm 2.7) (Figure 1). Individuals not implanted with radio-transmitters were sampled to assess physiological condition (see below).

Treatments

Marias Treatment

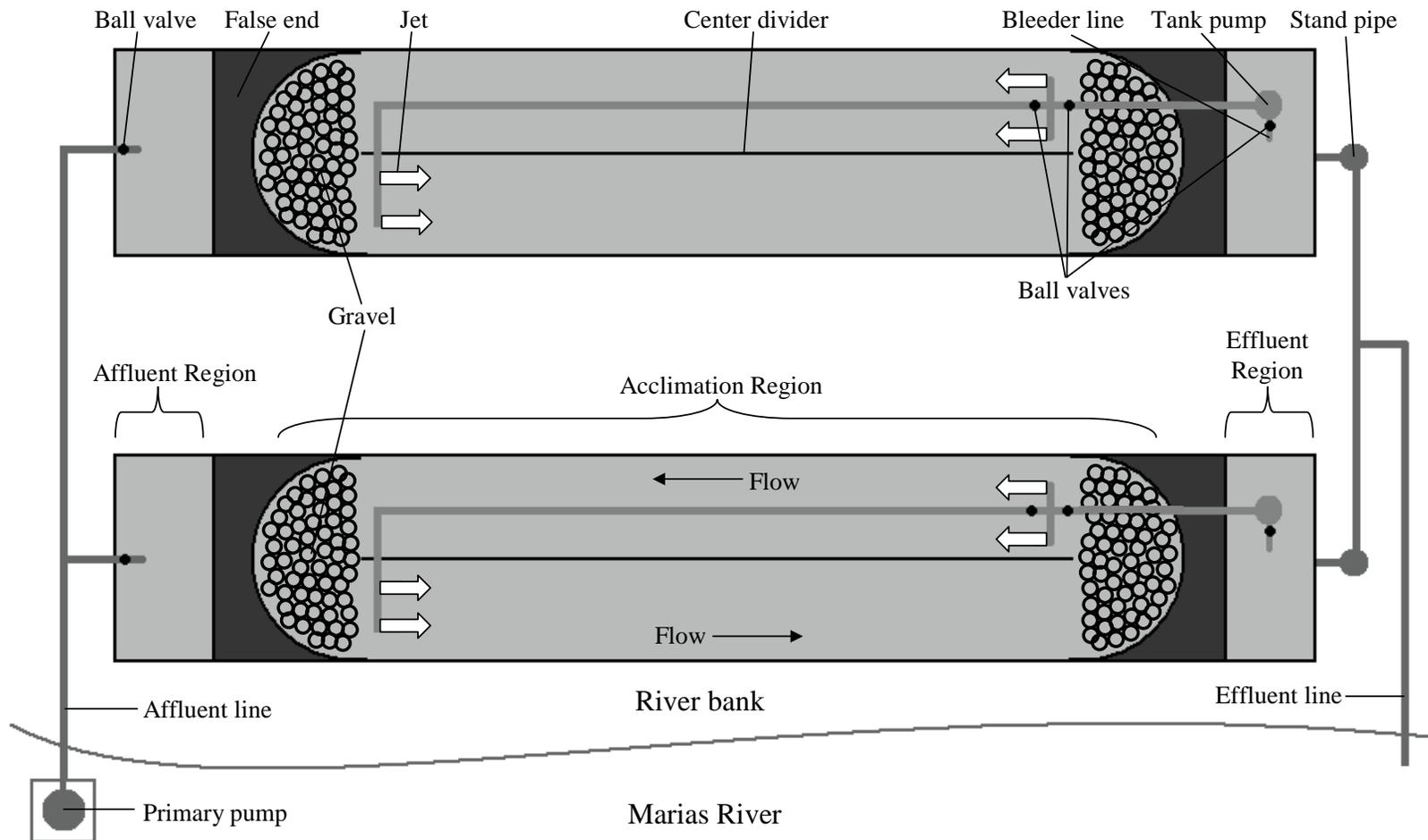
Marias treatment pallid sturgeon were acclimated to flow and site-specific physicochemical water conditions in tanks near the Marias River. The Marias treatment was implemented in both 2005 and 2006.

Tanks were constructed of lumber and lined with Firestone PondGard EPDM Geomembrane. Tanks were 7.3-m long, 1.5-m wide, and had a water depth of 0.5 m. Each tank had semi-circular false ends. The false ends were constructed of lumber and sealed with Sweetwater Epoxy Paint. Fiberglass reinforced plastic (FRP) lined the half-circle. Each half-circle had 1.9-cm washed gravel substrate. The remainder of the tank

had no substrate. The false ends divided the tanks into three regions: affluent, acclimation, and effluent (Figure 2). Water flowed through each region via 6-mm holes drilled in the FRP and 6-mm plastic mesh screening located on top of the FRP panel. The false ends were centered in the tank with their apices 4.6 m apart. A 3.0-m center divider constructed of FRP was centered in the acclimation region of the tanks. The half-circle false ends and center divider created an enclosed system where water could continually be replaced concurrently with flow being generated around the center divider.

A single 1/3 hp (249 W) Goulds Model 3886 Submersible Sewage Pump pumped Marias River water to the tanks. The pump in the Marias River was inside a filter constructed of a 0.6-m³ redwood frame screened with 6-mm plastic mesh. A 2-in (5.08-cm) polyvinyl chloride (PVC) pipe transported water from the pump to the tanks (Figure 2). Each tank had a ball valve at the end of the affluent line to regulate discharge into the tank. The tanks were each supplied with 102 L/min, which provided a replacement every 50 minutes. A stand pipe kept the water level constant and returned effluent water to the Marias River. Each tank had a single 1/3 hp Goulds Model 3886 Submersible Sewage Pump in its effluent region. The pump discharged water through a 2-in PVC pipe and supplied water within the acclimation region at two equidistant locations. Each location had two 2.5-cm jets. Three ball valves controlled the partitioning of discharge from the pump (Figure 2). The first ball valve was on a perpendicular line located 30 cm from the pump outtake. This served as a bleeder for the system and, in conjunction with the first jet ball valve, allowed 0-100% of the discharge to be returned to the effluent region. The second ball valve was immediately proximal to the perpendicular line that supplied the

Figure 2. Schematic of Marias treatment tanks illustrating continuous water supply with concurrent flow around the center divider.



first set of jets, and the third ball valve was located immediately distal to the aforementioned perpendicular line and controlled discharge to the second set of jets. The discharge from these four jets circulated the entire volume within the acclimation region in a counterclockwise direction. Each tank was covered with 6-mm wire mesh lids. One half of the acclimation region and all of the affluent and effluent regions were covered with white cotton shade cloth. A 4-m high by 6-m wide by 9-m long canopy was erected over the tanks to provide additional shade.

Pallid sturgeon ($N = 30$ in 2005 and $N = 90$ in 2006) were transported from the BFTC and placed in Marias treatment tanks on 28 July in 2005 and 12 June in 2006. Fish densities within the Marias treatment tanks were 0.33 kg/m^2 in 2005 and 0.78 kg/m^2 in 2006. The circulation pumps were on, but all discharge went through the bleeder line. The jet ball valves were opened one-half turn at hour 24 and again at hour 48 to increase water velocity and slowly acclimate the pallid sturgeon to flow. To further increase water velocity, the bleeder ball valve was closed one-half turn at hour 72 and again at hour 96. At this time, discharge to the acclimation region was at maximum and the jet ball valves were adjusted so that equal discharge was supplied at both locations. Mean bottom velocity was $0.23 \pm 0.04 \text{ m/s}$ in both 2005 and 2006 and was maintained for the remainder of the acclimation period.

Fish were fed a mix of 50% Silver Cup No. 4 (2.00 mm - 3.36 mm) Crumbled Salmon Feed (sinking) and 50% BioProducts 3 mm BioDiet Grower. In 2005, immediately following tank construction, preliminary acclimation trials were conducted in order to ensure survival within the acclimation tanks and calculate feed rates. I

decided to feed 1% of the mean fish weight (mean of initial weights) of food per fish per day to all of the tanks rather than the 2.5% recommended in the Upper Basin Pallid Sturgeon Propagation Plan (UBPSPC 2005) because of fungal growth on uneaten food and mortality of pallid sturgeon in the preliminary acclimation trials at 2.5% of the mean fish weight of feed. Thus, each tank was supplied with approximately 1% (10 g) of fish weight per fish per day of feed in 2005. Due to low and negative growth rates that were observed in 2005 (see results), tanks were supplied with approximately 2.5% (60 g) of mean fish weight per fish per day of feed in 2006.

Bozeman Treatment

Bozeman treatment pallid sturgeon were acclimated to flow at the BFTC using spring water (i.e., not stocking site-specific physicochemical water conditions). Due to mortality of unknown origin in Bozeman treatment tanks during preliminary trials, the Bozeman treatment was not conducted in 2005. Thus, the Bozeman treatment was implemented only in 2006. On day 1 of the acclimation period, Bozeman treatment pallid sturgeon ($N = 90$) were placed in the hatchery truck at the BFTC and transported for four hours to keep Marias treatment transportation time from being an independent variable. They were returned to the BFTC and placed in two tanks (45 fish per tank) located in the containment building. Fish density within the Bozeman treatment tanks was 0.78 kg/m^2 . All design specifications, conditions, and monitoring were identical to the Marias treatment tanks except for tank length and water source. The tanks were 9.1-m long, 1.5-m wide, with water depth of 0.5 m. Although overall tank length was greater in the Bozeman treatment than Marias treatment tanks, the acclimation region of all tanks

had identical dimensions. A warm spring (22 °C) and a cold spring (8 °C) were used at the BFTC allowing manipulation of the temperature of the water supplied to the tanks. Water temperatures in the BFTC were adjusted to follow the thermograph of the Marias River as closely as possible. The tanks were each supplied with 22 L/min, and replacement was five hours.

