Effects of Traveling Screen Operation on Impingement and the Survivability and Latent Health of Impinged Catfish at Barry Steam Generation Plant

by

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Abstract

Two studies were conducted concurrently at cooling water intake structures at a coal fired power plant on the lower Mobile River basin. The first study observed the effects of varying operation times of traveling screen on impingement rates of fish and shellfish. The second study observed the survival rates of blue catfish (*Ictalurus furcatus*) and channel catfish (*Ictalurus punctatus*) for seven days post-impingement. Observations were made and recorded of gross lesions, parasite load and presence of known fish pathogens of impinged fish at impingement, of mortalities of the survival study and survivors of the survival study. The travelling screen study results showed no significant difference in freshwater species impinged at the cooling water intake structures but significantly increased impingement of salt-water species when screens were operated intermittently. The survival rates for impinged blue catfish (6.5%) and channel catfish (12.5%) were significantly lower than the channel catfish controls (91.0%). The health study found 87% of the impinged fish had known fish pathogens present. *Flavobacterium columnare* was the most common pathogen and was found on 66.7% of fish sampled at impingement, on 71.6% of the survival study mortalities, and on 66.7% of the impingement survivors. This was significantly higher than the 4.8% infection rate of the surviving control fish in the study.

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List of Abbreviations

AFS   American Fisheries Society

BHI   Brain-Heart Infusion

BTA   Best Technology Available

CWA   Clean Water Act

CWIS   Cooling Water Intake Structure

DO   Dissolved Oxygen

EPA   Environmental Protection Agency

EPRI   Electrical Power Research Institute

HZI   Hydraulic Zone of Influence

ISOH   Isopropyl Alcohol

MGD   Million Gallons Daily

MW   Mega-Watt

ML/day   Million Liters per Day

NTU   Nephlometric Turbidity Units

SU   Standard Units

TPWD   Texas Parks and Wildlife Division

USGS   United States Geographic Survey

WSU   Washington State University

I. INTRODUCTION

Steam electric generating power facilities produce the majority of electricity used in the United

States with a large percentage of these power plants using a once-through cooling water process.

Water is withdrawn from a body of water, such as a river or reservoir, pumped through

condensers to provide cooling and condensation of waste steam by heat exchange, and then

discharged back into the same or a nearby water body (Veil et al. 2002). Impingement occurs

when larger organisms are retained on the traveling water screens located at the entrance of the

intake structure (Lohner et al. 2000; Dey 2002). Entrainment takes place when smaller aquatic

organisms such as fish eggs, juvenile fish, fish larvae or shellfish larvae pass through the intake

screens and enter the cooling-water circuit. There are concerns that adverse environmental

impacts may result if aquatic organisms enter the cooling water intake structure (CWIS) and

become impinged or entrained (Lohner et al. 2000).

During impingement, organisms may be subject to gill compression leading to suffocation and

other mechanical damage such as scale loss or skin lacerations (Hadderingh 1979). However,

recent studies have shown that a significant number of impinged fish may have pre-existing

diseases that may have made them susceptible to impingement (Baker 2007).

Due to the concerns over the potential effects of impingement and entrainment losses, the Clean

Water Act (CWA) Section 316(b) requires that the U.S. Environmental Protection Agency (U.S.

EPA 1977) regulate the location, design, construction and capacity of CWISs so that the structures reflect the best technology available (BTA) for minimizing adverse environmental impact (U.S. EPA 1977, Super and Gordon 2002). Under CWA Section 316(b), the EPA categorizes facilities that use cooling water into one of three groups, with corresponding rules associated with each. The rules (promulgated as "phases") for each group are based on when the facility was constructed, the type of operation for which cooling water is used (e.g., electricity generation, refining, pulp and paper production), and the amount of surface water withdrawn for cooling.

Specifically, the Phase II Rule published on 9 July 2004 (since remanded as subsequently discussed) applied to existing facilities that, as their primary activity, generate electric power, withdraw ≥189.3 million liters (50 million gallons) per day, and use 25% or more of that water for cooling purposes. The 2004 Phase II rule required existing facilities to reduce impingement mortality by 80% to 95% from a calculated baseline where the impingement mortality would hypothetically occur if the facility had a shoreline, near-surface intake with a standard 9.5 mm (0.4 in.) mesh traveling screen (U.S. EPA 2004).

However, facilities using closed-cycle cooling were considered to have BTA for minimizing impingement (U.S. EPA 2004) and entrainment. Also, facilities that had through-screen design velocities of < 0.5 fps were considered to have BTA for impingement only. In addition, the Phase II Rule required facilities located on the Great Lakes, tidal estuaries, or small rivers (power plant cooling water withdraw >5% of the mean annual flow) to reduce the number of entrained aquatic organisms by 60% to 90% from a calculated baseline (U.S. EPA 2004).

Approximately one-third of the existing power plants in the U.S. subject to the Phase II rule withdraw cooling water from freshwater reservoirs or large rivers. These power plants would have been subject only to impingement reduction evaluations (Federal Register 2002).

On 25 January 2007, the Second U.S. Circuit Court of Appeals remanded several provisions of the Phase II Rule back to the U.S. EPA (Riverkeeper, Inc. v. U.S. EPA, No. 04 - 6692, 2d Cir. 25 Jan. 2007). As a result, the U.S. EPA suspended the entire rule and is in the process of rewriting it to comply with the Second Circuit's decision (Federal Register 2007).

Attempts to reduce impingement rates have included the development of exclusion devices that can be grouped in one of two categories: physical and behavioral. Physical devices typically surround an intake structure and physically block the entrance into the CWIS.
Physical devices used to reduce impingement rates include: wedge wire screens, barrier nets, diversion dikes and deep water intakes. Other physical methods that can be employed to reduce impingement rates include: flow reduction (e.g. variable speed pumps and closed cycle cooling), collection and transfer of live organisms to the source water (e.g. traveling water screens) and modification of existing equipment operation (e.g., traveling screens) times to lower impingement. The use of a fish return channel allows the return of impinged fish to the water body. However, physical methods, such as traveling water screens, have limitations which include occlusion due to the selection of small mesh sizes (EPA 1976).

Behavioral devices are designed to repel fish away from a physical barrier and include: louver barriers, fish elevators, velocity cap intakes, light, sound and electrical devices. Behavioral

devices rely on the swimming ability of the various species to avoid the artificial stimuli. Swimming ability is related to fish size within a species and is highly variable between different species of fish. Swimming ability is significantly affected by temperature with reduced swimming ability corresponding to the colder winter months (EPA 1976). Parasitized, diseased or unhealthy fish have been shown to have decreased swimming ability or aberrant swimming behaviors (Barber et al. 2000, Seppälä et al. 2004). Any adverse effects parasites have as they invade, move around or grow inside a host may have consequences that affect growth, survival and reproductive fitness. Behavioral changes affected through pathology, physiological imbalance or general malaise can affect choice of habitat, schooling ability and the ability to resist current. The parasites also use host resources to exist and the host, in ridding itself of the parasites, may expend resources needed for survival (Barber et al 2000).

From January 22, 2007 to August 15, 2007 a screen rotation study was conducted at Plant Barry at Bucks, Alabama to determine BTA for the CWIS. The objective of the study was to assess if intermittent operation of traveling screens increased impingement versus traveling screens in constant operation. Concurrently, from June 01, 2007 through August 15, 2007, a survival study was conducted using impinged blue catfish (*Ictalurus furcatus*) and channel catfish (*Ictalurus punctatus*). A health assessment determined the prevalence of parasites and pathogens in the impinged catfish at the time of impingement, at the time of death in the study and the 7-day survivors of the wild population and two sets of controls. One set of controls was used to assess mortality rates of non-impinged catfish. The other controls were used to assess the effects caused by the capture gear itself.

The overall objectives of this study were to: 1) determine the effect varying operation schedules of rotating screens had upon impingement rates, and 2) evaluate the health and survival rates of impinged blue catfish and channel catfish.

II. LITERATURE REVIEW

Many advances have been made in technology to physically deter fish from CWIS and modern designs of traveling screens that reduce the effect of impingement on aquatic organisms. There have been no studies on the effect of intermittently operating traveling screens versus keeping the screens in constant operation. There are a large number of studies on the survival of impinged organisms. The types of studies and the objectives have become standardized. But, because of variation in rivers, seasons, fish populations, and chemical variables, studies have to be performed at each site before survival rates can be estimated. There have been very few impinged fish health studies at CWIS due to the difficulties of preserving samples, expertise and lab facilities.

Screen Rotation Study

Data from laboratory, prototype, and full-scale studies of modified traveling screens have demonstrated that survival of fish is specific to species, life stage, and site, making it difficult to estimate survival of species that have not been tested with modified traveling screens. Intra-specific differences in survival observed among study sites also make it difficult to project an accurate and reliable estimate of survival at a new site where the screens have not been previously evaluated. Many studies of impingement survival at power plants have been performed. EPRI (2003) published a summary of 35 survival studies located in 15 different states

and the province of Ontario. The studies covered three major types of traveling screens and the majority of the studies were conducted to evaluate a modification to enhance fish survival.

One lab study conducted for EPRI (Black et al. 2006), evaluated the injury and mortality caused by a modified travelling screen using 10 species of freshwater fish and 3 water velocities. Survival rates in the 48-hour study exceeded 95% for all species and velocities tested. Fish length played a significant role in several of the species and as the fish grew in length survival rate increased along with a decrease in injuries and scale loss. There was a tendency toward greater survival at lower velocities but it was only significant for one species. Additional tests were conducted on channel catfish (*Ictalurus punctatus*), fathead minnow (*Pimephales promelas*) and golden shiner (*Notemigonus crysoleucas*) to assess the effect of longer durations of impingement on survival, injury and scale loss. Longer periods of impingement resulted in higher rates of mortality, injury and scale loss.

Yet, gaps remain in the available data and need to be filled before the data can be used to make accurate predictions of survival at cooling water intake structures (CWIS) where this technology is being considered to meet the 316(b) performance standard.

Regulations and technology change over time creating the need to balance BTA with what is economically viable to industry (EPA 2002). Many new technologies are considered BTA, but retrofitting power plants may be prohibitively expensive, reduce the efficiency of the power plant and, in the end, have a negligible effect on the aquatic population. Varying screen operation

times is a potential use of existing technology to comply with the 316(b) regulations requiring reduction of impingement.

Currently the belief is that constantly rotated traveling screens have lower impingement rates when compared to intermittently operated screens (EPA 1976). Studies (King et al. 1978, Tatham et al. 1978) have shown that reduced time between screen washes corresponds with a significant increase in organism survival. Debris build-up against a stopped screen may provide shelter from predation and current and some species may forage in the debris (EPA 1977). As fish grow, their ability to swim faster and longer increases. Small fish can become exhausted while trying to maintain orientation in the current and may become pressed against the screens due to plant inflow. In all cases, when the traveling screen is operated again, the fish population in the debris could then be entrapped increasing impingement (EPA 1976).

Survival Study

Many traditional traveling water screens are designed to spray debris from the screens into a collection pile. Periodically the refuse is removed to a disposal site (EPA 1976). Survival studies combined with impingement data have been keystone measurements of whether modifications to the traditional disposal system are warranted or economical. If few fish are being impacted or survive impingement, the cost to install fish returns to the CWIS may not be warranted.

One study of modified Ristroph screens (Black et al. 2006) evaluated injury, scale loss and survival of 10 species of freshwater fish exposed to the impingement and collection processes.

Survival for all 10 species was greater than 95%. In most cases water velocity was not correlated to survival. For many species survival was significantly correlated to fish length, with larger fish surviving better than smaller fish. Tests indicated impingements lasting more than 6 minutes significantly increased mortality. Many fish actively respond to being impinged by trying to free themselves. The force of the water holding them to the screens combined with the movement of the fish can result in de-scaling, eye injury, and other soft tissue injuries. Impinged organisms also encounter the spray wash system that removes debris from the screens which may be another source of physical abrasion. Finally the presence of debris may result in abrasive contact throughout the impingement process (EPA 1976).

Because the environment is rarely static, impingement can be influenced by a variety of diurnal, seasonal, and periodic events. Influential factors that may affect impingement rates are temperature, time of day, wind action, dissolved oxygen, turbidity, water velocity, habitat type, life stage, overall health, disease prevalence and spawning events.

Turbidity is defined as an optical property of water wherein suspended and dissolved materials such as clay, silt, small organic and inorganic matter, plankton, and other microscopic organisms cause light to be scattered and absorbed. Turbidity likely influences impingement when lights are used as deterrents (Baker 2008, McIninch and Hocutt 1987, Taft 2000). Increasing turbidity would diminish the ability of fish to see and avoid the travelling screens. Diurnal factors can also influence the impingement of specific species. A study by Swanson et al. (2005) using delta smelt (*Hypomesus transpacificus*) to observe the time of contact and the effect of contact with wire mesh screens determined that contact with the screens rose significantly in darkness. Simple

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contact with the screens was not fatal but if a fish became injured by contact with the wire mesh it was always fatal.

Low temperatures or a change in temperatures may cause fish mortalities that drastically increase impingement. Cold temperatures adversely affect clupeid species. Studies conducted with threadfin shad (*Dorosoma petenense*) at southeastern power plants has shown significant increases in impingement rates as the temperature drops below 15°C (Griffith and Tomljanovich 1975, Loar et al. 1978, McLean et al. 1985). Cooler temperatures also cause other temperate water species to be more sluggish and have reduced swimming ability (Griffith and Tomljanovich 1975, Grimes 1975, Hoyt 1979). Low temperatures have been shown to cause a loss of equilibrium, disorientation, and mortality in juvenile freshwater drum (*Aplodinotus grunniens*) (Bodensteiner and Lewis 1992). With reduced swimming abilities, alewives and other species lose the capacity to effectively avoid behavioral deterrents and thus, reduce the deterrent's efficiency.

Wind and wind-induced effects have been correlated to fish impingement (Lifton and Storr 1978). When the fetch of a lake is large, wind can have significant effects on fish location. Lifton and Storr (1978) also found that fish could be passively moved by wind-created currents toward intake structures leading to increased impingement rates. They also concluded that increased turbidity associated with wind action caused fish to be at higher risk to impingement due to decreased visibility.

Impingement also tends to vary inversely with dissolved oxygen (DO) concentrations (Lewis and Seegert 2000). Decreases in DO concentration generally stimulate fish to search for higher concentrations in adjacent areas. The search for higher concentrations of DO may expose fish to other variables such as low temperatures or cause them to be displaced closer to the CWIS (Bodensteiner and Lewis 1992). Fish with reduced physical conditions resulting from low DO or low temperature stress may become subjected to suboptimal conditions rendering them incapable of producing the avoidance reactions. These impaired reactions would cause the deterrent system to become less effective (Bodensteiner and Lewis 1992, Knights et al. 1995).

The effectiveness of behavioral technologies has been evaluated through a variety of methods in field and laboratory settings. Passive techniques are generally used to monitor fish in a laboratory setting. The majority of behavioral guidance studies use visual observations and video cameras as the primary methods for evaluation under laboratory conditions. The predominant method of actively evaluating the effectiveness of behavioral technologies at power production facilities has been through impingement rate measurements. When measuring impingement rates, fish are first collected from the intake screening device, usually a rotational screen. The fish are then physically counted and/or examined to the researcher's specifications. After measurements have been taken, the fish can be either returned to a safe location in its environment or discarded.

Physical characteristics of the CWIS affect the survivability and health of impinged fish. Mechanical injury can occur at the fixed structure, the rotating screens, in the return chute or by debris in the water. Pathogens can gain entry into the fish through mechanically caused lesions

incurred during the impingement process. Finally, stressors such as exposure to air, immobilization on the screens and high pressure jets could cause decreased immune response.

If rotating screens are inactive, small fish in the influence of the CWIS, may seek debris buildup as defensive habitat from predation or as a buffer from fast flowing water. Screen activation would then increase mortalities by trapping fish in the debris (EPA 1976). Hanson et al. (1977) reported the accumulation of debris on intake screens can affect impingement by serving to entrap and entangle fish and altering the hydraulic flow and intake velocity.

Barge traffic mortalities (shear and propeller entrainment) can disorient, injure or kill fish (Wolter and Arlinghaus 2003) near the CWIS hydraulic zone of influence. From the available investigations of navigation forces, it has been concluded the direct entrainment of individuals through the propeller zone of vessels is the main source of navigation-induced fish mortality for eggs, juveniles up to 32 mm total length (Killgore et al. 2001), and adults (Gutreuter et al. 2003) due to shear stress created by towboat propellers. Fish injured in the propeller zone of barges can become trapped on the screens and counted as impinged.

Physical factors such as river discharge, elevation of the water body (stage) and the amount of debris influence impingement rates. Mathur et al. (1977) found the number of fish impinged increased as Susquehanna River discharge increased and pool elevation decreased. Suspended sediments and debris loading may cause increases in impingement rates (Hanson et al. 1977).

Species-specific behavior can determine how effective a deterrent will be for a given location and targeted species or suite of species. For example, the greatest avoidance to strobe lights was shown to be above 300 flashes per minute (Sager et al. 2000). Flash rates below 200 per minute were found to be significantly less effective than higher flash rates. Given the wide range of hearing capabilities among species, appropriate sound frequencies should also be considered when choosing sound as a deterrent. In addition, sufficiently elevated sound pressure levels are necessary to cause a deterrent response at these appropriate sound frequencies.

Health indicators of impinged fish, such as gill condition, lesions, presence of parasites, bacterial infection and length to weight ratios can be evaluated. These health characteristics can indicate fish that were impaired, moribund or dead prior to being impinged. The ability to sample the health of impinged fish quickly and accurately has been a primary difficulty.

Previous fish health studies at Alabama power plants (Baker 2007, Knight 2008) found a significant number of impinged fish sampled were diseased or dead. Critical evaluations of the survival after impingement for impinged fish by power plant traveling screens had not been done. Time of death, prior to or because of impingement, is a critical factor needing to be understood. Fish that have died in the open water prior to entering a CWIS zone of influence bias evaluation of technology designed to reduce fish impingement. Current practice assumes all impinged fish were the direct result of the CWIS. Studies trying to account for dead, morbid or diseased fish being pulled into intakes thus far have been lacking, especially in relation to survival after the impingement event.

The current impact that impingement has on ecosystems and the fish community is unknown. Fish impingement rates depend on the time of year, water demand and population densities of the organisms in question (Spigarelli et al. 1981). Natural fish mortality can be extremely high depending on the species and season. It is certain that many of the fish impinged during the winter would not survive to reproduce due to starvation, disease or predation. According to the U.S. EPA (2002), impingement mortality occurs in several ways including starvation, exhaustion, asphyxiation and physical damage.

Swimming performance of fish is characterized by the relation of swimming speed and endurance time and has been classified by Webb (1984) and Beamish (1978) into three categories: sustained, prolonged and burst swimming. A fish's swimming performance is species specific and increases as the size of the fish increases (Hammer, 1995; Domenici, 2001). Current at the CWIS can exceed the sustained ability of small fish and draw them into the screens.

Impingement studies of alewives indicated impinged fish had significantly lower body and liver weights. This is suggestive of exhaustion of the impinged alewives (Rodgers, 1995). Exhaustion can occur from prolonged swimming at speeds higher than normal cruising speed (Beamish, 1978). Also, fish in poor health due to breeding stress, disease and parasites may not be able to escape the current at the intake structure (Barber 2000).

Sprengel and Luchtenberg (1991) showed infestations of microsporidians, endoparasites and muscle infesting nematodes reduce the swimming speed of European smelt (*Osmerus eperlanus*)

and European eel (*Anguilla anguilla*). In European smelt, adults with the parasite *Pseudoterranova decipiens*, were found to have slower swimming speeds. The decreased swimming ability increased the number of contacts with mesh screens at power plants and increased the external damage to impinged smelts. CWIS selectively impinge parasitized smelt and reduce the number of parasitized smelt in the population.

The Mobile River at Plant Barry experiences tidal fluctuations. During high tide and low flows, the water has the capability of running up stream. The average daily water flow at Plant Barry on October 16, 2006 was -4,230 ft3/sec and on September 22, 2007 was -775 ft3/sec (USGS 2006, USGS 2007). During low-flow periods, high tide can also cause a gravity-fed salt wedge to move upriver under the fresh water (Maes et al, 1998). It also introduces the problem of warmer water from the outflow downstream migrating upstream to the intake and recirculating through the power plant cooling system.

Immunological responses of fishes can be significantly affected by temperature (Avtalion et al 1973, Casterlin and Reynolds 1977). The upper critical levels for fish are species-specific and vary greatly. If the temperature becomes too high the fish will experience hyperthermia and stress. Theoretically, a fish moving into a area warmer than the general environment could affect hormone or enzyme activity and increase the fish survival rate. Higher temperatures could also increase phagocytosis in the early stages of infection (Ellis 1981). Survival of infections can be affected by relatively short term temperature increases (Covert and Reynolds 1977). Data also shows the febrile response is effective even if the elevated temperature is not constant or indefinitely maintained. Avtalion et al. (1973) demonstrated fish developing the immune

response at elevated temperatures retained an antibody memory response at lower temperatures.

This suggests the febrile response is most useful in naïve fish and early immune response.

III. METHODS

Site Description

Barry Steam Plant (Plant Barry; Bucks, AL) is owned and operated by Alabama Power Company and has a nominal rating of approximately 2,625 MW. Five coal-fired Units (Units 1-5) can generate up to 1,525 MW and use once-through cooling water. Additionally, Plant Barry has two combined cycle electric generating Units (Units 6-7) with a heat recovery steam generator. These combined cycle Units use closed-cycle cooling and have a combined nominal rating of approximately 1,100 MW. The plant is on the Mobile River (Mobile County, AL), approximately 49 km (30.5 mi) upstream from the confluence of the river with the Gulf of Mexico (Figure 1). River stage is monitored by USGS 02470629 (Mobile River at River Mile 31.0 at Bucks, AL) located at Latitude 31°00'56", Longitude 88°01'15" NAD27 (Figure 2). The Mobile River at this location is fresh water. River stage is influenced by tidal fluctuations and, during high tide and low flows, the water has the capability of running up stream. During low-flow periods, high tide can also cause a gravity-fed salt wedge to move upriver under the fresh water (Maes et al. 1998).

Two CWISs, one for Units 1-3 and one for Units 4-5, are used to withdraw cooling and service water for the five coal-fired units and makeup water for the two combined cycle generating units. Both CWISs are located within a man-made barge canal that is adjacent to the main river channel

and separated by less than 60 meters (<200 ft) (Figure 2). At low tide the canal has a maximum depth of 5 m (17 ft) and the Mobile River at the junction with the intake canal has a depth of 13 m (42 ft) and a width of 198 m (650 ft).

The CWISs are equipped with floating debris buffers, trash racks, and traveling screens to remove the high volume of debris from the Mobile River (Figures 3, 4). The pontoon-supported debris buffer consists of a series of floating pontoon structures with vertical rods extending to a depth of 2 m (6.5 ft) and spaced 20 cm (8 in) apart. The pontoons are located about 6.1 m (20 ft) upstream (i.e., north) of the trash racks (i.e. in front of the intake structure). Six traveling screen bays for the Units 1-3 CWIS and five traveling screen bays for the Units 4-5 CWIS are located immediately downstream of the trash racks. Each screen bay is about 3.4 m (11 ft) wide and houses a stainless steel trash rack with 8.9 cm (3.5 in) x 2.1 cm (0.8 in) bars and spaced 10.2 cm (4 in) on-center with 8 cm (3 1/6 in) clear openings. The trash racks are cleaned on a daily to weekly frequency depending on the extent of debris blockage. Debris is removed manually from the Units 1-3 trash racks. However, a mechanical Backett-Bosker trash rake is used to remove debris from the Units 4-5 trash racks. The design through-screen velocity using normal water surface elevation of 0.6 m (2 ft) above mean sea level is approximately 0.5 m/s (1.66 ft/s) and 0.6 m/s (1.98 ft/s) for Units 1-3 and Units 4-5, respectively. A high-pressure front spray wash system is used to remove fish and debris from the screens. This wash water then flows down a concrete sluiceway into a basket which collects the debris for disposal.

At full load, Units 1-3 withdraw 1,772 ML (468 MGD) and Units 4-5 withdraw 2,532 ML/day (669 MGD) of cooling water from the intake canal. Water passes through the trash rack and into

the plant via the intake structure underflow opening. Screened cooling water for each CWIS then flows into an intake tunnel that conveys water via circulating water pumps to the condensers for cooling.

Screen Rotation Study

The screen rotation study evaluated the effect of periodically stopping the rotating screens compared to constant operation of traveling screens on impingement at Plant Barry from January 22, 2007 to August 15, 2007. Both intakes at Plant Barry were used to produce paired data samples where the control CWIS was in operation and the treatment CWIS was stopped during the sampling period. Under normal operating conditions the rotating screens at both CWIS are in operation all of the time. The environmental parameters of water temperature, dissolved oxygen, pH, specific conductivity and turbidity were monitored to ensure that these variables were not interfering with the evaluation of the study.

There were three seasonal sampling phases: winter, spring and summer. There were two breaks between the three phases, from March 15 to April 08, 2007 and May 31 to June 17, 2007. These periods were used for initial data evaluation and coordination with the plant engineers to increase the sampling times. In the winter phase 3 daily 4-hour samples twice per week were scheduled to coincide with a day, dusk, and night sample. In the spring phase the screens were stopped for 3 daily 6-hour sample periods twice per week again timed so that a day, dusk, and night sample were taken. In the summer phase the sample periods were extended to twice daily 11-hour

samples twice per week. The summer samples were timed to provide a day sample and a night sample. The CWIS units were alternated weekly between control and treatment schedules.

The treatment CWIS rotating screens were stopped during the sampling period for 4 hours, 6 hours or 11 hours depending on the phase of the experiment and then rotated for 30 minutes to collect the sample. This added 30 minutes to every treatment sample time to insure collection of all organisms and debris trapped on the screens. The control CWIS sample was collected continuously throughout the period. The CWIS units were alternated weekly between control and treatment schedules. The traveling screens at Plant Barry are normally kept in operation at all times due to the high debris load. The length of the sampling was increased over the length of the experiment as the operators of Plant Barry evaluated whether there was a negative impact on the flow of water from debris buildup on the screens that were stopped for the experiment.

Since the experiment involved changing the operations of existing rotating screens, no modifications to either CWIS was needed. Aluminum baskets composed of 9.5 mm mesh screen (Figure 5) were designed to fit the existing screen-wash trash pits to collect the samples. For both the treatment and control CWIS the baskets were placed in the discharge flume from the screens at the beginning of the sample period.

Impingement sampling was used to determine any difference between treatment and control schedules. All fish and shellfish (clams, freshwater shrimp and snails) collected during each sampling event were backwashed off the traveling screen basket to the sampling basket. Organisms were removed from the sampling basket, sorted, identified to species, enumerated,

and weighed. Total count and weight were recorded for each species. Severely decayed animals were discarded and not included in the sample.

Impingement numbers and weights were standardized to fish impinged per hour for the sample period. When Unit 5 went off-line it reduced the flow at that CWIS to 35 percent of the full operating inflow. This both reduced the current at the intake and reduced the hydraulic zone of influence at the CWIS for Units 4-5. All of the impingement numbers for Units 4-5 during the period that Unit 5 was down were divided by 0.35 to adjust the impingement numbers closer to the impingement would have been if the CWIS was as full operating flows.

When screens became inoperable but not stop logged, the water continued to flow through the screen with the potential for impinging organisms. Any organisms impinged while the screen was inoperable were not collected. So, a screen adjustment factor was applied to the number and weights of organisms to account for organisms not recovered from inoperable traveling screens. The counts and weights for organisms recovered from the operating screens in the sample were then divided by the number of functional screens over the total number of screens with water flow.

Water quality samples were collected at both intakes during each impingement sampling event. The water sample was collected directly in front of the CWIS from the water surface using a 38 L bucket. The water in the bucket was then immediately sampled. Water quality parameters recorded included were water temperature (ºC), pH (SU), dissolved oxygen (mg/l), turbidity (NTU) and specific conductance (µS/cm). Water quality measurements were taken from surface

water samples directly in front of the trash racks. Water in front of the CWIS was extremely turbulent and assumed to be representative of the whole water column within the intake forebay area. A YSI 85 meter (Yellow Springs Instruments, YSI Incorporated, Yellow Springs, OH) was used to measure dissolved oxygen and temperature. A LaMotte 2020 (LaMotte Company, Chestertown, MD) was used to measure turbidity. A WTW 340i meter (Wissenschaftlich-Technishe Werkstatten GmbH, Weilheim, Germany) was used to measure specific conductance and pH.

River stage and discharge data were obtained from the United States Geological Survey (USGS) gauge (02470629) located at the debris buffer on CWIS Units 1-3. In addition, the CWIS flow volume (m³/s), through-screen flow velocity (m/s) and the number of circulating water pumps in operation were recorded for each collection period. River stage, amount of surface area of the screen, and the volume of water withdrawn from the CWIS were used to calculate the CWIS flow velocities.

Survival Study

An impingement survival study was conducted at Plant Barry at the CWIS for Units 1-3 from June 01, 2007 to August 15, 2007 utilizing blue and channel catfish. Sample collection for the study was conducted daily with the exception of Tuesdays and Thursdays to avoid conflicting with the screen rotation study described above. Duration of collection times of fish from the basket varied from 15 minutes, 30 minutes and 60 minute intervals depending on the amount of debris and the number of catfish being collected.

A sample collection basket of plywood bolted to a reinforcing framework of 2" by 4" studs (Figure 6) was designed to hold water and minimize turbulence. Flow into the tank was controlled by a hinged shunt that when in place would block water flow into the basket. Overflow holes were built into the basket to prevent overflowing the sides of the tank and losing impinged samples. A drain was placed in the bottom of the tank to allow the tank to drain slowly once the flow of water was removed from the basket.

Live, impinged catfish were placed in water containing a 20 mg/L of MS-222 (Finquel®) buffered with equal parts sodium bicarbonate. All dead catfish suitable for necropsy were placed on ice and were diagnosed for disease. A suitable fish was determined by gross observation of the gills, a red gill indicates a freshly dead fish, eyes were checked for clarity and the skin was checked for general necrosis. Necrosis around a lesion is not uncommon in fish but is localized around the lesion and did not disqualify a fish. Severe degeneration of the skin along with the paleness of the gills and absence of eye clarity disqualified fish for use. All usable fish were checked for lesions, parasites and bacterial infections. All impinged organisms were recorded to the lowest practical taxa and whether live or dead. All blue catfish and channel catfish were retained for the survival and health studies and the other impinged organisms were disposed of to prevent re-impingement.