Traditional Treatment

Traditional treatment was no acclimation. Traditional treatment was implemented in 2005 and 2006. On day 1 of the experiment, traditional treatment fish ($N = 30$ in 2005 and $N = 90$ in 2006) were placed in the hatchery truck at the BFTC and transported for four hours to keep Marias treatment transportation time from being an independent variable. They were returned to the BFTC and placed in two 1.8-m diameter circular tanks located in the containment building. Fish densities within the traditional treatment tanks were 0.64 kg/m^2 in 2005 and 1.53 kg/m^2 in 2006. The tanks were supplied with spring water at a rate of 20 L/min and replacement was every 70 minutes. Fish were reared for 36 d in 2005 and 74 d in 2006 under the protocol of the Upper Basin Pallid Sturgeon Propagation Plan (UBPSPC 2005).

Physicochemical Parameters

Physicochemical parameters were measured every 14 d in all tanks and in the Marias River at the Marias treatment pump location. Conductivity (μS), salinity (‰), nitrate ($\text{mg/L NO}_3^- \text{-N}$), nitrite ($\text{mg/L NO}_2^- \text{-N}$), ammonia ($\text{mg/L NH}_3 \text{-N}$), alkalinity (mg/L as CaCO_3), and hardness (mg/L CaCO_3) were measured to ensure that the concentrations

of these parameters remained within the suggested physiological tolerance limits of similar species (see Mims et al. 2002; Portz et al. 2006). A Hach DR/850 Datalogging Colorimeter was used to measure nitrate, nitrite, and ammonia. Nitrate was measured using the cadmium reduction method with NitraVer® 5 Reagent Powder Pillows. Nitrite was measured using the diazotization method with the NitriVer® Nitrite Test 'N Tube™ reagent set. The salicylate method using the AmVer™ Low Range Ammonia Test 'N Tube™ reagent set was used to measure ammonia. Alkalinity and hardness were measured using a Hach Model 16900 Digital Titrator. Alkalinity was measured using the phenolphthalein and total method with the Hach Model AL-DT test kit. Hardness was measured using a total hardness reagent set.

Temperature (°C), dissolved oxygen (mg/L), turbidity (NTU), and water velocity (m/s) were measured to ensure that levels of these parameters remained within pallid sturgeon physiological limits; however, these parameters were also measured to define the varying conditions of acclimation among treatments (see below). Temperature, conductivity, and salinity were measured using a YSI 30 Salinity, Conductivity, and Temperature meter. Temperature was also recorded at 1-h intervals using Onset Optic StowAway® Temp data loggers. Temperature data loggers were placed in each acclimation tank and in the Marias River near the Marias treatment pump location. A YSI Model 50B dissolved oxygen meter was used to measure dissolved oxygen. Turbidity was measured using a Hach 2100P turbidimeter. A Marsh-McBirney Model 2000 portable flow meter was used to measure water velocity. Water velocity was measured 10 cm from the bottom of the tanks (bottom velocity). All equipment was

calibrated and tested prior to the study and once a month throughout the acclimation period. In addition to the 14-d measurements, 24-h profiles of temperature and dissolved oxygen were taken in 4-h intervals every 30 d.

Dissolved oxygen, temperature, turbidity, and water velocity were used to characterize treatments (Table 1). Mean dissolved oxygen concentrations and turbidity levels were greater in the Marias treatment tanks than the Bozeman and traditional treatment tanks (Table 1). Mean daily temperature was similar among treatments (Table 1); however, diel fluctuations in temperature were greater in the Marias treatment than in the Bozeman and traditional treatments (Figure 3). Mean water velocity was greater in Marias and Bozeman treatments than in the traditional treatment (Table 1).

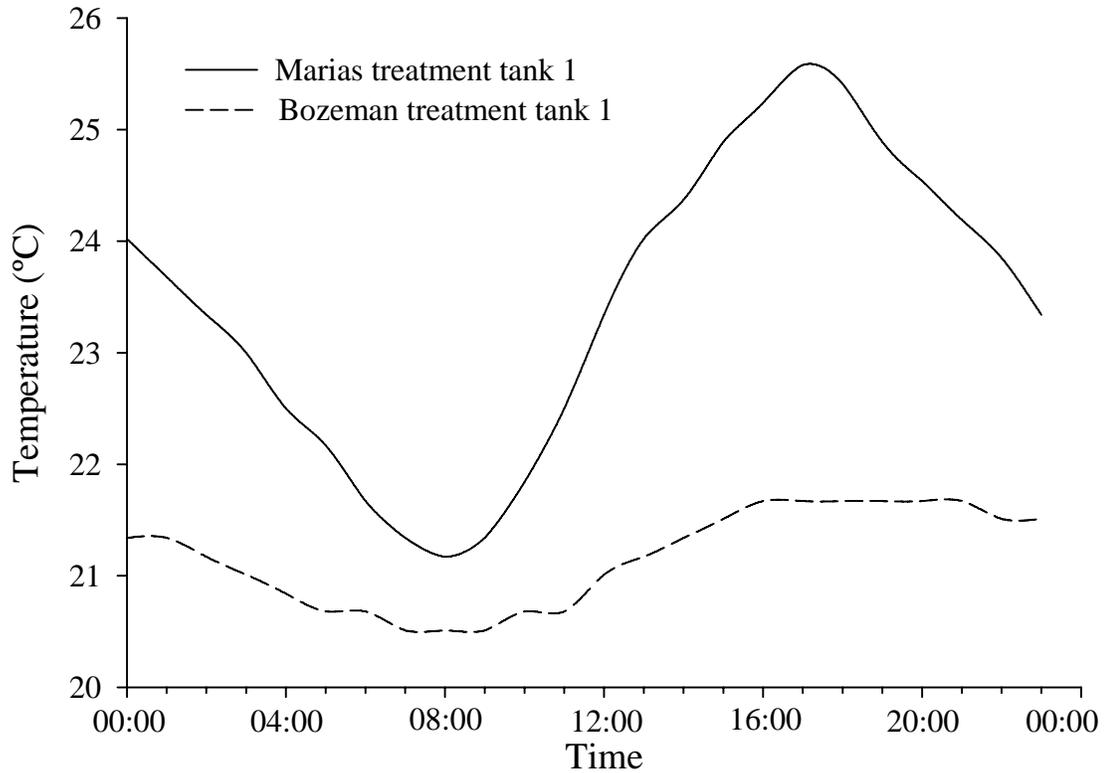
Table 1. Mean (SE) physicochemical water parameters by treatment for 2005 and 2006.

Year	Parameter	Treatment		
		Marias	Bozeman	Traditional
2005 ^a	Dissolved Oxygen (mg/l) ^b	8.19 (0.01)		
	Daily Temperature (°C)	21.0 (< 0.1)		17.8 (< 0.1)
	Turbidity (NTU) ^b	12.0 (< 0.1)		
	Velocity (m/s)	0.23 (0.03)		0.01 (< 0.01)
2006	Dissolved Oxygen (mg/l)	8.11 (0.03)	6.23 (0.01)	6.90 (0.02)
	Daily Temperature (°C)	21.9 (< 0.1)	21.4 (< 0.1)	21.1 (< 0.1)
	Turbidity (NTU)	17.1 (1.1)	1.0 (0.1)	0.6 (0.1)
	Velocity (m/s)	0.23 (0.04)	0.20 (0.03)	0.01 (< 0.01)

^aBozeman treatment was not implemented in 2005.

^bData not collected for traditional treatment in 2005.

Figure 3. Twenty-four hour temperature profile for Marias and Bozeman treatments on 10 July 2006.



Physiological Condition

Fin Curl

Prior to treatment assignment, all individuals were examined for fin curl. Fin curl is a deformation primarily in the pectoral fins (can be expressed in all fins) that is characterized by curling of the pectoral fins in a dorsal direction (Figure 4). Fin curl is unique to the BFTC (for pallid sturgeon) and mechanisms that cause fin curl are currently unknown. Although varying severity of fin curl existed among individuals, fin curl was measured as present or absent.

Figure 4. Dorsal and lateral views of pallid sturgeon pectoral fins with moderate fin curl (upper row) and without fin curl (lower row).



Relative Growth Rate

Growth rate by treatment was determined for the acclimation period by year.

Length and weight were measured immediately prior to the acclimation period and near the end of the acclimation period (day 33 in 2005 and day 63 in 2006). Growth rate was calculated using relative growth rate (Isely and Grabowski 2007):

$$G_{\text{relative}} = (L_2 - L_1) / [L_1(t_2 - t_1)] \times 100$$

where L_1 = length or weight at day 0 of the acclimation period; L_2 = length or weight at the end of the acclimation period; and $(t_2 - t_1)$ = number of days elapsed between

measurements. A nested analysis of variance (ANOVA; tank nested within treatment) was used to compare growth rate for length and weight among treatments.