Live catfish from the impingement collection tank were moved to a temperature-controlled (21°C) trailer and placed individually into 1 of 24, 57 L flow through aquaria (Figure 7). At the trailer, the fish were assigned a number and data recorded for time of capture, time in collection

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basket, water quality and time entering the aquarium. Each aquarium had an independent air and water supply. River water was pumped directly from the CWIS so the fish were subjected to any water quality changes present in the CWIS. Water entered the aquaria at the top from the manifold and was removed from the bottom through a stand pipe. Water flow to the aquaria was checked every two hours when the fish were inspected for mortalities. No fatalities were attributed to these temporary stoppages.

Health indicators of impinged fish were determined over the course of the survival study impinged fish. Fish necropsied were assigned one of four categories: Dead Impinged, Live Impinged Initial, Impinged Latent Mortality and Impinged Latent Survivor. The Dead Impinged group consisted of the freshly dead (showing red or pink gills) catfish retrieved from the sample basket. The Live Impinged Initial category consisted of live catfish larger than 20 cm (too large for the aquariums) and every fifth live impinged catfish to establish health at impingement. The Impinged Latent Mortality were impingement survivors that were placed into aquaria but died before 7 days. The Impinged Latent Survivors were impingement catfish that survived the 7 days and then were necropsied to assess their health.

The aquaria water quality parameters recorded were water temperature, turbidity, dissolved oxygen, salinity and operational traveling screens. Data was recorded daily, when adding fish to aquaria, and during mortality checks.

Control fish for the experiment were channel catfish that had been raised indoors at the E. W. Shell Fisheries Center, North Auburn Unit. The controls came in two size ranges 50 mm to 95

mm and 133 mm to 204 mm. The control used was the closest to the length of the impinged fish. The control fish were monitored and fed daily. The control fish were maintained in a holding tank in the study mobile laboratory connected to the same water and air supply as the aquariums. Tank and basket controls were assigned the number of the fish they were matched with. There were two control treatments consisting of tank controls and basket controls. Tank controls (non-stressed) were removed from the control holding tank and placed in an aquarium at the same time as the impinged catfish. Basket controls were dip-netted, fin clipped for marking purposes and placed in a transport bucket. The basket control fish were placed in the impingement capture basket once it was full of water. The basket control was subjected to the turbulence and abrasion an impinged fish would encounter by entering the capture tank at the start of a 1-hour sample period. After 1-hour, the basket control was then removed with any additional impinged fish, placed in a bucket with buffered MS-222, and transferred to the holding aquaria.

Control fish were fed daily in the large common holding tank. Once fish entered the study aquaria they were fed 3-2 mm pellets of standard catfish feed every other day. This was done to avoid the effects of starvation while in the study.

From June 01, 2007 to July 08, 2007 fish were observed for 14 days. During this time, all of the fish that survived the first 7 days were alive at 14 days. In early July, all 24 aquaria were in use. To facilitate more fish being sampled, beginning on July 08, 2007, the study fish were held in the aquaria for only 7 days before being euthanized to increase the number of fish sampled in the study.

Study fish were inspected every 2-hours. Directly prior to observation, temperature, dissolved oxygen specific conductivity and pH were recorded. Since the air and water supplies came from common mains, the assumption was made that the conditions in one tank would be the same as the conditions in any other tank.

The aquaria were composed of a translucent, white plastic. The turbidity of the water was also high enough to prevent easy observation of the bottom of the aquaria. To determine if fish in the tanks were alive, a light was shined through the aquaria to allow direct observation of the fish. If no movement was observed, a net was then inserted into the aquarium to induce the fish to move. If no detectable motion was observed the fish was netted and observed directly to determine morbidity. Dead fish were removed from the aquaria, individually bagged, placed on ice and necropsied. Times of death and fish identification were recorded. Aquaria were sanitized with sodium hypochlorite when a fish was removed to prevent cross-contamination by parasites and bacteria. All samples intended for necropsy were placed in plastic bags then put onto ice. Fish were designated as live or dead when removed from the capture basket. All fish were then measured to the nearest millimeter and weighed to the nearest gram. Gross observations of mechanical damage, lesions or parasitism were measured, described and recorded.

A skin scrape was made from around the head, tail and any lesions and placed on a glass microscope slide. A small sample of the fish gill filaments was obtained, placed on a slide, and a second gill sample was streaked onto Hsu-Shotts agar supplemented with tobramycin (Griffin 1992, Decostere et al. 1997) for detection of *F. columnare*. Samples were observed on 10X, 20X, and 40X for crustaceans, monogenetic trematodes, protozoans, fungi and presence of long rods

characteristic of *F. columnare* (Griffin 1992). The presence of fungi was decided if hyphae were present.

Samples from the kidney and liver were aseptically obtained and streaked onto brain heart infusion agar (BHI). Additional samples from the kidney were streaked onto Hsu-Shotts media supplemented with tobramycin (Griffin 1992, Decostere et al. 1997). Once the agar plates were inoculated they were placed in a $30°C$ incubator. The abdominal cavity was then examined for gross internal lesions, parasites or abnormalities of the internal organs. If internal parasites or granulomas were present, the sample was observed under the dissecting microscope for classification of the parasite. Finally, samples of gill, trunk kidney and liver were preserved in Bouin's Fixative or 10% buffered formalin for additional histological verification if needed.

All bacterial plates were incubated for a minimum of 24 hours at $30°C$. Hsu-Shotts agar was incubated for 48 hours and then inspected under the dissecting scope for yellow-orange, rhizoid growth. BHI agar was incubated for 24 hours and then inspected for growth. Plates with less than 5 colonies were considered to not have significant growth. When BHI agar plates contained multiple bacterial morphologies, the dominant colony type was re-streaked for isolation and identification.

To presumptively identify the bacterial isolates, a dichotomous key was designed based upon biochemical and phenotypic characteristics to presumptively identify the pathogen to the lowest possible tax. Fish isolates were initially assessed using the potassium hydroxide string test (Powers 1995) as an alternative to the gram stain, and cytochrome oxidase tests (Brenner et al.

2001; Buller 2004) to determine the appropriate biochemical tests needed. Further biochemical tests used were glucose metabolism (Furuwatari et al. 1994, Brenner et al. 2001), d-mannitol utilization (Abbott et al. 2003), sensitivity to vibriostat (Holt et al. 1994, Brenner et al. 2001, Buller 2004), growth in 0% NaCl (Furuwatari et al. 1994, Austin and Austin 1999, Brenner et al. 2001) and sensitivity to novobiocin (AFS-FHS 2005). When it became impractical to identify the samples in the field, isolates were frozen and stored in the -80ºC ultra-freezer for subsequent identification. All of the biochemical tests and the expected results for the pathogens expected in the study are shown in Table 1 and Table 2.

Samples to be frozen were cultured in BHI broth for 24 hours. Then 0.5 ml of glycerol was added to the 5 ml of broth culture and vortexed for 30 seconds to facilitate the glycerol coating the bacteria. The 2.0 ml of culture was placed in cryotubes (Denville Scientific, Inc., Metuchen, NJ). Samples were then flash frozen in liquid nitrogen and stored at -80ºC.

Statistical Analyses

All of the statistics in this project were analyzed using the Statistical Analysis System software (SAS Version 9.2, SAS Institute, Cary, North Carolina). The screen rotation study used PROC GLM procedure to analyze total impingement and individual species impingement. The GLM procedure was used to take advantage of the ANOVA with repeated measures. The study was blocked by seasons. The GLM procedure was run as a 2x3 table to account for differing numbers of samples per season. The Tukey's studentized range test (HSD) was used to determine

statistically significant differences between treatments and sample times. P values of ≤ 0.05 were considered statistically significant.

Survival analysis is a class of statistical methods for studying the occurrence and timing of events and is most often applied to the study of deaths (Allison 1995). The Survival study was analyzed using SAS PROC LIFETEST. The initial data provided by LIFETEST is the estimated distribution of failure times using the survival distribution function (SDF) to describe the lifetimes of the treatments. Then the survival functions are compared using the Savage (log rank) test or Wilcoxian test to determine whether significant differences exist and produce approximate Chi-Square statistics. All fish that survived 7 days were right censored and not considered in the analysis. P values of ≤ 0.05 were considered statistically significant.

The Health study was analyzed using PROC FREC to determine the probabilities using a Chi-Square analysis of the data. The treatment groups were analyzed for differences in presence of lesions, parasites and bacterial infections. P values of ≤ 0.05 were considered statistically significant. Individual pairwise comparisons were performed when significant differences were indicated within groups.

IV. RESULTS

Screen Rotation Study

Over 29,500 fish and 6,000 non-fish organisms (e.g. shrimp, muscles, and Asian clams) comprising 46 species (Figure 8) were collected during the Screen Rotation Study. Nine species (Figure 9) comprised more than 96% of the total impingement and accounted for 91.1% of the total weight impinged. The median daily impingement during the study was 1877 organisms per day, with a mean daily impingement of 1691 organisms. Impingement varied greatly by season, species, CWIS and treatment over the study (Tables 3, 4). During the study, 122 paired impingement samples were obtained. To account for seasonal and diurnal variability, screen rotation samples were evaluated on a weekly basis, whereby the two different treatments (intermittent or constant operation) would be paired and evaluated during individual weeks. The sample design also produced equal pairs of samples from each intake allowing comparison of species between intakes. The median impingement rate was used to differentiate between high and low impingement rates for each species.

The majority of winter impingement consisted of freshwater species at Units 1-3 and was comprised primarily of young-of-the-year (YOY) or over-wintering fish. Spring phase had very low fresh water impingement. In the summer phase threadfin shad at Units 4-5 was the only fresh-water species impinged in high numbers. While very few eurihaline fish were impinged in

the winter phase, euryhaline species showed the highest seasonal impingement during the spring or summer phases. Summer impingement rates were highest at Units 4-5.

Intermittent operation of traveling screens (Table 3) did not significantly increase overall impingement ($p \leq 0.1469$; Figure 10) or overall impingement of the major freshwater species threadfin shad (*Dorosoma petenense*) ($p \leq 0.2114$; Figure 11), blue catfish ($p \leq 0.3117$; Figure 12), channel catfish ($p \leq 0.7299$; Figure 13), freshwater drum (*Aplodinotus grunniens*) ($p \leq 0.2084$; Figure 14), and gizzard shad (*Dorosoma cepedianum*) ($p \leq 0.7000$; Figure 15). The euryhaline species, bay anchovy (*Anchoa mitchilli*) ($p \leq 0.0194$; Figure 16) and hogchoker (*Trinectes maculatus*) ($p \leq 0.0040$; Figure 17) showed overall significant differences. In the non-fish species shrimp did not show an overall increase in impingement ($p \leq 0.3552$; Figure 18) while the blue crab ($p \leq 0.0001$; Figure 19) showed significantly higher overall impingement with intermittent screen operation. All of the eurihaline species showed seasonal impingement peaks during the spring or summer phases.

The nine most impinged species showed significant seasonal differences (Table 4). When impingement rates were evaluated by intake, bay anchovy ($p \leq 0.0038$), blue catfish ($p \leq 0.0001$), blue crab ($p \leq 0.0010$), channel catfish ($p \leq 0.0005$), freshwater drum ($p \leq 0.0001$) and gizzard shad ($p \leq 0.0001$) showed significant differences in impingement by intake. In the winter the impingement consisted of mostly blue catfish, channel catfish, freshwater drum, threadfin shad and gizzard shad at Units 1-3. The spring impingement was split relatively evenly between the intakes and mainly consisted of channel catfish, bay anchovy, hogchoker and shrimp. The

summer impingement was highest at Units 4-5 and the peak species were threadfin shad, bay anchovy, and blue crab.

The winter phase impinged 82.3% of the total blue catfish, 62.4% of the total channel catfish, 70.6% of the freshwater drum, 95.2% of the gizzard shad, and 39.4% of the threadfin shad. Threadfin shad were an exception, while demonstrating high impingement during the winter, their impingement was highest in late summer and accounted for 54.9% of their total impingement for the study. Bay anchovy impingement was relatively evenly split between spring and summer accounting for 45.6% and 54.3% respectively. Blue crab impingement was extremely heavy in the summer accounting for 90.7% of the total. The shrimp species impingement was heavy in the spring accounting for 92.2% of the total.

The seasonal water quality means for the Screen Rotation Study are presented in Table 5. A pair wise comparison of these parameters (using a Least Significant Difference) showed significant differences between the CWIS in pH ($P \leq 0.0056$), Temperature ($P \leq 0.0003$), and Turbidity ($P \leq 0.0115$). The differences between the two CWIS may be due to the influx of water from the river near Units 4-5 while Units 1-3 are closer to the end of a closed channel. The turbidity inside the channel is likely caused by the coal barges within the channel. The temperature ranged from a low of 8.9 ℃ in January to a high of 34.8 ℃ in August with the coldest temperatures between January 29 and February 15 (Figure 20). The pH ranged from 6.59 to 7.79 with little correlation to season (Figure 24). Turbidity ranged from 2.70 Nephlometric Turbidity Units (NTU) to 69.07 NTU (Figure 23) with a general decrease from January to August with four readings exceeding 30 NTU. Dissolved oxygen varied from a high in January of 11.45 mg/L to a low in August of

6.01 mg/L (Figure 21). Conductivity (Figure 22) increased from January to August ranging from 140 µmhos/cm to 306 µmhos/cm). Dissolved oxygen (P≤0.3398) and conductivity (P≤0.2057) showed no significant differences between intakes.

Survival Study

A total of 169 impinged catfish were captured alive in the sampling basket and categorized as latent impinged in the survival study (Table 6). The sample of 105 impingement survivors consisted of blue catfish and channel catfish. The controls consisted of 64 channel catfish raised at the Auburn University North Station (S-6). The impinged fish 7-day survival rate was 9.5% with survival rates in the two control groups of 90.2% in the aquaria controls and 78.3% in the basket controls (Figures 25, 26). A significant difference (p<0.0001) was found in mortality rates between the control groups and impinged catfish. There was no significant difference in mortality rates between the impinged blue catfish and channel catfish (p≤0.93) or the two control groups (p≤0.09).

Among blue catfish and channel catfish impingement survivors at Plant Barry, 60% died within the first 24-hours post impingement and 90.5% died within 72-hours. After 72-hours, there were no additional impingement mortalities.

Environmental variables monitored in the aquaria include water temperature, dissolved oxygen (DO), specific conductivity and pH and were recorded every two hours and when a new impinged fish entered the system.

Aquaria water temperature varied from 21.8ºC to 34.9 ºC with large fluctuations within the course of a day (Figure 26). The greatest fluctuation was on 23 June 2007 when the water temperature increased from 28.4 ºC to 34.9 ºC from 0640 to 1400. By 2200, eight hours later, the water temperature in the tanks was back to 28.4 ºC.

DO ranged between 5.3 mg/L and 8.31 mg/L with a general decrease of roughly 1.0mg/L over the course of the study. Specific conductivity ranged between 244µs to 360µs during the study. The pH ranged from 6.45 to 7.58.

Throughout the study over fourteen different known bacterial fish pathogens were isolated from internal organs and lesions of both the impinged and control fish (Table 7). Bacterial fish pathogens were found in 91.7% of the dead impinged catfish found suitable to be necropsied and 89.7% of the live impinged fish compared to 31.0% of the aquaria controls and 54.5% of the basket controls. Total bacteria isolated from liver samples of impinged fish were observed to be significantly higher than the aquaria controls in the dead impinged ($p \leq 0.0001$) and in the impinged latent mortality ($p \leq 0.0001$). The basket controls had significantly less liver bacteria than the impinged latent mortality ($p \leq 0.001$). Dead impinged was significantly greater than the live impinged initial ($p \leq 0.024$) but less than impinged latent mortality ($p \leq 0.001$). Live impinged latent also had significantly fewer bacteria than impinged latent mortality ($p \leq 0.0001$).

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For bacterial pathogens present in the trunk kidney, the aquaria controls were significantly lower in the aquaria controls than in the impinged latent mortality ($p \leq 0.0001$). The basket controls were significantly higher than the live impinged initial ($p \leq 0.031$). The dead impinged were significantly higher than the live impinged initial ($p \leq 0.042$) but significantly less than the impinged latent mortality ($p \leq 0.0001$). The live impinged initial was also significantly lower than the impinged latent mortality ($p \leq 0.0001$).

Flavobacterium columnare on the gills was significantly less in the aquaria controls than in the basket controls ($p \leq 0.0031$), dead impinged ($p \leq 0.0001$), live impinged initial ($p \leq 0.0001$), impinged live mortality ($p \leq 0.0001$) and impinged live survivors ($p \leq 0.0017$). Also on the gills the *F. columnare* was significantly lower in the basket controls than the dead impinged ($p \leq 0.0001$), live impinged initial ($p \leq 0.0174$) and impinged latent mortality ($p \leq 0.0028$), but was not significantly different than the impinged latent survivors ($p \leq 0.4382$). In the kidney, there was no significant difference between the occurrence of *F. columnare* in the aquaria controls and the basket controls ($p \leq 0.085$) and impinged latent survivors ($p \leq 0.659$). The aquaria controls had significantly less *F. columnare* than dead impinged ($p \leq 0.0001$), live impinged initial ($p \leq 0.003$) and impinged latent mortality ($p \leq 0.0001$). The basket controls had significantly less *F. columnare* than dead impinged ($p \leq 0.0001$) and impinged latent mortality ($p \leq 0.003$). The dead impinged group had significantly less *F. columnare* than the live impinged initial ($p \leq 0.0001$) but greater than the impinged latent survivors ($p \leq 0.001$). The impinged latent mortalities was also significantly greater than the impinged latent survivor ($p \leq 0.008$).

Prevalence of *Aeromonas veronii biotype sobria* in the aquatic controls was significantly lower than the basket control (p≤0.008), dead impinged (p≤0.050) and impinged latent mortality (p≤0.001) but not significantly different than the live impinged (p≤0.716) and impinged latent survivors (p≤0.309). The occurrence of *A. veronii biotype sobria* was significantly greater in the basket controls than in live impinged initial (p≤0.021) and impinged latent survivors (p≤0.034) but was not significantly different from the dead impinged (p≤0.117) and impinged latent mortalities (p≤0.954). The frequency of *A. veronii biotype sobria* in the dead impinged was significantly lower than the impinged latent mortality (p≤0.025) along with live impinged initial being significantly less than the impinged latent mortalities (p≤0.002). The presence of *Aeromonus schubertii* was significantly higher in the aquaria controls than in the dead impinged (p≤0.032) and significantly higher in the impinged latent mortalities than the dead impinged (p≤0.004). There were no other significant differences with this pathogen.

Parasites (Table 8) found in wet mounts of gills during the study include monogenes, copepods and seven genera of protozoan parasites. Overall presence of parasites was highest in dead initial (78.7%), live impinged initial (80.0%) and impinged latent mortalities (69.3%). For overall parasites there was no significant difference between the aquaria control and the basket control (p≤0.198). The total parasite present was significantly lower in the aquaria controls than in the dead impinged (p≤0.0001), live impinged initial (p≤0.0001) and impinged latent mortality (p≤0.0001). The basket controls were significantly lower than the dead impinged (p≤0.005) and live impinged initial (p≤0.013).

For the presence of monogenes the aquaria controls were significantly less than the dead impinged (p≤0.0001), live impinged initial (p≤0.001) and impinged latent survivor (p≤0.011). There were no other significant differences in monogene presence between the other groups.

The presence of *Trichodina* spp. was significantly lower in the aquaria controls than in live impinged initial (p≤0.024) and impinged latent mortality (p≤0.018). *Trichodina* spp. were also significantly less in the basket controls than in live impinged initial (p≤0.041) and impinged latent mortality (p≤0.039).

The presence of *Capriniana* spp. was significantly lower in the aquaria controls than in dead impinged (p≤0.0001), live impinged initial (p≤0.0005) and impinged latent mortality (p≤0.027). *Capriniana* spp. was also significantly less in the basket controls than in dead impinged (p≤0.01) and live impinged initial (p≤0.041). *Capriniana* spp. was also significantly higher in the dead impinged than in the impinged latent mortality (p≤0.013).

Nematodes were observed in the abdominal cavity during gross observation in dead impinged, live impinged initial and impinged latent mortality at 0.1%, 6.7% and 1.0%. No nematodes were observed in the aquaria control, basket control and impinged latent survivors. There were no significant differences between the treatments.

All fish in the health study were observed for external health indicators such as mechanical lesions, non-mechanical lesions, eroded fins, eroded caudal fins, depigmentation and eye lesions as part of the health assessment (Table 9). Caudal fins were separated from the other fins because

of the greater potential for degrading sustained swimming performance. Total lesions, eroded fins, eroded caudal fins and depigmentation showed significant differences between groups.

For total lesions, the aquaria controls had significantly less than basket controls ($p \leq 0.0001$), dead impinged ($p \leq 0.001$), live impinged initial ($p \leq 0.0001$) and impinged live mortality ($p \leq 0.0001$). The basket controls were statistically less than live impinged initial ($p \leq 0.019$) and impinged latent mortality ($p \leq 0.0001$). Dead impinged also had significantly fewer total lesions than live impinged initial ($p \leq 0.031$) and impinged latent mortality ($p \leq 0.0001$).

Fin erosion on the pectoral fins, pelvic fins and dorsal fins of the aquaria controls was significantly less than live impinged initial ($p \leq 0.029$) and impinged latent mortality ($p \leq 0.004$). Dead impinged was also significantly less than impinged latent mortality ($p \leq 0.032$). Caudal fin erosion was significantly less in the aquaria controls than the basket controls ($p \leq 0.026$) and the live impinged initial ($p \leq 0.029$). Caudal fin erosion was significantly higher in impinged latent mortality than aquaria controls ($p \leq 0.0001$), basket controls ($p \leq 0.015$), dead impinged ($p \leq 0.0001$) and live impinged initial ($p \leq 0.001$).

Depigmentation can be an indicator of a *F. columnare* or other bacterial infection. Depigmentation was significantly higher in live impinged initial than in the aquaria controls ($p \leq 0.0001$), basket controls ($p \leq 0.034$) and dead impinged ($p \leq 0.0001$). Depigmentation was also significantly higher in impinged latent mortalities than in the aquaria controls ($p \leq 0.0001$), the basket controls ($p \leq 0.022$) and the dead impinged ($p \leq 0.0001$).

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There were no significant differences in the relative weights (W_r) of the channel catfish

treatments including the control groups (Table 11). The W_r for blue catfish impinged latent

mortalities (153.03) and impinged latent survivors (161.98) were significantly different ($p \le 0.05$)

from the aquaria controls, basket controls, dead impinged and live impinged initial with 113.79,

115.14, 121.84, and 120.1. A W_r of 100 is considered the 75th percentile of all fish of that species

(Wege and Anderson 1978). The only group in this study with a W_r less than 100 was the

channel catfish LII (93.18).

V. DISCUSSION

In 2007, Alabama experienced extreme drought conditions (Figure 27) with January being the only month to exceed the previous 4-year average monthly discharge. From February to September, the mean monthly discharge rates of the Mobile River were less than 50% of the previous 4-year mean and from May to September the monthly rates were below 22% of the 4-year mean discharge. The minimum mean daily discharge during the experiment was 1,130 ft^3/sec on 10 September, 2007 (USGS 2006, USGS 2007). This combination of tidal fluctuation and extremely low flow rates may have caused abnormal impingement in both numbers of animals and composition of the sample.

The tidal activity of the Mobile River at Plant Barry causes a backflow of heated water from the discharge canal 7-miles downstream. The discharge water moves upstream as the tide moves in and is re-circulated into the CWIS. The effect that periodic changes in water temperature have on fish disease outbreaks, susceptibility and associated impingement is unclear. The effects could vary from an altered metabolism and resilience to infection to thermal shock incapacitating or killing some species.

Screen Rotation Study

The EPA is concerned about BTA in both equipment and methods to preserve the ecology of natural ecosystems. In the case of the rotating screens, the operating of the screens to decrease the impingement of organisms counts as BTA. Plant Barry, due to its position in the Mobile River Basin and specifically the nearby river geography, experiences unusually high debris loads. A diversion dike was constructed to alter the current in the river bend in an attempt to divert much of the debris. Because of the high debris loads, Plant Barry is in the habit of operating its rotating screens at all times. Even with the screens running, Steam Generation Unit 5 went off line during this study when the CWIS for Units 4 and 5 became clogged with debris.

The screens at Plant Barry were originally designed in the 1970s and have been modified and repaired over time. To replace one unit, the estimated cost is $86,000 and the plant has 11 units between the two CWIS. Constant operation of the rotating screens wears them out quicker. Many of the equipment breakages take weeks to fix. When a screen is stop logged the same amount of water is flowing into the CWIS but the current is faster through the remaining screens and the debris load on the remaining screens is higher. So, it is to the benefit of power plants to only operate the screens as needed.

The traveling screens at Plant Barry were often non-functional during the sampling. The traveling screens were often stopped for minor repairs or were stop logged and drained to allow removal of the damaged screen. For two samples on February 14, three of the six screens on the CWIS for Units 1-3 were out of service. Two of the screens were stoplogged and the third screen

was not running but still had water flowing through it. Over the course of the study 90.2% of the screens were operational during sampling. Of the non-function screens stoplogs were in place 9.0% of the time and 0.8% were stopped but not stoplogged. All of the stoplogged screens occurred at Units 1-3. Traveling screen 6 was offline from 14 February to 18 July and accounted for 70.2% of the stoplogged screens. Screens 4 and 5 at Units 1-3 comprised the rest of the stoplogs. No screens were stoplogged during the study for Units 4-5.

During the study power generation Unit 5, the largest at Plant Barry, went offline from 22 January to February 15, 2007. This reduced the water flow in CWIS 45% to 35% of normal causing a reduction of the hydraulic zone of influence and the current. It is not known if the reduction in impingement was proportional. Unit 5 was reactivated between the paired samples for week 6 of 2007. So, for that week, the first samples were obtained at 35% flow and the second samples were obtained at 100% flow.

Other unusual events included stoppages due to icing, operators cleaning the trash rack during sampling periods, and sample collection pit flooding during a sample. However, impacts to sampling were considered minimal. Sampling was halted and times recorded with details for data analysis.

The water temperature increased from winter to summer (Figure 20). The lowest temperatures in winter occurred during the highest impingements of the freshwater fishes. Low water temperature has been correlated with high impingement rates (Griffith and Tomljanovich 1975, Mclean et al. 1985). In January and February, water temperatures were periodically below

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10 ℃. Lifton and Storr (1978) performed studies at CWIS on the Great Lakes that demonstrated an inverse correlation between water temperature and impingement rates. Other studies have shown threadfin shad have increased mortality rates in temperatures below 9 ℃ (Strawn 1965, Griffith and Tomljanovich 1975, Griffith 1978). Threadfin shad, in laboratory studies, have exhibited decreased feeding behavior (Griffith and Tomljanovich 1975, Griffith 1978) and loss of equilibrium (McLean et al. 1985, Griffith 1978). Griffith and Tomljanovich (1975) demonstrated that threadfin shad exhibited changes in swimming ability when exposed to water temperatures below 12 ℃. Also, gizzard shad impingement has been correlated with low temperatures (LaJeone and Monzingo 2000). Swimming performance of channel catfish in laboratory experiments (Hocutt 1973) were positively correlated to water temperature.

The Mobile River experiences a seasonal "salt wedge" that is influenced by tidal height and discharge of the river which this phenomenon has been well documented (Geyer and Farmer 1989) at the Fraser Estuary, British Columbia. Conductivity at Plant Barry increased from 140 µmhos/cm in January to 306 µmhos/cm in September (Figure 22). The conductivity and salinity never reached proportions equivalent to brackish or sea water. The Plant Barry intakes use surface water from a side channel off of the main River. The increased conductivity could be attributed to ion concentration from the low flows or a dilution of the salt wedge as it mixed with the fresh water at the surface. During the mid to late summer there was an increase in the numbers of brackish and salt water species in the impinged fish. The advance of the salt wedge could provide a conduit for marine species (Geyer and Farmer 1989) to move up river as supported by the number of euryhaline species impinged in the spring and summer phases.

The most highly impinged freshwater species (threadfin shad, blue catfish, channel catfish, freshwater drum and gizzard shad) demonstrated the highest impingement rates in the winter phase at Units 1-3, a short period in late winter where impingement was equal at both CWIS, before the impingement at both CWIS dropped and stayed low throughout the summer phase. This could correspond with overwintering in the intake canal and a movement to the river in warmer temperatures.

Both of the highly impinged eurihaline species (bay anchovy and hogchoker) had high impingements rates for a very short period either in the spring or summer phase with low impingement throughout the rest of the study.

The two most impinged non-fish species were shrimp and blue crab. Shrimp impingement was highest in the spring extremely low impingement rates during the winter and summer phases (Figure 18). Blue crabs were impinged throughout the study but impingement was only heavy in the summer phase (Figure 19). There was a significant increase in impingement of blue crabs when traveling screens were inactive. It is likely blue crabs climb inactive screens to forage in the debris trapped on the screens. When the screens are activated crabs may hold on instead of trying to escape thus increasing impingement.

Survival Study

Another possible application of BTA at Plant Barry is employing a fish return chute from the discharge flume of the CWIS that currently does not exist. This allows fish that survived the

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impingement process to be returned to the body of water. Due to the design of the CWIS this would be a lengthy and expensive operation. This is one of the reasons the survival and health study was performed. By determining the percentage of fish surviving the impingement, it allows the EPA and the power plant to evaluate the efficacy of a fish return chute. If a large number of the commonly impinged fish would not survive being returned to open water, a fish return chute may not be warranted.

Most of the previous survival studies used scaly fish with high survivability. This study used blue catfish and channel catfish because they are two of the most impinged freshwater species at Plant Barry and very few survival studies have been conducted on catfish or bullheads.

Another reason for conducting the Survival and Health Study was other researchers (Baker 2007, Knight 2008) have noted the unusually high prevalence of bacterial infections in the impinged fish at Plant Barry. Common survival studies determine survival over 48 or 72 hour periods. By looking at the health of impinged fish from impingement out to 168 hours (7 days), we evaluated the latent health effects caused by impingement.

The control fish were added to the health assessment to resolve questions raised by the Baker (2007) and Knight (2008) studies. Both studies looked at health parameters of impinged fish at Plant Barry. The samples were collected from the debris basket at the end of the discharge flume. The fish used in both studies were determined fresh by examination of gills for color, clearness of the eye and absence of general necrosis. Since there was no indicator of how long the fish had

been in the capture basket the question has been raised that maybe the pathogens developed after death while the fish were in the basket.

In this study the maximum time a fish was in the capture basket was 1 hour. It is unlikely the presence of fish pathogens infected fish within that time frame or caused advanced lesions that were observed. The aquaria controls provided us a baseline of background pathogens and the basket controls allowed us to evaluate the effect the capture basket might be having on the impinged fish. While the prevalence of bacteria was higher in the basket controls than in the aquaria control, there was not a significant different.

The survival study was conducted during an extreme drought (Figure 27). There is no way to evaluate what affect the drought had on the numbers of catfish impinged in this study. The river flow for the duration of the study was less than 20% of the 4-year average prior to 2007.