Energy Reserves

Percent crude fat was used to quantify energy reserves. In 2006, mean percent crude fat was measured on six randomly selected individuals prior to treatment assignment (day 0; pre-treatment level) and from six randomly selected individuals for each treatment on day 63 to obtain post-treatment levels. Treatments were Marias, Bozeman, and traditional. After collection, pallid sturgeon were euthanized and weighed (wet weight; g). All fish were frozen at -80°C until sample preparation. Individuals were partially thawed, sectioned (< 5mm), placed in a 500 mL plastic beaker, and weighed (g). Deionized (DI) water was added at a ratio varying from 1:1 to 3:1 (g DI water:g fish) and the sample homogenized using a Janke and Kunkel IKA® Labortechnik Ultra-Turrax® T25 disperser. A sample of the homogenized material (60 – 190 g) was placed in a Labconco FreeZone¹² lyophilizer. Samples were lyophilized for 48 h and weights taken every two hours until all moisture was extracted from the samples. Three sub-samples were collected from each sample. Sub-samples were placed in Ankom XT4 one micron filter bags, heat sealed, and placed in an aluminum weigh dish. Weights for the filter bag, weigh dish, and sub-sample were measured to the nearest 0.0001 g. Sub-sample weights varied from 0.8427 – 1.4926 g. Sub-samples remained in the weigh dish and were baked at 105°C for three hours in a Yamato DKN 900 forced convection, constant temperature oven. Sub-samples were allowed to cool to room temperature in a desiccator. Sub-samples were weighed, five sub-samples per cycle were placed in an Ankom^{XT10}

extractor, 350 mL of petroleum ether added, and extraction occurred for one hour at 90°C. Compounds extracted were triacylglycerols and a small quantity of related lipids, traditionally termed “crude fat.” After the extraction cycle was complete, sub-samples were baked at 105°C for 15 minutes, placed in the desiccator until cool, and weighed. Crude fat loss was quantified as the loss of mass from the extraction of fat and oil from the sub-sample. Proportion crude fat in dry material was calculated using the following equation:

$$P_{fat} = 1 - (W_2 / W_1)$$

where W_1 = sub-sample dry weight and W_2 = sub-sample post-extraction weight. Mean proportion crude fat in dry material (\bar{P}_{fat}) of the three sub-samples was then calculated for each sample. Total body percent crude fat was obtained by calculating percent dry material in the sample and incorporating mean proportion crude fat in dry material using the following equation:

$$\text{Total body percent crude fat} = (W_2 / W_1) \times 100 \times \bar{P}_{fat}$$

where W_1 = sample wet weight; W_2 = sample dry weight; and \bar{P}_{fat} = mean proportion crude fat in dry material. Total body percent crude fat was square root arcsine transformed (Zar 1984), and ANOVA was used to compare percent crude fat among treatments (pre-treatment was not included in the ANOVA).

Liver Fat Score

Liver samples were collected from six randomly selected individuals prior to the acclimation period in 2006 to obtain a pre-treatment liver fat level. Liver samples were

also collected from six individuals from each treatment on acclimation period day 63 to obtain post-treatment levels. Pallid sturgeon were euthanized, weighed (wet weight), and tissue collected. Liver tissue was collected and fixed in Davidson's solution (acetic acid, ethyl alcohol, formalin, distilled water). Liver tissue was embedded in paraffin, sectioned at 5 μm , and stained using hematoxylin and eosin (Luna 1968). Histological slides were scored according to the Upper Basin Pallid Sturgeon Propagation Plan (UBPSPC 2005) based on fat vacuolation of hepatocytes. Figures representing liver fat score criteria can be found in Appendix A. A Fisher-Freeman-Halton test was used to evaluate differences in liver fat score among treatments (pre-treatment was not included in the Fisher-Freeman-Halton test).

Poststocking Dispersal

Lotek Coded Radio NanoTag transmitters weighing 2.1 g with signals at the 150 to 151 MHz range (150.300, 150.420, or 150.680 MHz) were surgically implanted in hatchery-reared age-1 pallid sturgeon to assess dispersal. Each transmitter had a unique code and burst rates varied from 5.0 s to 5.3 s. Guaranteed battery life was 74 d and this is what determined the maximum duration of the dispersal period. Surgical methods can be found in Gerrity (2005).

Pallid sturgeon poststocking dispersal was quantified using remote telemetry stations placed at four locations along the Missouri River and one location along the Marias River in 2005, and at five locations along the Missouri River in 2006 (Figure 1). The remote station along the Marias River (Loma; 2005) was placed at rkm 0.8. Remote

stations along the Missouri River were at Coal Banks (rkm 3,270; 2005), Virgelle (rkm 3,274; 2006) Judith Landing (rkm 3,190; both years), Power Plant Ferry (rkm 3,118; 2006), Mauland Island (rkm 3,076; both years), and Squaw Creek (rkm 3,001; both years) (Figure 1). Remote stations had a Lotek SRX 400A receiver with W31CT firmware. Power to the receiver was supplied by two 12-volt batteries inside a Lotek Solar Powered Environmental Enclosure. Solar panels kept the batteries charged for the duration of the study. Two Lotek four element Yagi antennas were mounted on each station. All remote stations were field tested to ensure that direction of movement could be deciphered, transmitters could be decoded (tested in the thalweg and opposite river bank), and for interference prior to the release of pallid sturgeon.

After the 36-d (2005) and 74-d (2006) acclimation period, 15 of the radio-tagged pallid sturgeon from each treatment (seven from one tank and eight from the other were randomly selected) were released in the Marias River (Circle Bridge in 2005, Marias River acclimation location 2006). The remaining 15 radio-tagged pallid sturgeon from each treatment (seven from one tank and eight from the other were randomly selected) were released in the Missouri River at the Fred Robinson Bridge (Figure 1). Poststocking dispersal was quantified using remote stations from release until day 68 post-release in 2005 and day 64 post-release in 2006 (dispersal period).

Downstream dispersal rate was calculated as kilometers per day (km/d). Dispersal rate was calculated for each individual from the release site to the first remote station encountered and for each reach between remote stations thereafter. A two-way ANOVA with repeated measures and a nested design (tanks nested within treatments)

was used to analyze dispersal rate. The main effects were treatment and release site. The interaction term for the main effects was included in the model.

Diurnal and nocturnal movements were quantified using remote station data. Sunset and sunrise tables for Fort Benton, Montana were used to establish diel periods. The diurnal period was defined as 30 minutes before sunrise (06:00 in 2005; 05:46 in 2006) to 30 minutes after sunset (18:23 in 2005; 18:44 in 2006) for the dispersal period. Conversely, the nocturnal period was defined as 30 minutes after sunset to 30 minutes before sunrise for the dispersal period. Each time an individual pallid sturgeon transmitter was detected by a remote station the initial time of detection was recorded. Time of fish detection was reported as the total number of detections for the nocturnal and diurnal periods by one-hour period.

Poststocking dispersal was also analyzed using fish locations at the end of the dispersal period. For each year-release site combination, remote station data were used to determine the number of pallid sturgeon from each treatment that remained in a reach (between remote stations) at the end of the dispersal period. In 2006, these locations were validated by comparing them to the fish locations observed during active tracking by jet boat at the end of the dispersal period. A Fisher-Freeman-Halton test was used to test for differences among treatment distributions by release site.

Pallid sturgeon were actively tracked by jet boat at the end of the dispersal period each year to validate remote station locations and identify mortalities. Once a fish was found, its location (river kilometer) was recorded using a Garmin GPSMAP 168 Sounder and attempts were made to make the fish move by driving repeatedly over the fish

location, dragging a steel t-post over the fish location, or wading at the location if water depth was less than one meter. Fish were identified as alive if they moved laterally or in an upstream direction greater than 10 m after being disturbed. Fish were identified as mortalities if the transmitter was recovered from the substrate, or the transmitter was located in shallow water (< 1 m) and did not move after repeated disturbance attempts.

In 2005, 37% (22/60) of fish released were located through active tracking by jet boat at the end of the dispersal period. All individuals were located in the reach (between remote stations) that was consistent with the end location data obtained from remote stations. Three fish were identified as mortalities and were excluded from all analyses. All three mortalities were from the Marias River release.

In 2006, 82% (74/90) of fish released were located through active tracking by jet boat at the end of the dispersal period. For the Marias River release, 78% (35/45) of the individuals were located. Thirty-three fish were located in the reach (between remote stations) that was consistent with the end location data obtained from remote stations. The remaining two individuals were not detected by the Virgelle remote station and located in the reach immediately downstream to that remote station. In addition, the Virgelle remote station did not detect five fish that were later detected by downstream remote stations. Based on these data, the Virgelle remote station did not detect 24% (7/29) of the fish that dispersed downstream of that location. For the Missouri River release 87% (39/45) of the individuals released were located through active tracking by jet boat at the end of the dispersal period. All fish were located in the reach that was

consistent with the end location data obtained from remote stations. No mortalities were observed in 2006.

Data Analysis

All data were analyzed using Statistical Analysis System (SAS Institute, Inc. 2003) and alpha was 0.05 for all analyses. Years were analyzed separately for all data analysis because several extraneous variables differed between years (e.g., acclimation period duration, release site locations, remote station locations, amount of feed, post-surgery recovery time). Mixed models were used for all ANOVAs with repeated measures [for analyses with repeated measures, general linear models require data to be balanced within subjects (Wolfinger and Chang 1995)] and general linear models were used for all ANOVAs not containing repeated measures. All models containing two main effects were run with the interaction term included. Multiple comparisons were conducted using least-squares means (lsmeans) with Tukey-Kramer adjustment. Model assumptions of normality and homogeneity of variance were evaluated for all ANOVA models through visual examination of normal probability and residual plots.

RESULTS

Physiological Condition

Fin Curl

One hundred percent (60/60) of the pallid sturgeon had fin curl in 2005. In 2006, 27% (74/270) of all the pallid sturgeon had fin curl and 26% (23/90) of the fish used in the poststocking dispersal experiment had fin curl (10 from Marias treatment, 7 from Bozeman treatment, and 6 from traditional treatment). Fin curl was most prevalent in the pectoral fins in both years.