The total number of catfish impinged from 01 June to 06 August 2007 was 257 (Table 6). The impinged sample consisted of 108 dead catfish, 44 live catfish sacrificed for the fish health study and 105 live catfish used in the survival study. Approximately 58% of the catfish survived impingement and collection and were used in the aquaria survival study.

Many of the nightly sampling periods yielded no catfish. The dates of the survival study corresponded with the lowest catfish impingement times from the screen rotation study. By conducting the study during the summer months the confounding variables associated with winter mortality and rain events were avoided. So, in theory, starvation, low metabolism,

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suppressed immune systems and river conditions should not have affected the survival of the impinged catfish. A relative weight (W_r) of 100 is considered to be the 75th percentile for that species of fish (Wege and Anderson 1978) and the standard parameters used as suggested by Murphy et al. (1991). In support of this, all of the W_r (Table 11) of the impinged fish and controls were above 100 except for the live impinged initial channel catfish (93.18). Baker (2007) found a significant difference ($p \leq 0.006$) in the length weight regressions for blue catfish in the spring sample. There was no significant difference between the population's length-weight regressions for channel catfish in the spring or in both species of catfish during the fall.

Due to the difficulties of obtaining blue catfish and channel catfish of the same length as the impinged fish, the decision was made to use channel catfish raised at the Auburn University North Station (S-6).The rearing water supply was well water and the fish were raised in tanks with a standard, commercially available fish feed. The transport from Auburn University to Plant Barry caused little mortality in transit and the fish were acclimated to the river water for one week prior to being used in the experiment. The control catfish were not representative of the native river population in potential predisposition and exposure to pathogens present in the Mobile River. The study used these controls to test for stress associated with impinged basket collection and potential pathogen exposure in this time period. The aquaria controls provided a baseline of healthy fish living in the Mobile River water. The basket controls were a basis for determining what effect the capture gear caused to the fish and what latent health effects were due to the added stress and pathogens present in the water.

Statistically there were no differences in survival times (Figure 26) between the impinged blue catfish and impinged channel catfish ($p \leq 0.93$). There was no significant difference between the survival rates of the two control groups ($p \leq 0.086$). There was a significant difference between the control groups and the impinged blue catfish ($p \leq 0.0001$) and the control groups and the channel catfish ($p \leq 0.0001$). The basket controls (18.9%) experienced roughly double the mortalities of the aquaria controls (9.5%). While this suggests that being placed in the turbulence of the collection basket increased the mortality rate, there were no immediate mortalities due to the basket. The first mortality in the basket controls occurred after 42 hours.

This survival study was unusual in the length of time fish were observed for latent mortality and the use of control fish. Many studies only observe the fish for three days instead of seven. The fact no mortalities occurred in the impinged fish after three days validated the use of three day observations for survival studies. This study was also unusual in the use of control fish to establish baselines of survival and effect of capture gear.

The fluctuations in water temperature due to the heated discharge water recirculating from downstream into the CWIS appeared to be extreme. To address this question a study (Bevelhimer and Coutant 2007) performed to determine exactly what effect fluctuating water temperatures near the extreme upper limits had on black crappie (*Pomoxis nigromaculatus*), mosquitofish (*Gambusia affinis*) and striped bass (*Morone saxatilis*). One trial varied the temperature from $31°C$ to $36°C$ for two hours before raising the temperature to loss of equilibrium. The results suggested that exposure to temperatures near the critical upper temperatures of these species are not necessarily harmful and that full recovery from heat stress

causing loss of equilibrium may occur in as little as an hour. One study (Currie et al. 1998) determined the critical thermal maxima of channel catfish to be 40.3 °C for catfish acclimated to a constant temperature of 30 °C.

The prevalent bacterial pathogens found in this study were *F. columnare*, *A. veronii biotype sobria* and *A. schubertii* which are in line with the findings of Baker (2007). Of the fourteen known fish pathogens these three also showed significant differences between the control groups and the impinged fish. The techniques used in this study were suited to the detection of parasites and bacterial infections but not for quantifying the degree of infection or effect of the pathogens on the catfish.

The pathogen surveys are from the different stages in the impingement process and are only representative of the impinged fish. The sample is representative of the immediate area of the CWIS. Prior studies (Baker 2007; Knight 2008) have indicated higher rates of pathogens among fish in the intake structures when compared to fish in the intake channels and rivers.

 Living and dead impinged catfish in this study were immediately surveyed to determine pathogen prevalence at impingement and if there was a difference in prevalence between the two groups. The latent mortality group showed how many fish died within one week and what the pathogen prevalence was in that group. When comparing prevalence of bacterial infections (Table 7), there was a general tendency for the basket controls to have a higher prevalence of bacterial pathogens than the aquaria controls. This difference was only significant with *F. columnaris* infections of the gills ($p \leq 0.0031$) and *A. veronii biotype sobria* ($p \leq 0.008$).

The Dead Impinged and Live Impinged Initial groups represent the impinged population at the time of impingement. Presence of pathogens was significantly higher in the dead impinged in the presence of overall bacterial infections, *F. columnare*, and *A. veronii biotype sobria*. Between these two groups *A. schubertii* was not statistically different and occurred in one fish per group. The greater prevalence of pathogens in the Dead Impinged group would indicate the Live Impinged Initial group was healthier and more capable of surviving the stress of impingement.

Most striking is the incidence of bacterial infection in the Impinged Latent Mortality group. These fish survived impingement but died within the 7 day post-impingement observation period. All of the wild mortalities occurred within 72 hours. The impinged latent survivor is the only group to have no infections of these three species. It is likely the stress of impingement allowed pre-existing internal infections to overcome the fish's immune response in the impinged latent mortalities. The absence of the pathogens initially would explain why the prevalent pathogens were not found in the survivors.

Columnaris disease, first described by Davis (1922), is known to affect most species of fish in fresh water (Roberts 2001). The incidence of *F. columnare* (for blue catfish and channel catfish combined) at Plant Barry was 78.7% on the dead impinged and 66.7% on the impingement survivors in this study. Baker (2007) at Plant Barry reported *Aeromonas spp.* in 43.9% of blue catfish and 10.6% of channel catfish. The same study also found no *F. columnare* on reference fish captured from the Mobile River but found prevalence rates of 60.9% and 30.3% on impinged blue catfish and channel catfish. However, the study was conducted in 2005 under normal rainfall and river flows. Fujihara and Olsen (1962) compared the prevalence of *F.*

columnare on fish from three sample sites in the Columbia River to fish from an artificial

spawning channel. The fish collected were also categorized by temperature ranges that reflected

the sample period. Prevalence rates from the Columbia River samples ranged from 1.8% at the

lowest temperature range to 3.6% at the upper temperature range. In the artificial spawning

channel the incidence of *F. columnare* was 22% for fish held in less than 15.6 ℃ and 54% for

fish held between 15.6-20.6 ℃. The authors concluded that high fish densities contributed to the

levels of infection.

Prevalence rates for *F. columnare* on the gills of wild populations of fish have ranged as low as

0-1.5% to as high as 75-93% (Fujihara et al. 1964, Becker and Fujihara 1978, Bowser 1973,

Pacha and Ordal 1970). The types of media used in the studies varied and could explain a failure

to isolate *F. columnare*. Whereas the media used in this study was the Hsu-Shotts medium

selective for *F. columnare*.

Epizootic events in Alabama water bodies have been attributed to various pathogens. Hawke

(1974) investigated fish kills in seven Alabama impoundments and determined *Aeromonus*

hydrophila to be the etiological agent in 90% of diseased fish. Unfortunately detection of a

pathogen does not necessarily indicate disease in fish. In a survey of fish sampled from lakes

Martin and Logan from 1973 to 1974 by electrofishing, Hawke (1974) revealed a carrier rate of

A. hydrophila of 10% and 25% respectively.

From 01 June to 06 August 2007 water for the aquaria was pumped from Plant Barry Units 1-3.

The recorded water temperatures at the aquaria ranged from 24.8 ℃ to 34.9 ℃ and were highly

variable daily depending on whether the thermal discharge water from the plant outflow was being recirculated through the intakes. Normal procedures for transferring fish require acclimatization or tempering if a fish is subjected to more than a 2 ℃ temperature difference when moved from one body of water to another and to avoid a temperature change of more than 5 ℃ per hour (Stickney 1979). Tempering does not allow for complete acclimation to the new temperature in the time usually available and therefore likely represents a combination of rapid acclamatory adjustments and reduction of stress (Stickney 1979).

The preferred temperature of channel catfish in a lab study was been found to be 25.2 ℃ (Reutter and Herdendorf 1974) with the upper avoidance temperatures for channel catfish found to be 32 ℃ by Gammon (1973) and 34 ℃ by Proffitt (1969). While the water temperature never exceeded a 5 ℃ temperature change in 1 hour, however, it did frequently get above 32 ℃ during the survival study and exceeded 34 ℃ seven times. Short term acclimation uses a fish's energy resources that could be turned to growth and reproduction or the ability to resist disease. The constant temperature fluctuations of the CWIS at Plant Barry are likely causing an unusually large stress on the local fish population. It is also very likely the stress of adjusting to the constant temperature swings observed in this study could adversely affect the fish immune systems and could be adversely synergistic when combined with the act of impingement. Other impingement survival studies (Jinks et al. 2003) have reported high initial impingement survival of white catfish (Ameiurus catus) 89.4%, brown bullhead (*Ameiurus nebulosus*) 97.4% and channel catfish 84.3% with similar vertical rotating screens. The average extended survival rates for the same fish were 73.5%, 76.4% and 69.7% respectively. Very few, if any, extended

survival studies have used blue catfish and none of the other studies made reference to thermal pollution like is present at the CWIS at Plant Barry.

The prevalent fish pathogens at Plant Barry appear to be *F. columnare*, *A. veronii biotype sobria*, and *P. shigelloides* with prevalence rates of up to 78.7%, 26.9%, and 6.7% respectively, appear to be the prevalent pathogens present in the impinged catfish at Plant Barry. The *F. columnare* present on the gills appears to be high but is within ranges found by other researchers (Bowser 1973, Baker 2007). The prevalence rates of *A. veronii biotype sobria* and *P. shigelloides* may also represent the carrier rate of the local fish population.

This study should have added one more category to the health analysis. The control fish could have been surveyed for presence of pathogens prior to shipment, placement in the holding tank and exposure to the river water. This would have indicated what pathogens were present in the controls prior to exposure to water from the Mobile River. There would be no way to evaluate the effect of transport stress on the controls prior to being placed in the control tank. The controls may also represent a naïve population susceptible to Mobile River pathogens, whereas the local population may have had prior exposure and become resistant to or carriers of the local pathogens.

VI. CONCLUSION

In review, there were two questions this study was designed to answer. At the Plant Barry Cooling Water Intake Structures (CWIS), determine: 1) what effect varying the operation times of rotating screens had upon fish impingement rates and 2) the survival rates of impinged blue catfish and channel catfish along with the pathogen prevalence at impingement, mortality or one week post impingement as they may relate to impingement survival.

Screen Rotation Study

It appears that altering the operation times of rotating screens as a BTA could significantly affected the impingement rates of the bay anchovy, hogchoker and blue crab at Plant Barry. All three species had distinct periods where impingement levels were high in the spring or summer. The five most impinged freshwater species, threadfin shad, blue catfish, channel catfish, freshwater drum and gizzard shad did not show significant changes in impingement rate overall or by season.

Overall there was no significant difference in impingement if the screens were operated continuously or not. Balancing the need to reduce impingement with the need to clear debris, the operation schedules of the power plant could be adjusted to implementing BTA. The freshwater

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species were most impinged in the winter sample and their impingement did not appear to be significantly affected by screen operation schedules in any season.

Overall, impingement rates dropped from winter to summer. When looked at by species, two rough groupings stand out. The first group is the winter mortalities. Winter mortalities were overwhelmingly fresh water fish impinged at CWIS Unit 1-3. The highest impingement rates occurred during the coldest temperatures in the study. The second group is the seasonal eurihaline species. Shrimp and hogchoker were impinged in high numbers from early April until late May. Bay anchovy were impinged in high numbers from late April until mid July. In addition, blue crabs were impinged from mid June until the end of the study.

These two groupings are relevant when applied to the screen rotation experiment. There was no significant difference in impingement overall. There was also no significant difference in winter impingement rates which consisted of mostly fresh water species.

Three of the four salt water or brackish species had definite times that impingement rates increased and decreased. In the case of the blue crab the study covered the arrival of the blue crab in large numbers but ended before the high impingement of the crabs ended. There was a significant difference in all four of the eurihaline species impingement rates in units where screen operation was altered compared to continuous operation.

Stopping the rotation of the screens during the winter and spring months prior to the arrival of hogchoker and shrimp does not appear to affect the impingement rates. It is possible that

continuous operation of the traveling screens would reduce impingement when the salt water and brackish species are present in high numbers.

The impingement survival study was performed during the summer months when starvation, suppressed immune systems and low temperatures were not likely causes of mortalities. The blue catfish and channel catfish were also impinged in very low numbers compared to the winter impingement. The number of dead impinged catfish useable for bacterial samples was roughly the same as the impingement survivors during the collection period with 108 and 107 respectively.

Statistically there was no significant difference in mortality rates of the two species of catfish. Overall, 105 wild catfish survived impingement to be included in the study and only 10 or 9.5% survived for 7 days post impingement. The aquaria controls had a 90.5% survival rate and the basket controls had an 81.8% survival rate.

Survival Study

There was a very low survival of the fish in this study. The blue catfish and channel catfish had a 9.5% survival rate compared to the 90.5% survival of the controls. All of the mortalities in the impinged fish occurred prior to 72 hours. This study used only two of the commonly impinged species at Plant Barry. The low survival rate of the impinged catfish may not justify the added cost of BTA like installing fish return chutes at the current CWIS.

From comparing the results of the aquaria controls and the basket controls, the capture gear did not cause a significant increase in pathogen presence. That indicates a high prevalence of pathogens in the impinged catfish prior to their impingement. Previous studies (Baker 2007, Knight 2008) have indicated the unusually high prevalence of pathogens in many of the species impinged at Plant Barry but found a low occurrence of infected fish in the river adjacent to the power plant. The presence of a pathogen does not necessarily indicate a diseased fish. Some pathogens are endemic to the natural environment and may be present in small enough numbers that the fish are not negatively affected. The fact that so many of the fish impinged did have pathogens present suggests the fish were impaired and the CWIS are selectively impinging sick fish.

The results of the health study indicate a very high prevalence of *F. columnare* in the mortalities. Of the impinged mortalities, 71.6% had *F. columnare* infections. The impinged 7 day survivors still had *F. columnare* present on 50.0% of the gills although no *F. columnare* was detected internally. The survivors in the aquaria and basket control had 2.6% and 16.7% respectively F. columnare present on the gills and no internal infections detected. The aquaria and basket control mortalities had 25% and 100% *F. columnare* detected on the gills.

During the health analysis of the survival study thirteen bacterial fish pathogens were identified. Impinged fish were categorized as recently dead, live sacrificed immediately after capture, and live placed in aquaria. The prevalence of bacterial pathogens present in impinged fish was 91.7%, 86.0% and 89.7% respectively. The occurrence of bacterial infection was significantly less in the aquaria controls (31.0%) and basket controls (54.5%). It appears that exposure to the

capture gear for one hour may have increased the incidence of bacterial infection in the control fish by 23.5%. There does not appear to be a significant difference in infections between impinged fish placed in aquaria compared to the fish sacrificed immediately.

The controls were exposed to the Mobile River water for up to 6 weeks during the course of the experiment. There was no pathogen survey performed on the control fish before entry into the study. So it is difficult to determine what pathogens were present in the controls initially and what pathogens were acquired after exposure to the river water.

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Table 1. Biochemical tests used to determine known Gram negative and cytochrome oxidase positive fish pathogens in the Plant Barry Survival Study in 2007.

Gram negative/Cytochrome Oxidase Positive

Test/Species	Pseudomonas spp.	Vibrio spp.	P. Shigelloides	Aeromonas sobria	A. veronii bio sobria	A. caviae	A. media
Gram (KOH string test)	neg	neg	neg	neg	neg	neg	neg
Motility at 30 C				pos	pos	pos	pos
Cytochome Oxidase	pos	pos	pos	pos	pos	pos	pos
Glugose Fermentation	neg	pos	pos	pos	pos	pos	pos
Mannitol Utilization	neg	pos	pos	pos	pos	pos	pos
0/129 Vibriostat		sensitive	sensitive	resistant	resistant	resistant	resistant
Novobiocin		sensitive	resistant				
0% NaCl				pos	pos	pos	pos
Sucrose Utilization				pos	pos	pos	pos
Esculin Hydrolysis				neg	neg	pos	pos
Ampicillin				resistant	sensitive	resistant	sensitive
V-P						neg	neg
Ornithine Decarboxylase							
DL-Lactate Utilization							
Urocanic Acid Utilization							
Indole							

Gram negative/Cytochrome Oxidase Positive

Test/Species	A. veronii bio veronii	A. hydrophila	A. bestiarum	A. janaei	A. trotta	A. schubertii
Gram (KOH string test)	neg	neg	neg	neg	neg	neg
Motility at 30 C	pos	pos	pos	pos	pos	pos
Cytochome Oxidase	pos	pos	pos	pos	pos	pos
Glugose Fermentation	pos	pos	pos	pos	pos	pos
Mannital Utilization	pos	pos	pos	pos	pos	pos
0/129 Vibriostat	resistant	resistant	resistant	resistant	resistant	resistant
Novobiocin						
0% NaCl	pos	pos	pos	pos	pos	pos
Sucrose Utilization	pos	pos	pos	neg	neg	neg
Esculin Hydrolysis	pos	pos	pos			
Ampicillin						
V-P	pos	pos	pos	pos	neg	
Ornithine Decarboxylase	pos	neg	neg			
DL-Lactate Utilization		pos	neg			
Urocanic Acid Utilization		neg	neg			
Indole				pos	pos	neg

Table 2. Biochemical tests used to determine known Gram negative and cytochrome oxidase negative fish pathogens in the Plant Barry Survival Study in 2007.

Gram Negative/Cytochrome Oxidase Negative			
Test/Species	*E. tarda*	*Y. ruckeri*	*E. ictaluri*
Motility at 30 C		pos	neg
Indole	pos	neg	neg
MacConkeys	neg	neg	neg
Phenylalanine Deaminase	neg	neg	neg
Citrate	neg	neg	neg
Hydrogen Sulfide	pos	neg	neg

Table 3. Screen rotation comparisons by treatment and intake. An * indicates significant differences (p≤0.05).

Fish Species	Treatment p-value		Intake p-value	
Total Impinged	0.1469		0.0348	*
Freshwater Species				
Threadfin Shad	0.2114		0.5939	
Blue Catfish	0.3114		0.0001	*
Channel Catfish	0.7299		0.0005	*
Freshwater Drum	0.2084		0.0001	*
Gizzard Shad	0.7000		0.0001	*
Eurihaline Species				
Bay Anchovy	0.0194	*	0.0038	*
Hogchoker	0.0040	*	0.7905	
Non-fish Species				
Shrimp spp.	0.3552		0.5024	
Blue Crab	0.0001	*	0.0010	*

Table 4. Impingement comparisons of overall and the most impinged species by phase. Different letters within the same row indicates significant differences (p≤0.05).

Fish Species	Winter		Spring		Summer	
Total Impinged	7,724		6,398		7,048	
Freshwater Species						
Threadfin Shad	1,712	a	248	b	2,386	a
Blue Catfish	4,004	a	649	b	213	b
Channel Catfish	793	a	460	b	18	c
Freshwater Drum	653	a	226	b	46	b
Gizzard Shad	369	a	16	b	2	b
Eurihaline Species						
Bay Anchovy	1	a	2,309	b	2,747	b
Hogchoker	35	a	430	b	200	a
Non-Fish Species						
Shrimp spp.	123	a	1,949	b	44	a
Blue Crab	33	a	110	a	1,395	b

Table 5. Mean, minimum, maximum and counts of environmental data collected during impingement study.

		Total		Winter		Spring		Summer	
		Unit 1-3	Unit 4-5	Unit 1-3	Unit 4-5	Unit 1-3	Unit 4-5	Unit 1-3	Unit 4-5
Temp (C)	Mean	21.0	21.1	11.4	11.6	23.7	23.8	31.1	31.1
	Minimum	8.9	9.1	8.9	9.1	17.8	18.4	28.3	28.6
	Maximum	33.4	34.8	17.2	17.0	29.6	30.0	33.4	34.8
	N	115	115	42	42	45	45	28	28
DO (mg/l)	Mean	8.73	8.68	10.64	10.48	8.07	8.01	6.92	7.07
	Minimum	6.12	6.01	9.74	9.49	6.82	6.64	6.12	6.01
	Maximum	11.45	11.32	11.45	11.32	9.60	9.76	8.15	9.06
	N	115	115	42	42	45	45	28	28
pH(units)	Mean	7.19	7.15	7.23	7.24	7.11	7.06	7.24	7.18
	Minimum	6.68	6.59	6.80	6.76	6.68	6.59	6.91	6.61
	Maximum	7.77	7.79	7.77	7.79	7.61	7.57	7.61	7.60
	N	115	115	42	42	45	45	28	28
Specific Conductivity (microS/cm)	Mean	207	207	158	158	207	207	281	281
	Minimum	142	140	142	140	149	149	248	248
	Maximum	306	301	182	182	246	247	306	301
	N	115	115	42	42	45	45	28	28
Turbidity (ntu)	Mean	18.14	15.67	21.21	17.61	19.37	17.28	11.57	10.17
	Minimum	2.70	6.10	11.00	8.10	9.51	9.88	2.70	6.10
	Maximum	90.00	36.00	90.00	28.00	45.00	36.00	20.90	15.10
	N	115	115	42	42	45	45	28	28

Table 6. Total impinged blue catfish and channel catfish used in the survival and health studies at Plant Barry, AL from 01 June 2007 to 06 August 2007.

	Category of Impingement						
	Dead Blue Catfish	Dead Channel Catfish	Blue Catfish Sacrifice	Channel Catfish Sacrifice	Latent Impinged Blue Catfish	Latent Impinged Channel Catfish	Total
Mortality	84	24	37	7	74	21	247
Survivor	-	-	-	-	7	3	10
Total	84	24	37	7	81	24	257

Table 7. Prevalence of bacterial infections found in treatment groups from the Plant Barry Health Study in 2007. Different letters within the same row indicates significant differences (p≤0.05).

Bacterial Pathogen	Category					
	AC	BC	DI	LII	ILM	ILS
Liver Bacteria (all spp.)	9.8^a	21.7^{ab}	38.9^{bc}	20.0^{ab}	62.1^d	30.0^{abcd}
Kidney Bacteria (all spp)	26.8^a	43.5^{ab}	37.0^{abc}	24.4^{ad}	65.7b^e	30.0^{abcde}
F. columnare (gills)	4.9^a	30.4^b	78.7^c	66.7^c	68.4^c	50.0^{bc}
F. columnare (kidney)	2.4^a	13.0^{ab}	58.3^c	26.7^{bd}	47.4^c	0.0^{abd}
Aeromonus schubertii	4.9^a	4.3^{ab}	1.9^{bc}	2.2^{ad}	12.6^{abe}	0.0^{abcde}
A. veronii biotype sobria	17.1^a	39.1^b	26.9^{bc}	13.3^{acd}	42.1^{be}	0.0^{acde}
A. beastiarum	0.0	0.0	1.9	0.0	3.2	0.0
A. sobria	0.0	0.0	0.9	0.0	0.0	0.0
A. jandei	0.0	0.0	1.9	0.0	1.1	0.0
A. caviae	0.0	0.0	0.0	2.2	1.1	0.0
A. hydrophila	0.0	0.0	0.0	0.0	2.1	0.0
A. trotta	0.0	0.0	0.0	0.0	2.1	0.0
Plesiomonas shigelloides	4.9	0.0	6.5	6.7	2.1	10.0
Unidentified Bacteria	0.0	0.0	2.8	0.0	10.5	10.0
Vibrio spp.	0.0	0.0	1.9	0.0	4.2	10.0
Pseudomonas spp.	0.0	0.0	0.9	0.0	0.0	0.0
Proteus spp.	0.0	0.0	0.0	2.2	1.1	0.0
Edwardsiella tarda	0.0	0.0	0.0	2.2	0.0	0.0

AC = Aquaria Control
BC = Basket Control
DI = Dead Impinged
LII = Live Impinged Initial
ILM = Impinged Latent Mortality
ILS = Impinged Latent Survivor

Table 8. Prevalence of parasites by treatment during the Plant Barry Survival Study in 2007. Different letters within the same row indicates significant differences (p≤0.05).

Parasitic Pathogen	Category					
	AC	BC	DI	LII	ILM	ILS
Total with Parasites	33.3[a]	50.0[ab]	78.7[c]	80.0[d]	69.3[bcd]	62.5[abcd]
Monogene spp.	29.3[a]	39.1[b]	58.3[b]	62.2[b]	49.5[b]	50.0[ab]
Trichodina spp.	2.4[a]	0.0[a]	11.1[b]	15.6[b]	16.2[b]	0.0[a]
Capriniana spp.	0.0[a]	0.0[a]	20.4[b]	15.6[c]	10.5[c]	0.0[a]
Henneguya spp.	4.9	4.3	18.5	11.1	16.2	25.0
Ichthyobodo necatur	0.0	0.0	5.6	2.2	7.6	0.0
Apiosoma spp.	0.0	0.0	1.9	0.0	1.0	0.0
Ambiphrya spp.	2.4	0.0	1.9	0.0	1.0	0.0
Copepod spp.	0.0	4.3	8.3	6.7	8.6	0.0
Epistylus spp.	0.0	0.0	0.9	0.0	0.0	0.0
Glochidia spp.	0.0	0.0	4.6	4.4	1.0	0.0
Nematode spp.	0.0	0.0	0.9	6.7	1.0	0.0

AC = Aquaria Control

BC = Basket Control

DI = Dead Impinged

LII = Live Impinged Initial

ILM = Impinged Latent Mortality

ILS = Impinged Latent Survivor

Table 9. Prevalence of external lesions found during the Plant Barry Health Study in 2007. Different letters within the same row indicates significant differences ($p \leq 0.05$).

Physical Characteristic	Category					
	AC	BC	DI	LII	ILM	ILS
Total Fish with Lesions	7.1^a	22.7^b	34.3^b	53.7^c	70.3^c	25.0^{abc}
Eroded Fins (excludes caudal)	2.4^a	8.7^a	10.2^{ab}	16.2^b	22.2^b	12.5^{ab}
Eroded Caudal Fin	2.4^a	17.4^b	9.3^{ab}	42.9^c	45.5^c	12.5^{abc}
Depigmentation	2.4^a	13.0^b	12.0^b	38.1^c	38.6^c	12.5^{abc}
Non-mechanical Lesion	2.4	4.3	13.0	16.2	19.0	0.0
Mechanical Lesion	0.0	4.3	6.5	5.7	5.9	0.0
Eye missing or damaged	2.4	4.3	4.6	1.0	1.0	0.0

AC   = Aquaria Control
BC   = Basket Control
DI   = Dead Impinged
LII   = Live Impinged Initial
ILM = Impinged Latent Mortality
ILS = Impinged Latent Survivor

Table 10. Length and weight data of the treatment groups in the Plant Barry Fish Health Study in 2007.

Category		Maximum	Minimum	Mean	N	SD
AC						
Small	Weight (g)	6	1	2.7	21	1.1
	Length (mm)	95	54	70.5	21	9.5
Large	Weight (g)	60	17	31.2	21	8.9
	Length (mm)	204	133	164.4	21	15.6
BC						
Small	Weight (g)	4	1	2.9	16	1.0
	Length (mm)	86	50	72.8	16	11.7
Large	Weight (g)	47	25	34.2	6	8.9
	Length (mm)	185	151	165.8	6	13.0
Impinged						
DBCF	Weight (g)	314	1	15.1	84	39.5
	Length (mm)	337	49	104.9	84	49.5
DCCF	Weight (g)	408	1	27.8	24	83.1
	Length (mm)	349	52	116.7	24	63.1
LIBCF	Weight (g)	130	1	11.6	37	25.5
	Length (mm)	269	52	93.6	37	49.4
LICCF	Weight (g)	52	1	15.9	8	17.9
	Length (mm)	207	87	120.8	8	46.8
LILBCF	Weight (g)	173	1	9.9	81	22.0
	Length (mm)	287	42	88.4	81	44.0
LILCCF	Weight (g)	91	1	18.4	24	23.1
	Length (mm)	275	58	123.5	24	50.1

AC = Aquaria controls. All aquaria controls were channel catfish
BC = Basket controls. All basket controls were channel catfish
DBCF = Impinged blue catfish that were dead on collection
DCCF = Impinged channel catfish that were dead at collection
LIBCF = Impinged blue catfish alive at collection and necropsied
LICCF = Impinged channel catfish alive at collection and necropsied
LILBCF = Impinged blue catfish alive at collection used in survival study
LILCCF = Impinged channel catfish alive at collection used in survival study

Table 11. Relative weights (W_r) of blue catfish and channel catfish used in the Survival and Health Studies at Plant Barry, AL from 01 June 2007 to 06 August, 2007. W_r of 100 is considered the 75th percentile of all fish for that species. Different letters within the same row indicates significant differences ($p \leq 0.05$).