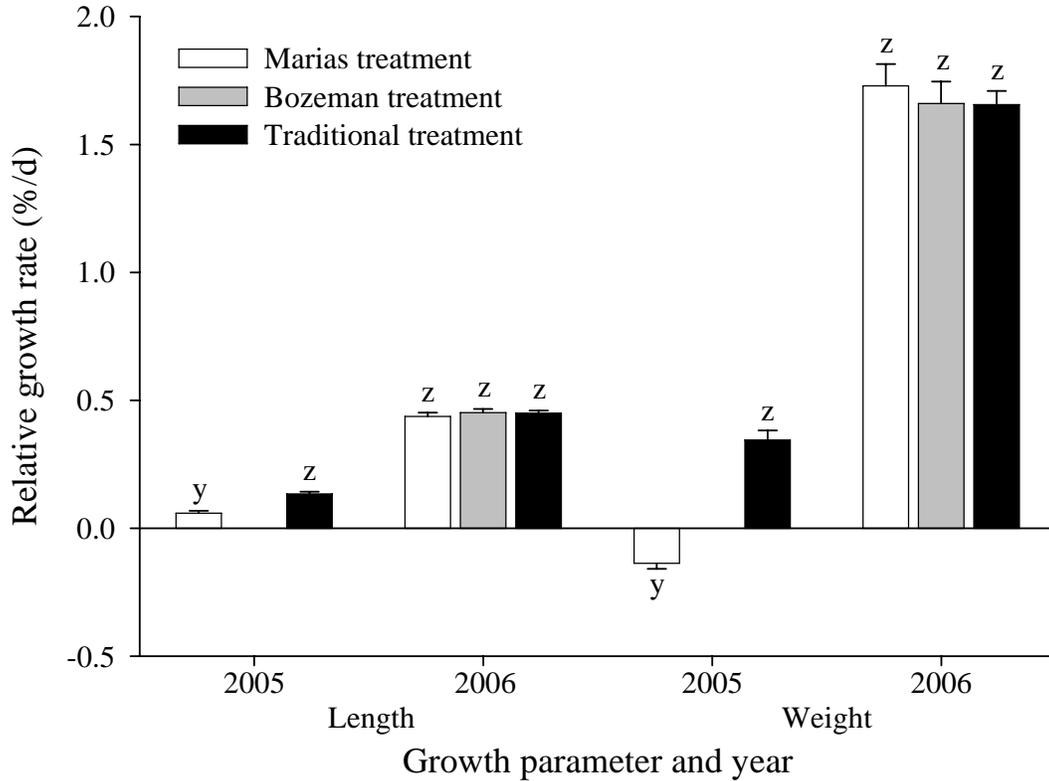
Relative Growth Rate

Relative growth rate differed significantly between treatments in length ($F_{3, 55} = 12.97, P < 0.01$) and weight ($F_{3, 55} = 58.50, P < 0.01$) in 2005. Relative growth rate for length and weight was lower for the Marias treatment compared to the traditional treatment (Figure 5). Relative growth rate did not differ significantly among treatments for length ($F_{5, 84} = 1.62, P = 0.16$) or weight ($F_{5, 84} = 1.43, P = 0.22$) in 2006 (Figure 5). Although statistical comparisons in relative growth rates were not made between years, mean relative growth rate in 2006 was approximately three times greater for length and five times greater for weight than in 2005 (Figure 5).

Energy Reserves

Crude fat differed significantly among treatments ($F_{2, 15} = 7.94, P = 0.04$; Figure 6). Percent crude fat was greater for the Bozeman treatment than the Marias treatment

Figure 5. Mean relative growth rate for length and weight in 2005 and 2006. Dissimilar letters indicate significant differences between or among treatments within growth parameter and year. Error bars denote standard error.

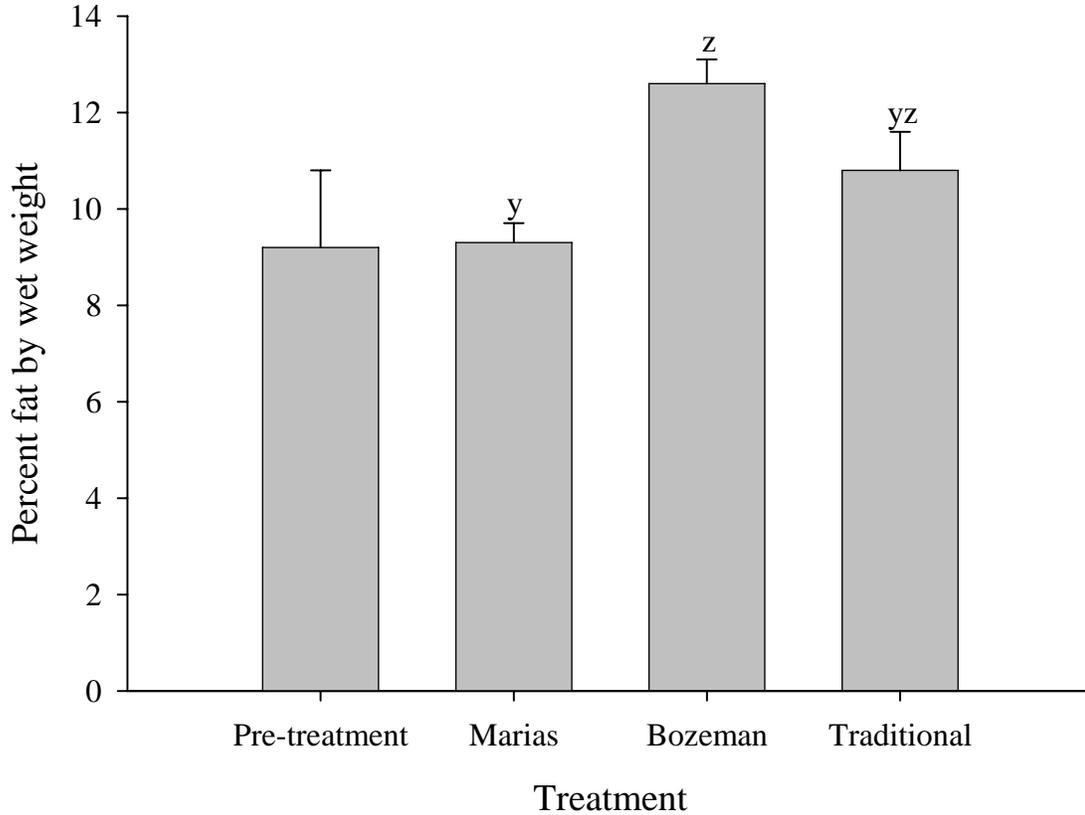


(lsmeans, $N = 12$, $P = 0.03$). Crude fat for the traditional treatment did not differ from the Marias or Bozeman treatments (Figure 6).

Liver Fat Score

The majority of pallid sturgeon sampled had a mean liver fat score 4, followed by liver fat score 3, and one fish had liver fat score 2 (Table 2). Liver fat score did not differ significantly among treatments ($N = 18$, $P = 0.81$). Although there were no significant differences among treatments and no livers had mean liver fat score 5, five of the six livers sampled from the traditional treatment contained localized areas of liver fat score 5. In those localized areas, fat vacuolation of hepatocytes was extreme and caused cell

Figure 6. Pallid sturgeon crude fat by wet weight. Dissimilar letters indicate a significant difference among treatments (pre-treatment was not included in the statistical analysis). Error bars denote standard error.



membranes to rupture (see Appendix A). No localized areas of liver fat score 5 were observed in the Marias treatment, Bozeman treatment, or pre-treatment.

Three pallid sturgeon were identified as mortalities throughout the study. All three fish were from the 2005 traditional treatment for the Marias River release. Transmitters from two fish were located and recovered in the Marias River at the end of the dispersal period. The other individual was also located in the Marias River; however, the transmitter was not recovered. These pallid sturgeon were removed from the dispersal rate and end location analyses.

Table 2. Percent of individuals in each liver fat content category by treatment for 2006 (liver fat score 0 = no fat vacuoles present, 1 = few hepatocytes contain fat vacuoles, 2 = less than 50% of hepatocytes contain fat vacuoles, 3 = most hepatocytes contain fat vacuoles but cells retain normal shape, 4 = hepatocytes are greatly enlarged and normal cytoplasm is displaced by fat but membranes are intact, 5 = numerous hepatocytes show ruptured cell membranes due to fat accumulation).

Treatment	Liver Fat Score					
	0	1	2	3	4	5
Pre-treatment	0	0	17	33	50	0
Marias	0	0	0	50	50	0
Bozeman	0	0	0	17	83	0
Traditional	0	0	0	33	67	0

Poststocking Dispersal

Remote stations detected pallid sturgeon 125 times in 2005. Seventy-seven percent ($N = 96$) of those detections occurred during the nocturnal period (Figure 7).

Remote stations detected pallid sturgeon 122 times in 2006. Ninety percent ($N = 110$) of those detections occurred during the nocturnal period (Figure 7). Modal hour of fish detection was 02:00 in 2005 and 00:00 in 2006.

In 2005, the treatment-release site interaction was non-significant ($F_{1, 66.4} = 0.53$, $P = 0.47$). Downstream dispersal rate differed significantly between treatments [$F_{1, 66.4} = 8.08$, $P = 0.01$ (pooled by release site)]. Dispersal rate was greater for the traditional treatment than for the Marias treatment (Figure 8). Further, downstream dispersal rate differed significantly between release sites [$F_{1, 66.4} = 11.43$, $P < 0.01$ (pooled by treatment)]. Pallid sturgeon released in the Marias River dispersed at a greater rate than fish released in the Missouri River (Figure 8).

In 2006, the treatment-release site interaction was non-significant ($F_{2, 56.9} = 0.94$, $P = 0.40$). Dispersal rate did not differ significantly among treatments [$F_{2, 56.9} = 0.77$, $P =$

Figure 7. Fish detection by time of day by remote stations for all pallid sturgeon in 2005 and 2006. Nocturnal and diurnal periods were estimated using mean daily time 30 minutes before sunrise and 30 minutes after sunset for the dispersal period (2 September through 9 November 2005; 25 August through 28 October 2006).