	Category					
	AC	BC	DI	LII	ILM	ILS
Blue Catfish	113.79[a]	115.14[a]	121.84[a]	120.1[a]	153.03[b]	161.98[b]
Channel Catfish	113.79	115.14	110.26	93.18	107.50	139.59

AC = Aquaria Control
BC = Basket Control
DI = Dead Impinged
LII = Live Impinged Initial
ILM = Impinged Latent Mortality
ILS = Impinged Latent Survivor
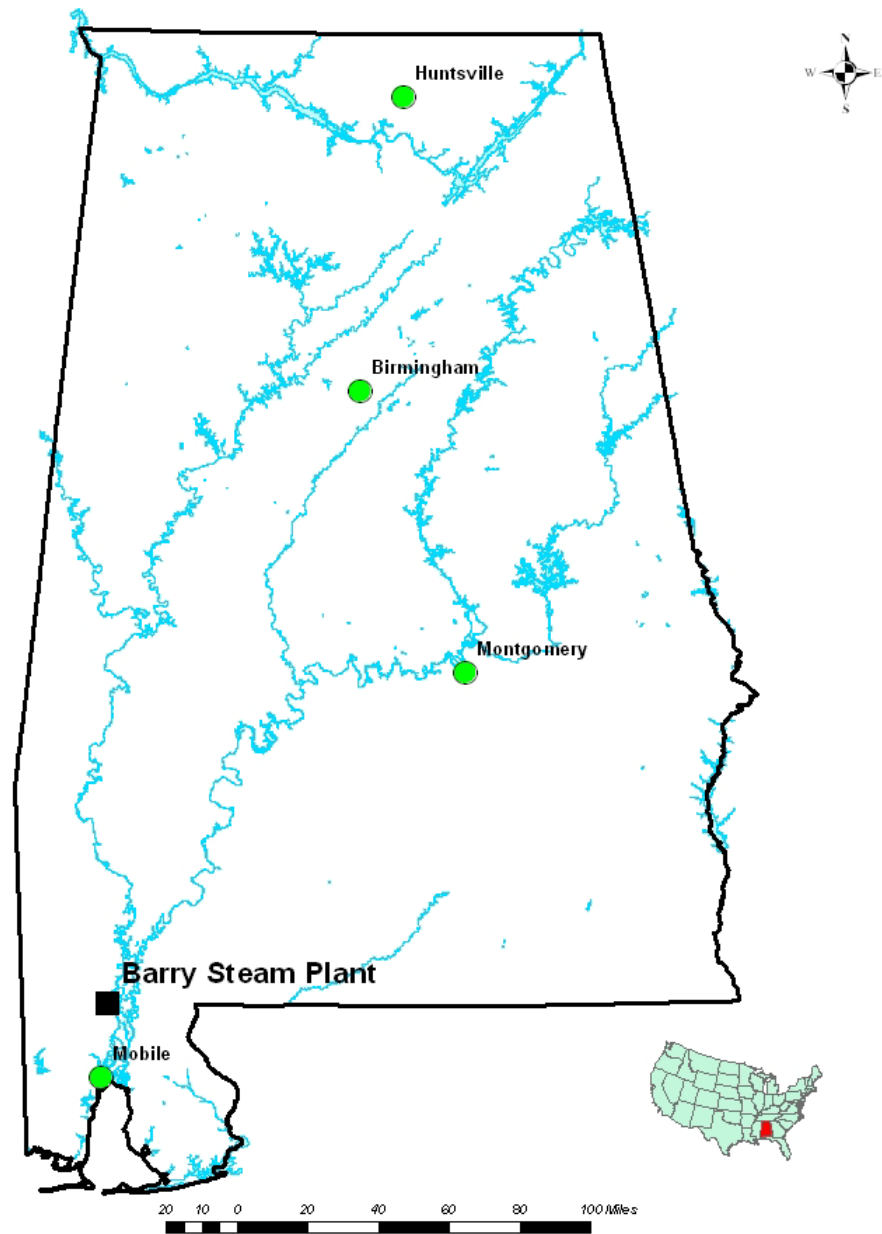
Figure 1. Site map of Plant Barry located on the Mobile River near Bucks, Alabama.
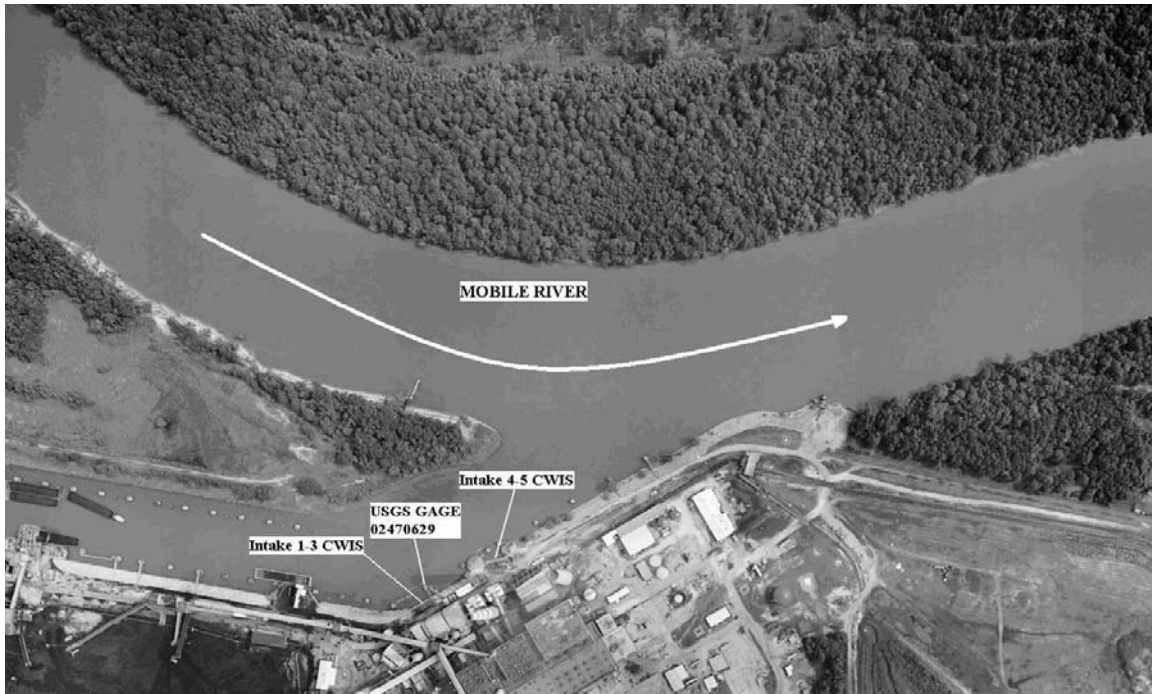
Figure 2. Overhead photo of Plant Barry intake structures and associated intake canal on the Mobile River at Bucks, Alabama with cooling water intake structures and USGS Gage indicated.

Figure 3. Plant Barry cooling water intake structure for steam generation Units 4 and 5.
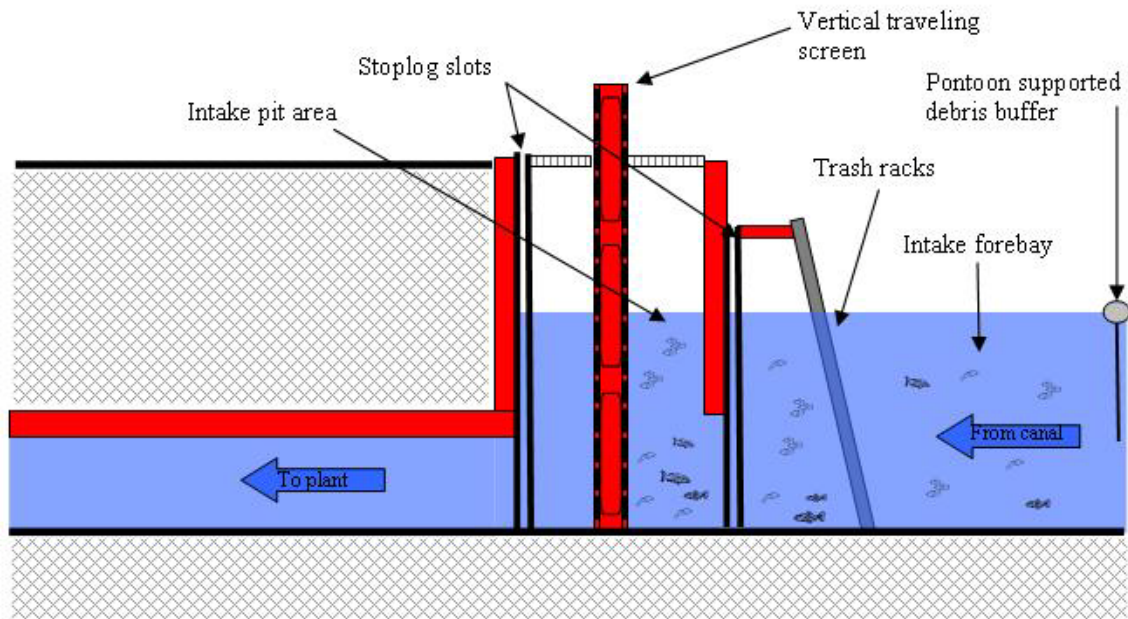
Figure 4. General schematic of the cooling water intake structures at Plant Barry.

Figure 5. Sample collection from the outflow flume at the cooling water intake structure steam generation Units 4 and 5 during the Screen Rotation Study at Plant Barry, Alabama in 2007.

Figure 6. Outflow flume and collection basket at the cooling water intake structure for steam generation Units 1 through 3 during the Plant Barry Survival Study in 2007.

Figure 7. Aquaria system and temperature controlled trailer used in the Survival Study at Plant Barry, Alabama in 2007.

Total by Species

Species	Count
Unidentifable Sunfish	1
Striped Mullet	1
Spotted Bass	1
Speckled Worm Eel	1
Smallmouth Buffalo	1
Silver Chub	1
Paddlefish	1
Longnose Gar	1
Harlequin Darter	1
Gulf Killifish	1
Golden Shiner	1
Chain Pickerel	1
Striped Bass	2
Spotted Sucker	2
Quillback	2
Crayfish	3
Blacktail Redhorse	3
American Eel	3
Longnose Shiner	4
Emerald Shiner	4
Spotted Gar	5
Redear Sunfish	5
Altantic Croaker	10
Violet Goby	11
Flathead Catfish	11
White Bass	14
Needlefish	14
Yellow Bass	19
Inland Silverside	24
Brook Silverside	25
Bluegill	27
White Crappie	28
Skipjack Herring	55
Black Crappie	60
Corbicula (Asiatic clams)	92
Sand Seatrout	200
Gulf Menhaden	221
Silverside Shiner	286
Gizzard Shad	399
Hogchoker	1074
Freshwater Drum	1094
Channel Catfish	1613
Blue Crab	2737
Shrimp spp.	3260
Blue Catfish	5273
Threadfin Shad	6537
Bay Anchovy	8858