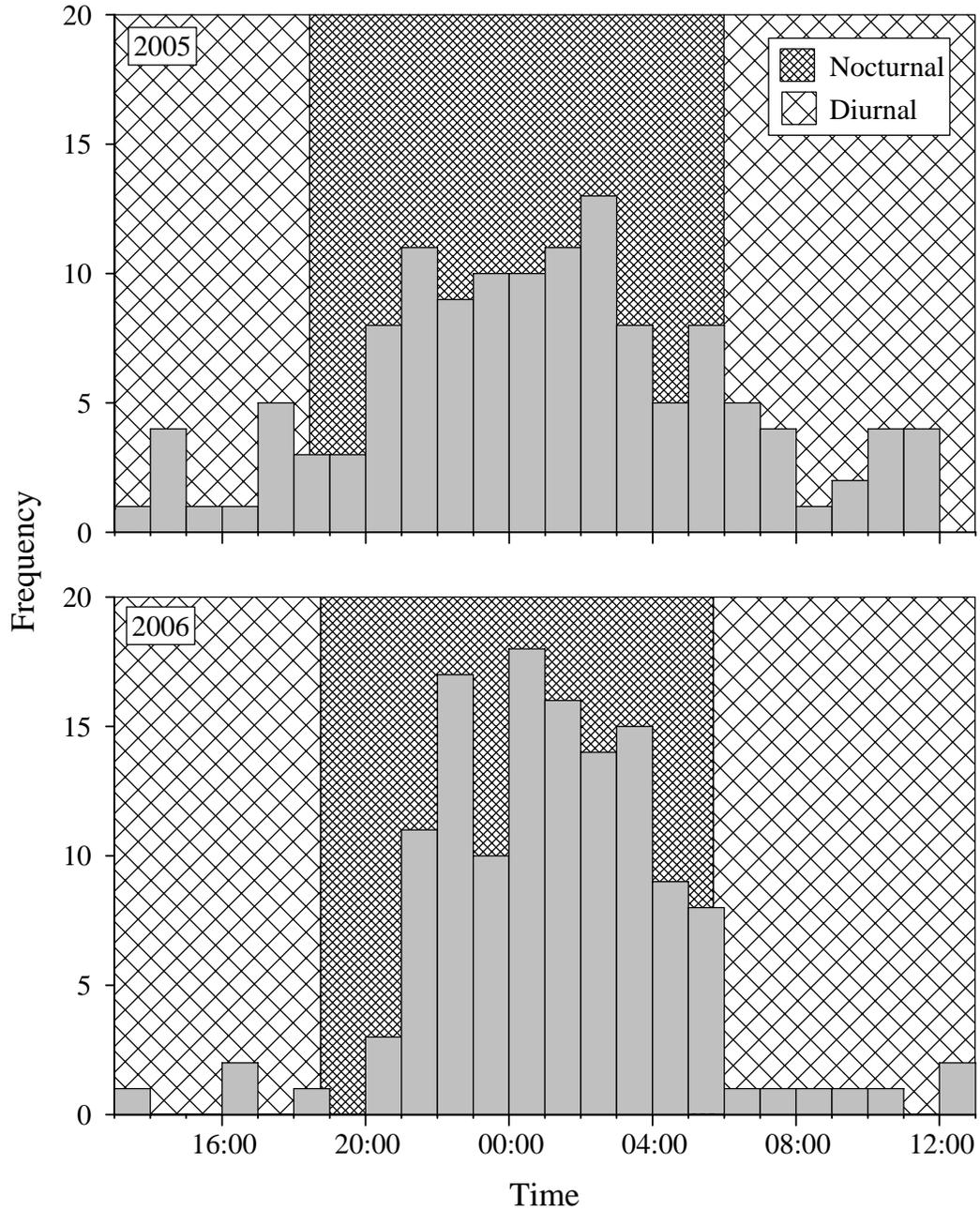
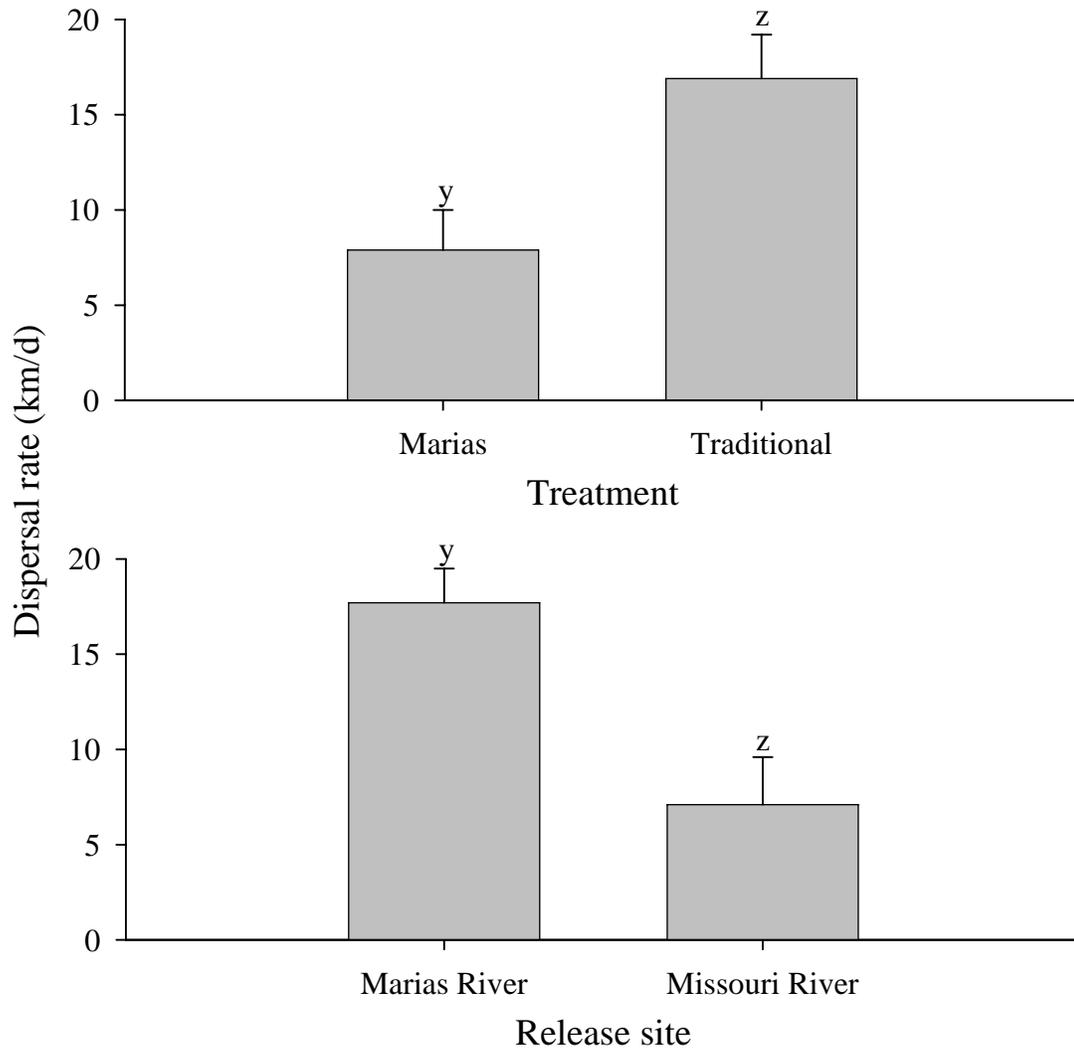
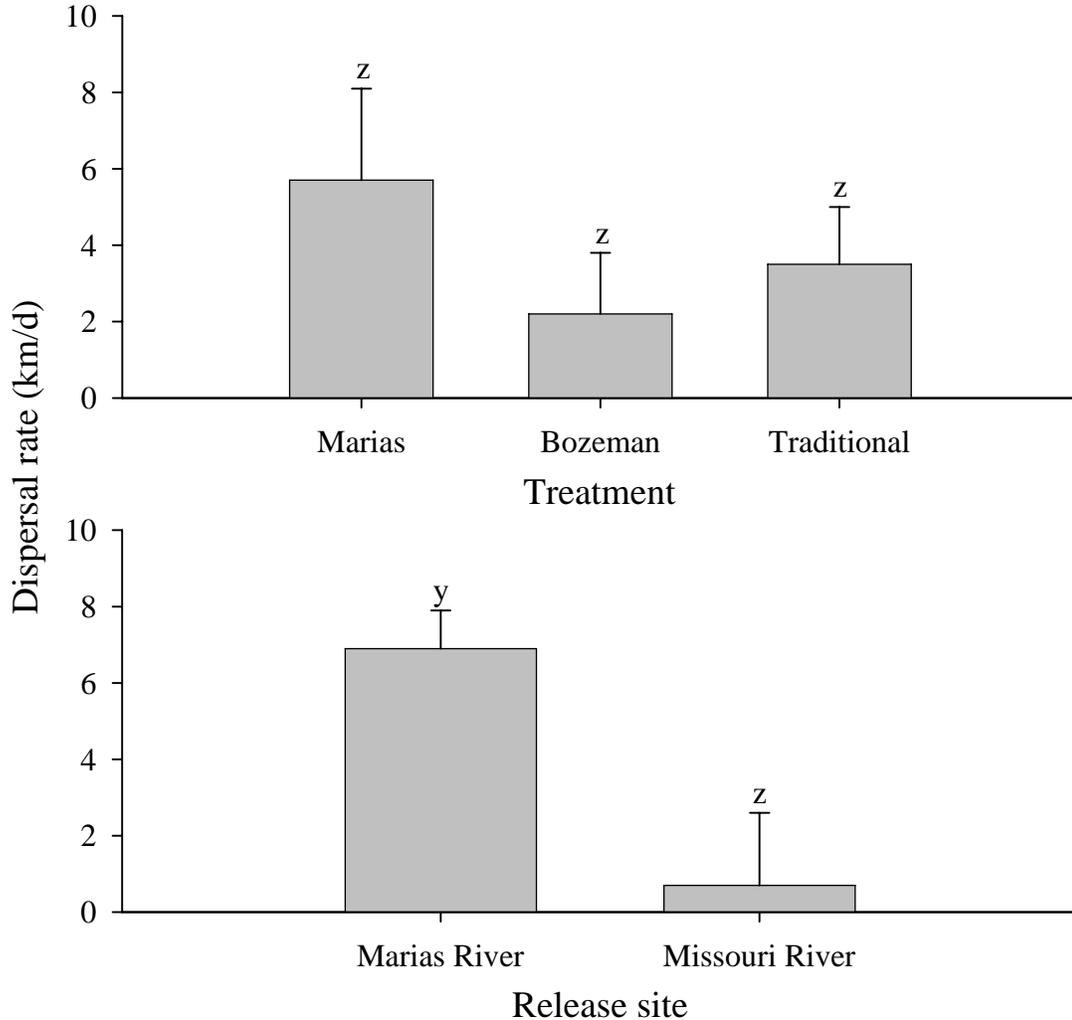


Figure 8. Downstream dispersal rate by treatment and release site in 2005. Dissimilar letters indicate significant differences between treatments or release sites. Error bars denote standard error. Bozeman treatment was not conducted in 2005.



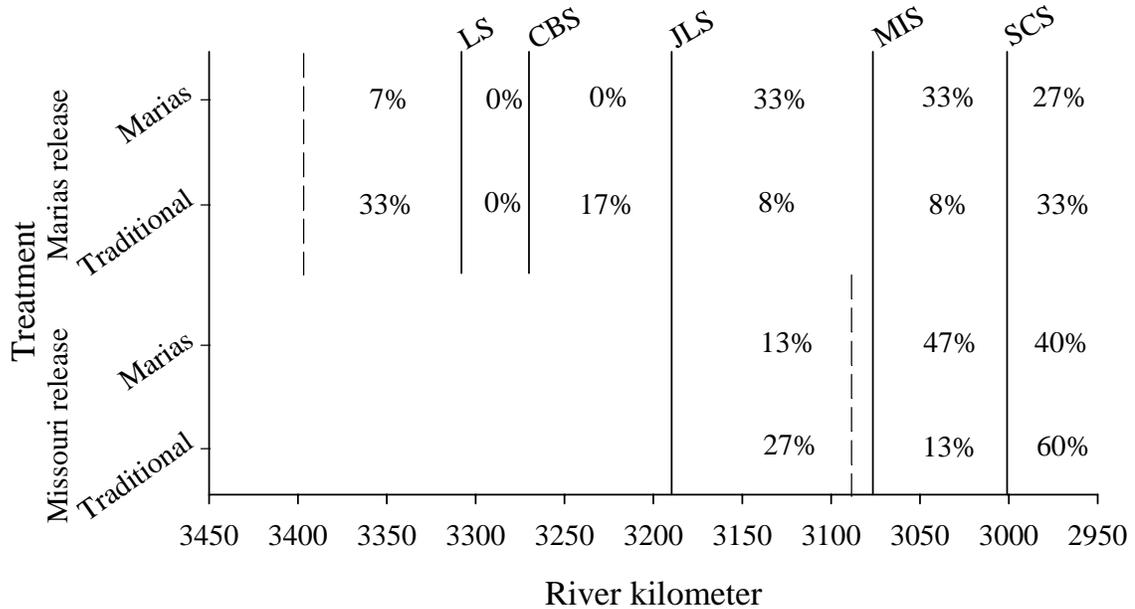
0.47; Figure 9 (pooled by release site)]. However, downstream dispersal rate differed significantly between release sites [$F_{1,58.1} = 8.24$, $P = 0.01$ (pooled by treatment)]. As in 2005, fish released in the Marias River dispersed at a greater rate than fish released in the Missouri River (Figure 9).

Figure 9. Downstream dispersal rate by treatment and release site in 2006. Dissimilar letters indicate significant differences among treatments or between release sites. Error bars denote standard error.



Treatments did not differ significantly with respect to end location distribution for a given release site in 2005 [Marias River release ($N = 27$, $P = 0.07$), Missouri River release ($N = 30$, $P = 0.16$); Figure 10] or 2006 [Marias River release ($N = 45$, $P = 0.22$), Missouri River release ($N = 45$, $P = 0.17$); Figure 11]. In 2005, 27% of the Marias treatment and 33% of the traditional treatment pallid sturgeon released in the Marias

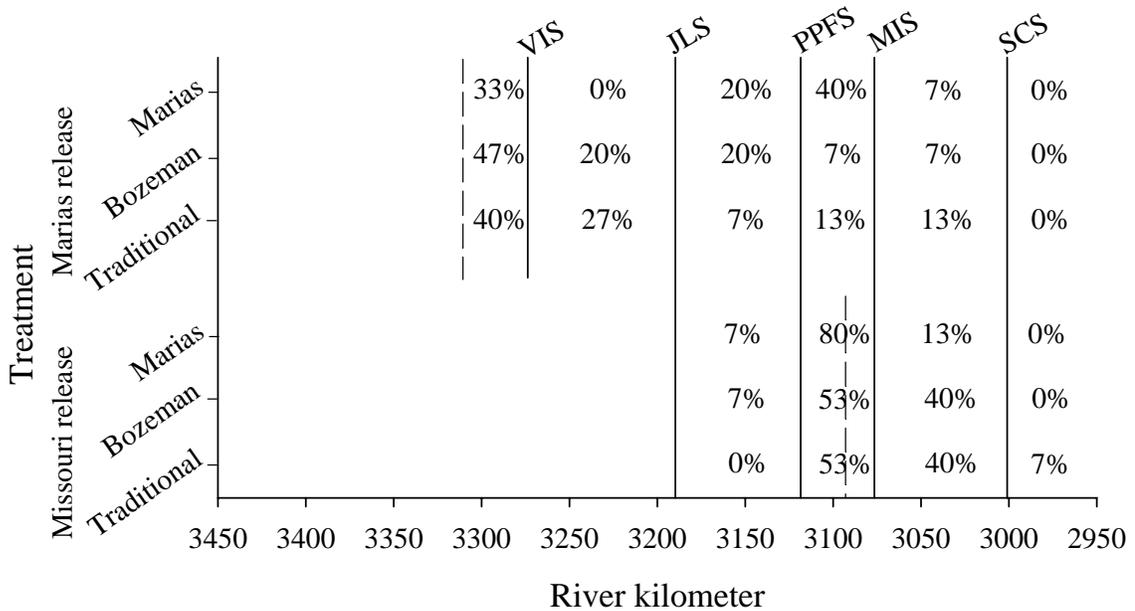
Figure 10. Percent of pallid sturgeon remaining in each reach at the end of the dispersal period by release site and treatment in 2005. Dashed lines represent release site locations. Solid lines represent remote station locations [LS = Loma station, CBS = Coal Banks station, JLS = Judith Landing station, MIS = Mauland Island station, SCS = Squaw Creek station (headwaters of Fort Peck Reservoir)]. Figure excludes three mortalities from Marias River release traditional treatment.



River dispersed downstream 400 km, and possibly greater, past the Squaw Creek remote station into Fort Peck Reservoir (Figure 10). Forty percent of the Marias treatment and 60% of the traditional treatment pallid sturgeon released in the Missouri River dispersed 92 km or greater into Fort Peck Reservoir over the same period (Figure 10). Overall in 2005, 40% (23/57) of pallid sturgeon released dispersed into Fort Peck Reservoir. In 2006, none of the 45 individuals released in the Marias River and one of the 45 individuals released in the Missouri River dispersed into Fort Peck Reservoir.

In 2006, five pallid sturgeon (two from the Marias treatment and three from the Bozeman treatment) traveled upstream past the Power Plant Ferry remote station (rkm 3,118). Two of these fish (one each from the Marias and Bozeman treatments) remained above the Power Plant Ferry remote station through the dispersal period (Figure 11). The remaining three fish dispersed back downstream; however, none of these fish dispersed downstream of rkm 3,075. No upstream movement was documented in 2005; however, the Power Plant Ferry remote station was not implemented that year.

Figure 11. Percent of pallid sturgeon remaining in each reach at the end of the dispersal period by release site and treatment in 2006. Dashed lines represent release site locations. Solid lines represent remote station locations [VIS = Virgelle station, JLS = Judith Landing station, PPFS = Power Plant Ferry station, MIS = Mauland Island station, SCS = Squaw Creek station (headwaters of Fort Peck Reservoir)].



DISCUSSION

Release site (i.e., habitat) appeared to influence poststocking dispersal rate more than treatment conditions. Thus, hatchery-reared pallid sturgeon that were acclimated to river conditions dispersed downstream similarly to non-acclimated pallid sturgeon and all treatments exhibited habitat selection. The river reach extending from slightly above the Fred Robinson bridge (rkm 3,098; Missouri River release site) downstream to the headwaters of Fort Peck Reservoir (rkm 3,001) retains many of the characteristics of a large warmwater river and is an important reach for hatchery-reared juvenile pallid sturgeon and their prey (Gerrity 2005). Hatchery-reared juvenile pallid sturgeon are primarily associated with fines and sand substrate in the upper Missouri River (Gerrity 2005), which is the most common substrate in this reach (Gardner 1994). Gravel and cobble are the primary substrates above that reach (Gardner 1994). Thus, the reach between rkm 3,001 and rkm 3,098 is thought to contain the most suitable habitat for juvenile pallid sturgeon in the upper Missouri River.

During both years dispersal rates were higher for pallid sturgeon released in the Marias River than for fish released in the Missouri river, regardless of treatment (i.e., no interaction). Further, pallid sturgeon released in the Marias River dispersed more rapidly above the suitable habitat as opposed to within this reach. For example, pallid sturgeon released in the Marias River were distributed more uniformly over the 309 km reach from the release site through the suitable-habitat reach. Conversely, most of the pallid sturgeon released in the Missouri River remained within the suitable-habitat reach.

For 2005, it appears that neither treatment was being fed to satiation as growth rates were lower than those observed in 2006. Further, pallid sturgeon from the Marias treatment grew at a slower rate than the traditional treatment in both length and weight. The difference between growth rates in 2005 may have been caused by feed in the Marias treatment tanks settling into the interstitial spaces in the gravel substrate where it was unavailable for consumption by pallid sturgeon. The traditional treatment contained no substrate. Thus, it is likely that the difference observed between the Marias and traditional treatments in 2005 was a function of the amount of available feed and not exercise conditioning because of the lack of differences in growth rates in 2006. It is not likely that density within the acclimation tanks negatively influenced growth rates. Growth rates are not affected by rearing densities less than 3.75 kg/m^2 for juvenile lake sturgeon (Fajfer et al. 1999). Further, none of the densities within tanks during this study exceeded the 3.59 kg/m^2 maximum density recommended for optimal fish growth for juvenile Atlantic sturgeon *Acipenser oxyrinchus* (Jodun et al. 2002).

Although dispersal rate for the Marias treatment was lower than the traditional treatment in 2005, no difference was observed in end location between these two treatments. However, end location between treatments for the Marias River release approached significance. Sixty-six percent of the Marias treatment pallid sturgeon were located in the suitable-habitat reach at the end of the dispersal period, whereas only 16% of the traditional treatment fish were located in that reach. Further, 33% of traditional treatment remained in the Marias River throughout the dispersal period. All three pallid sturgeon that were identified as mortalities were located in the Marias River and no fish

located in the Marias River were identified as living. Thus, it is highly likely that the remaining fish in the Marias River were mortalities.

In 2006, fish were fed the recommended 2.5% of the mean fish weight of feed per fish per day. There were no differences among treatments in growth rates for length or weight. This is consistent with the findings for brook trout *Salvelinus fontinalis* where final weight between fish exercised for 10 weeks and unexercised fish did not differ (Leon 1986). Crude fat differed among treatments, but all treatments had sufficient levels to aid in the transition between hatchery and river environment (Volkman et al. 2004). Thus, the acclimation treatments did not negatively influence crude fat levels (i.e., did not exhaust energy reserves). No differences were found in liver fat score among treatments. However, it is possible that a difference did exist and was not detectable due to low sample size (Type II error), the insensitivity of chi-square analysis for low sample size (Fisher-Freeman-Halton), or that a longer acclimation period is needed to express the variation in liver fat content. High liver fat scores were observed in localized areas of some livers. Five of the six livers sampled from the traditional treatment contained localized areas of ruptured hepatocyte membranes from fat accumulation in the cell (liver fat score = 5), resulting in loss of normal liver architecture. No ruptured hepatocyte membranes were observed in fish in the other treatments or pre-treatment. This difference may be biologically important as liver fat score 4 is considered borderline pathological and liver fat score 5 is pathological (UBPSPC 2005). Liver lipid accumulation in the liver causes metabolic disorder of the liver (Zhang et al. 2004) and survival decreases with increased liver lipid levels (Feng and Jia 2005). Therefore,

exercise conditioning (acclimation to flow) may be beneficial in reducing liver fat content; however, further investigation using longer periods of acclimation or greater velocities is warranted.

Dispersal rate and end location were similar among treatments in 2006. Only one individual dispersed into Fort Peck Reservoir and 90% of the detections occurred during the nocturnal period. Therefore, it appears that all fish, regardless of treatment, were physiologically capable of controlling their dispersal and selected for the suitable-habitat reach in 2006.

Release site influenced dispersal rate more than treatment conditions in both years, but there were slight differences in the results between years. Statistical comparison between years was not feasible given the addition of a treatment in 2006 and differences in release site, remote station locations, acclimation time, feed amount, and density in the tanks. Nevertheless, it is worthwhile to discuss some of the idiosyncrasies between years because it provides insight into the relation between physiological condition and dispersal rate.

Fin curl likely influenced dispersal rate between 2005 and 2006. Fin curl was more severe in 2005 than in 2006. Pectoral fins have been demonstrated to be critical to swimming performance and station-holding (maintaining station without actively swimming by using pectoral fins and body morphology to generate negative lift) efficacy of sturgeons (Adams et al. 1997, 1999, 2003; Wilga and Lauder 1999) and other benthic fishes (Webb 1989; Arnold et al. 1991; Webb et al. 1996). However, one study found no difference in station-holding ability between shovelnose sturgeon with and without the

anterior most fin ray of one pectoral fin removed (Parsons et al. 2003). However, the degree of fin curl observed in 2005 likely influences station-holding ability more than the removal of one fin ray. Further, the fish used in the Parsons et al. (2003) study were larger (510 - 648 mm) than those used in this study. The contention that pectoral fins influence station-holding and swimming ability in pallid sturgeon is supported by anecdotal evidence from pallid sturgeon released in the upper Missouri River in previous years. Pallid sturgeon from the 1997 year class had no fin curl and appear to have recruited to the population in the Missouri River (191 of 732 individuals released have been recaptured to date; Gardner 2007). However, individuals from the 2001, 2003, and 2004 year classes of pallid sturgeon released in the upper Missouri River have had fin curl and have shown little evidence of recruitment (34 of 5,977 individuals released have been recaptured to date; Gardner 2007). Moreover, 44 of 7,114 pallid sturgeon from the 2005 year class (no fin curl) were recaptured during 2006 (Gardner 2007). Further, pallid sturgeon appeared to be less capable of controlling their dispersal in 2005 than in 2006 as more fish were dispersing during the diurnal period and more fish dispersed into Fort Peck Reservoir. Therefore, the diminished pectoral fin condition of the pallid sturgeon in 2005 likely increased downstream poststocking dispersal relative to the pallid sturgeon in 2006.

In 2005, pallid sturgeon acclimated to river conditions along the Marias River dispersed at a slower rate than those not acclimated and fewer acclimated fish dispersed into Fort Peck Reservoir. In 2006, all treatments were similar with respect to dispersal rate. Thus, acclimation to river conditions may be important when fin curl is present. It

has been demonstrated that fish that are forced to swim for long periods of time (exercised fish) have greater swimming stamina than unexercised fish (Leon 1986; Ward and Hilwig 2004). However, pallid sturgeon spend approximately 18% of their time free swimming (Adams et al. 2003). Therefore, acclimation to flow may not be necessary for pallid sturgeon that have no physiological maladies because they are efficient at station-holding. Conversely, acclimation may be highly beneficial if pallid sturgeon have reduced swimming and station-holding ability due to fin curl or other physiological abnormalities.

Fish size may have also contributed to the differences in dispersal between years. Mean fish size at the time of release was greater in 2006 (length = 393 ± 2 mm; weight = 204 ± 3 g) than in 2005 (length = 341 ± 3 mm; weight = 124 ± 3 g). The maximum observed sustained swimming speed of age-0 pallid sturgeon was 0.25 m/sec for large fish (170 – 205 mm) and 0.10 m/sec for small fish (130 – 168 mm) (Adams et al. 1999). Although the pallid sturgeon used in Adams et al. (1999) were slightly smaller and eight months younger than the fish used in this study, they demonstrate that relatively small differences in size may have a great impact on swimming ability. Thus, the size difference in pallid sturgeon between years and the additional acclimation time in 2006 along with fin curl likely explains the differences in dispersal rate between years.

Eighty-three percent of pallid sturgeon detections occurred during the nocturnal period. This suggests highly nocturnal dispersal behavior for age-1 pallid sturgeon. Interestingly, one study found that adult pallid sturgeon exhibit predominantly diurnal behavior (Bramblett and White 2001). This discrepancy may be due to variation within

the life history of pallid sturgeon. The diel movement data also suggests that dispersal rate is influenced not only by habitat selection and physiological capabilities of the fish, but also by their swimming behavior. Three primary swimming behaviors have been demonstrated in pallid sturgeon; station-holding, substrate skimming (ventral body surface is in contact with substrate but propulsion is generated by body and caudal fin undulation), and free swimming (swimming occurs within the water column) (Adams et al. 1999, 2003). It is evident that pallid sturgeon without physiological abnormalities are able to maintain position within the river and that they likely utilize station-holding during the diurnal period, thus expending little energy while maintaining position within the river. During the nocturnal period pallid sturgeon are likely substrate skimming and free swimming as part of foraging activities, seeking more preferred habitats (e.g., velocity, substrate, turbidity, water temperature), or other behavioral activities. Therefore, the amount of time that pallid sturgeon spend within the water column (not appressed to the substrate) should be considered when thinking about dispersal rate.

Although it appears that there is little benefit in acclimating pallid sturgeon that have no physiological anomalies to flow and physicochemical conditions, the effects of acclimation to conditions other than those used in this study remain unknown and lead to alternative hypotheses. The largest pallid sturgeon from the BFTC were selected for use in this study. The effects of acclimating smaller pallid sturgeon to flow and physicochemical conditions remains unknown. Further, it has been suggested that it may take greater than one year for hatchery-reared pallid sturgeon to become acclimated to the river environment (Jordan et al. 2006). Thus, a longer acclimation period may be

desirable. Moreover, if fish were acclimated for a longer period of time or at greater velocities, swimming performance may be increased and liver fat content reduced. Further, acclimation to site-specific physicochemical conditions may have influenced pallid sturgeon dispersal. For example, pallid sturgeon may have selected for the upper reaches of the upper Missouri River (i.e., near Virgelle) if they were acclimated in this area rather than in the Marias River. Another hypothesis is that downstream dispersal may be reduced if individuals are released at an older age (e.g., age 2 or age 3). Lastly, it may not be desirable for pallid sturgeon to remain in the upper reaches of the upper Missouri River because of the lack of suitable habitat.

Management Implications

It appears that there is little benefit in acclimating pallid sturgeon to flow and site specific water conditions for 36 to 74 d unless they have physiological abnormalities. Acclimation may reduce poststocking dispersal in pallid sturgeon with fin curl and aid in reducing liver fat content of hatchery-reared individuals. Further, stocking at a single location (Fred Robinson Bridge) rather than multiple locations may be beneficial to pallid sturgeon because it would reduce dispersal distance and likely the stress associated with dispersal. This is consistent with the findings of Jordan et al. (2006) who found that stocking at one location was sufficient to support pallid sturgeon recovery efforts in the Missouri River below Fort Randall Dam.

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APPENDIX A

LIVER FAT SCORE CRITERIA

Figure 12. Pallid sturgeon liver cross sections (5 μm) magnified 40X. Numbers denote liver fat score (0 = no fat vacuoles present, 1 = few hepatocytes contain fat vacuoles).

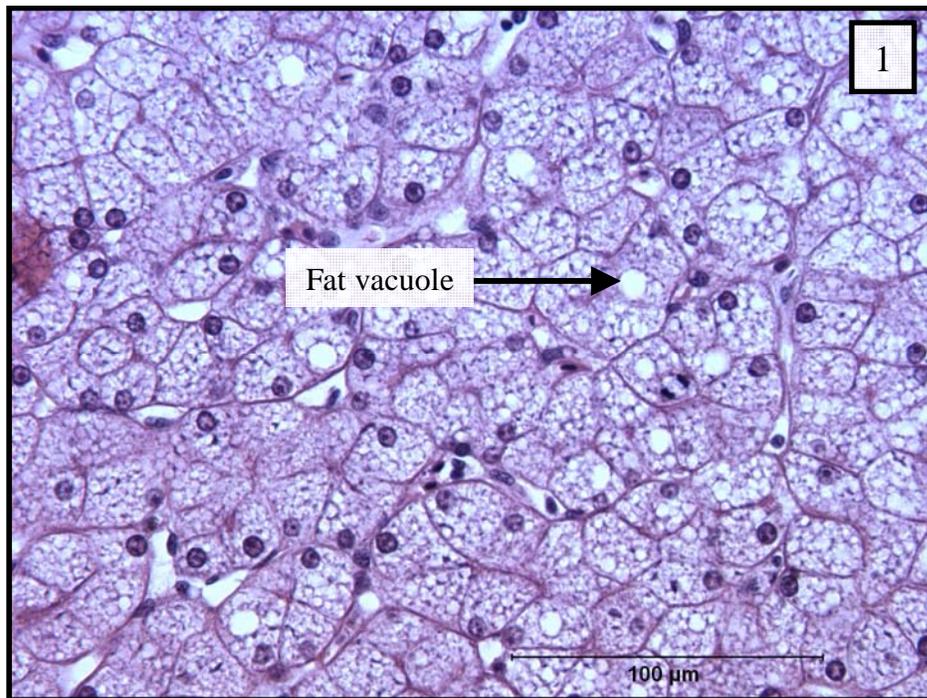
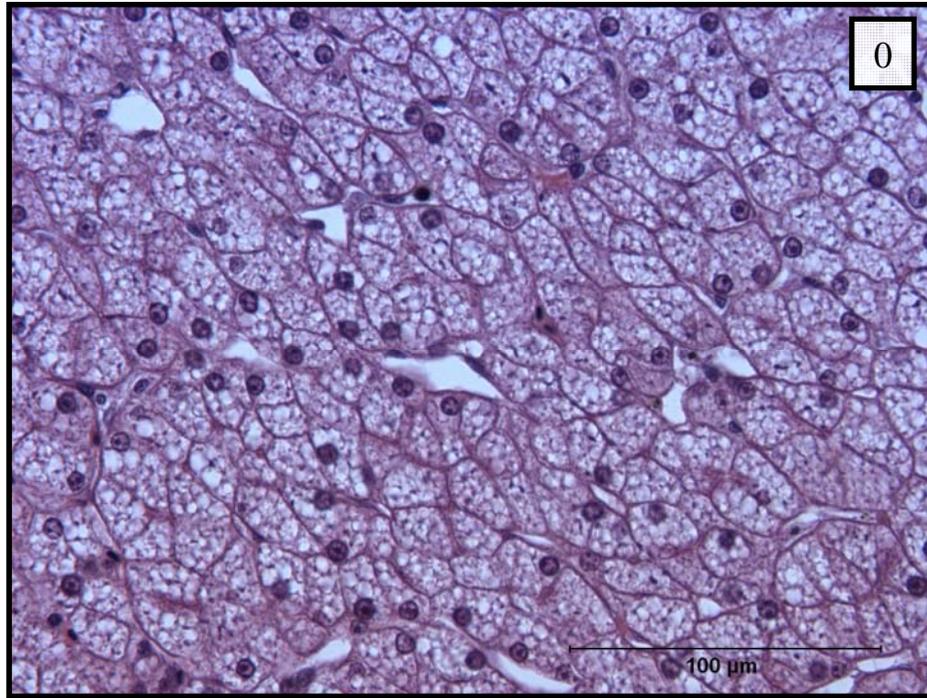


Figure 13. Pallid sturgeon liver cross sections (5 μm) magnified 40X. Numbers denote liver fat score (2 = less than 50% of hepatocytes contain fat vacuoles, 3 = most hepatocytes contain fat vacuoles but cells retain normal shape).

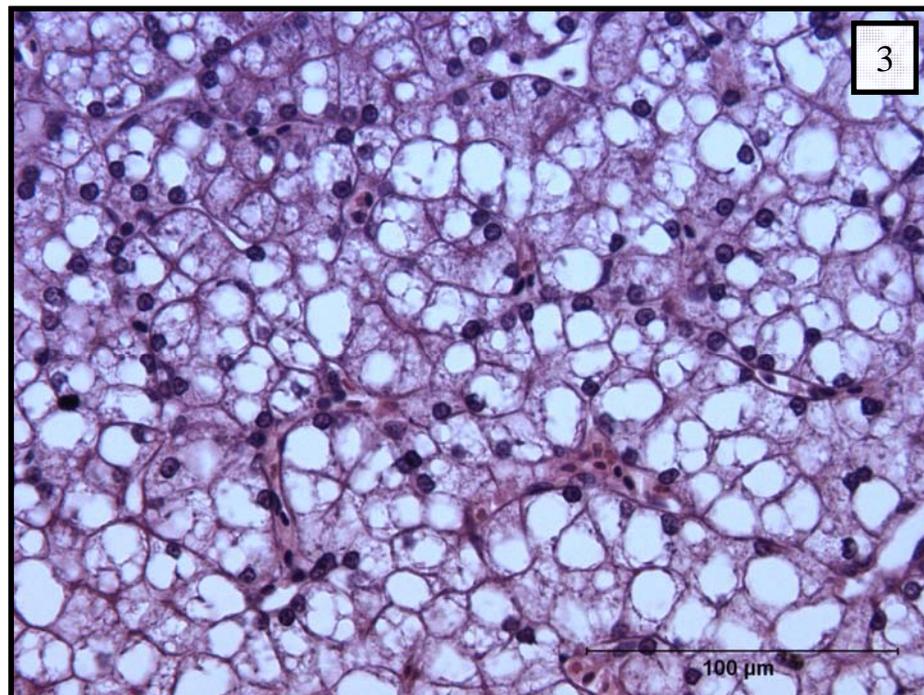
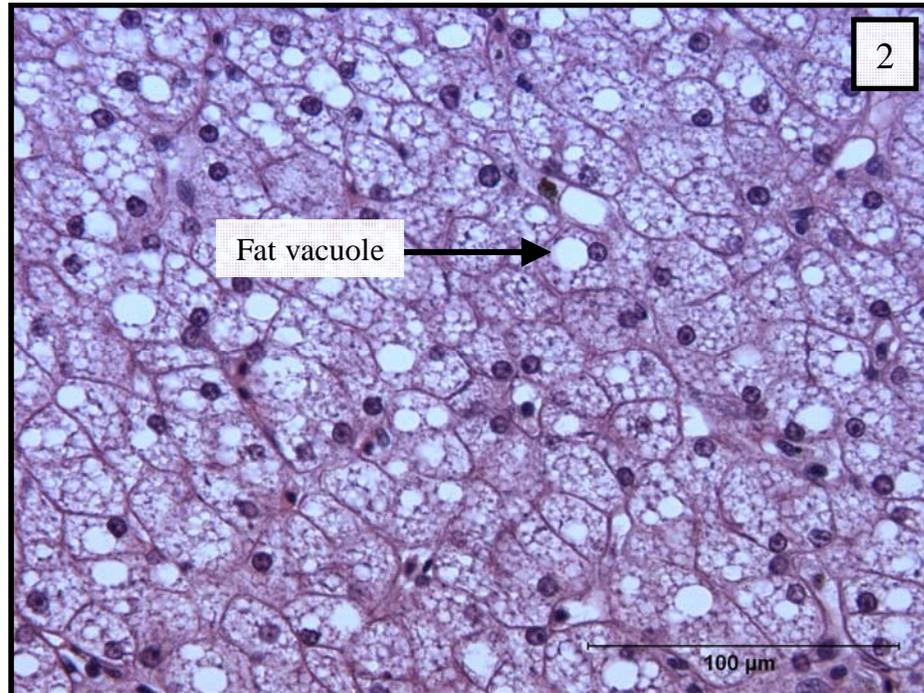


Figure 14. Pallid sturgeon liver cross sections (5 μm) magnified 40X. Numbers denote liver fat score (4 = hepatocytes are greatly enlarged and normal cytoplasm is displaced by fat but membranes are intact, 5 = numerous hepatocytes show ruptured cell membranes due to fat accumulation).