Figure 8. Numbers of fish by species impinged during the Screen Rotation Study at Plant Barry, Alabama in 2007.
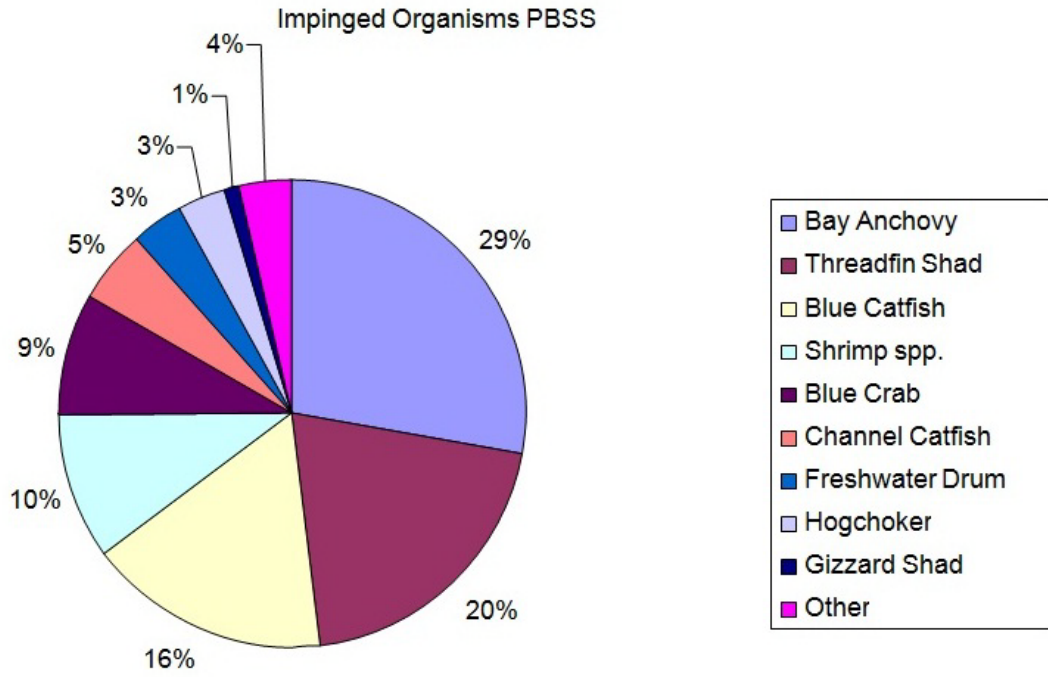
Figure 9. Percent number of the nine major species impinged during the Screen Rotation Study at Plant Barry, Alabama in 2007.
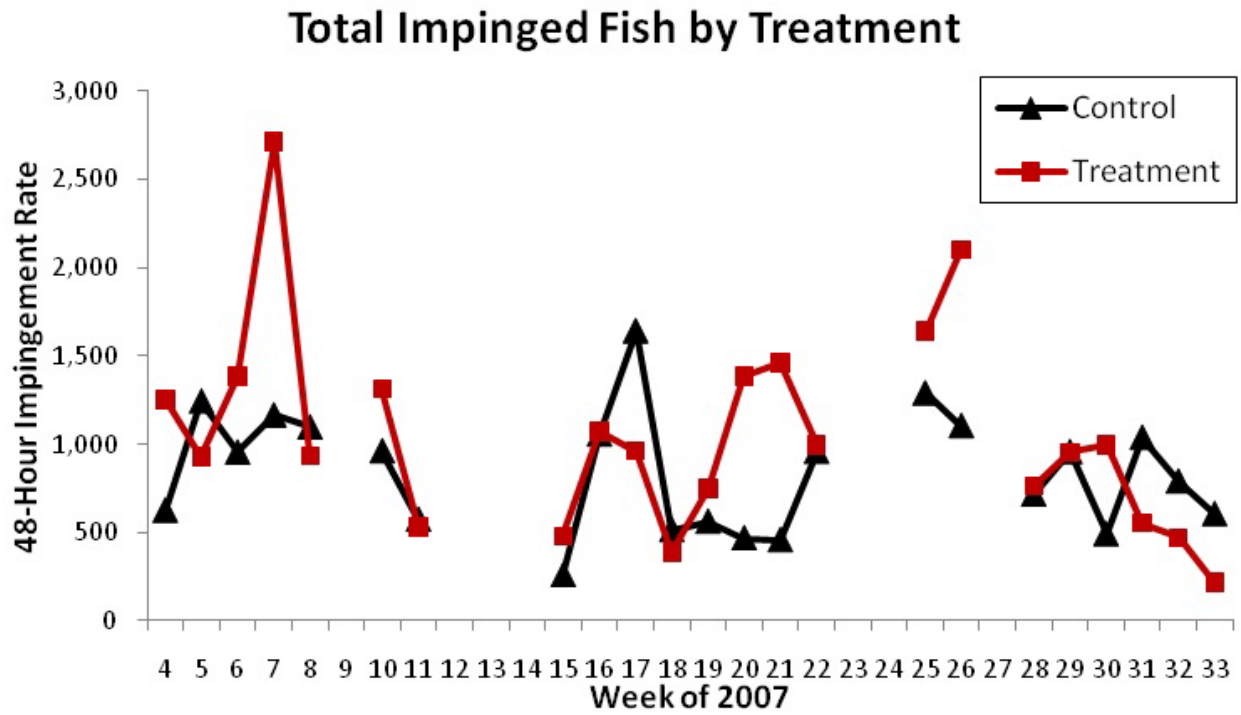
Figure 10. Total impingement of all species by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
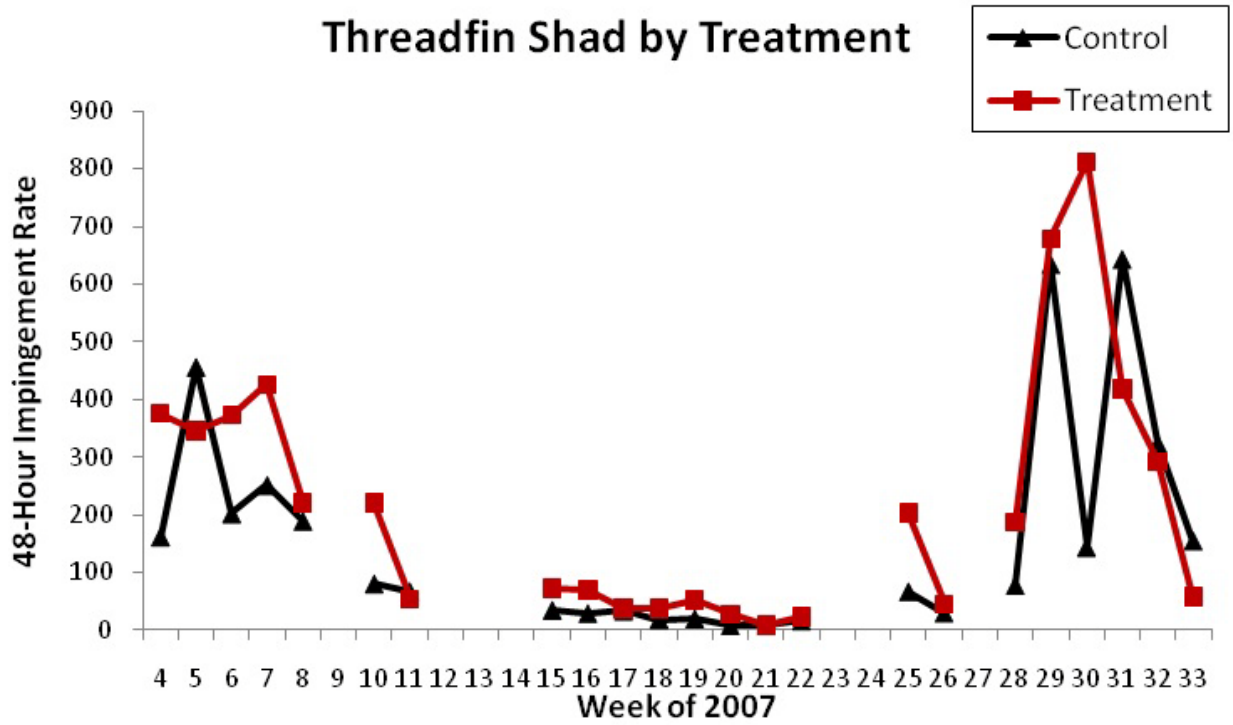
Figure 11. Impingement of threadfin shad by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
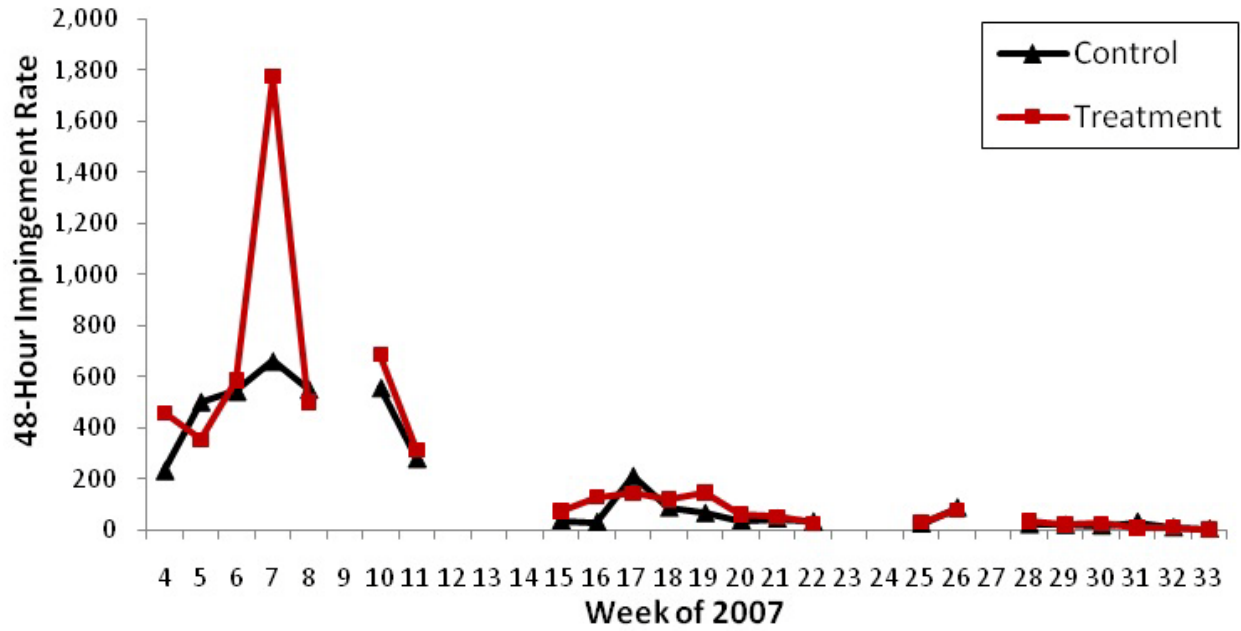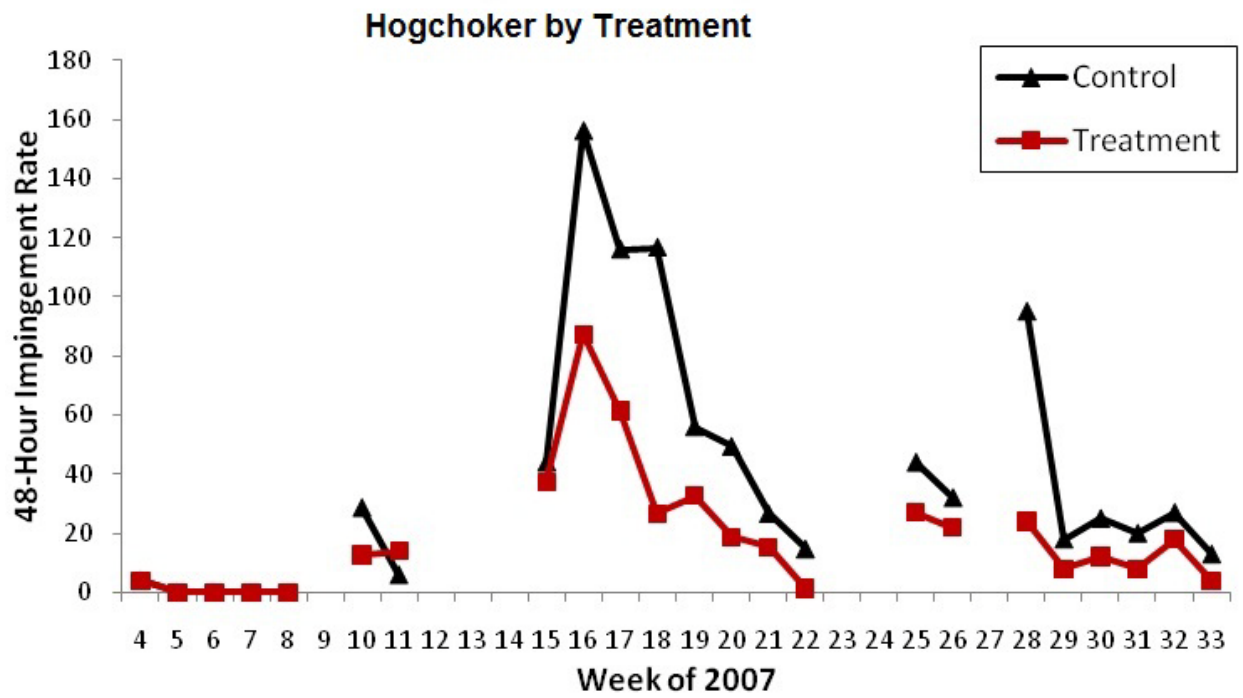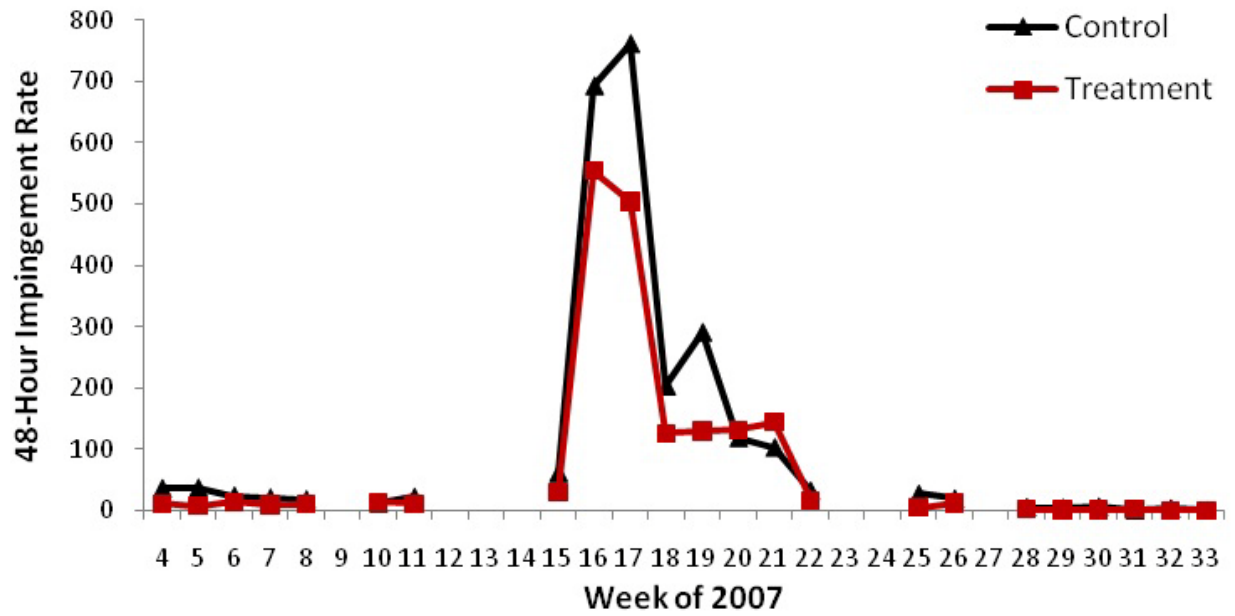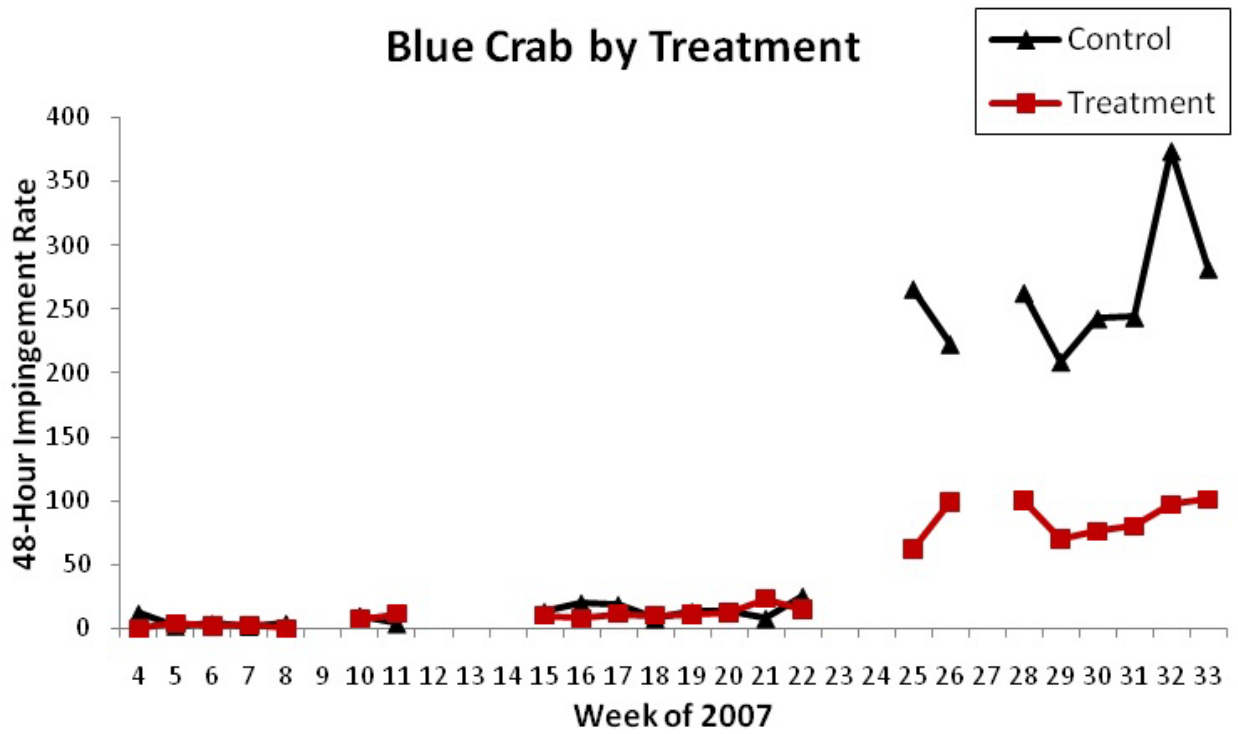
Figure 12. Impingement of blue catfish by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
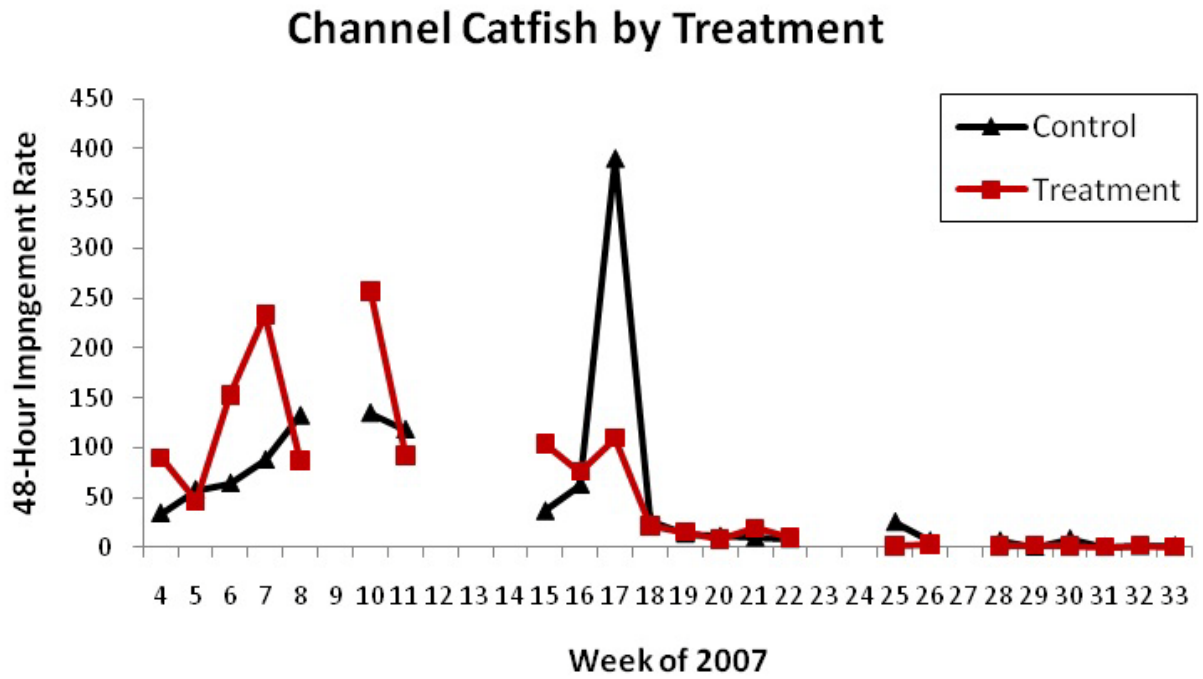
Figure 13. Impingement of channel catfish by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
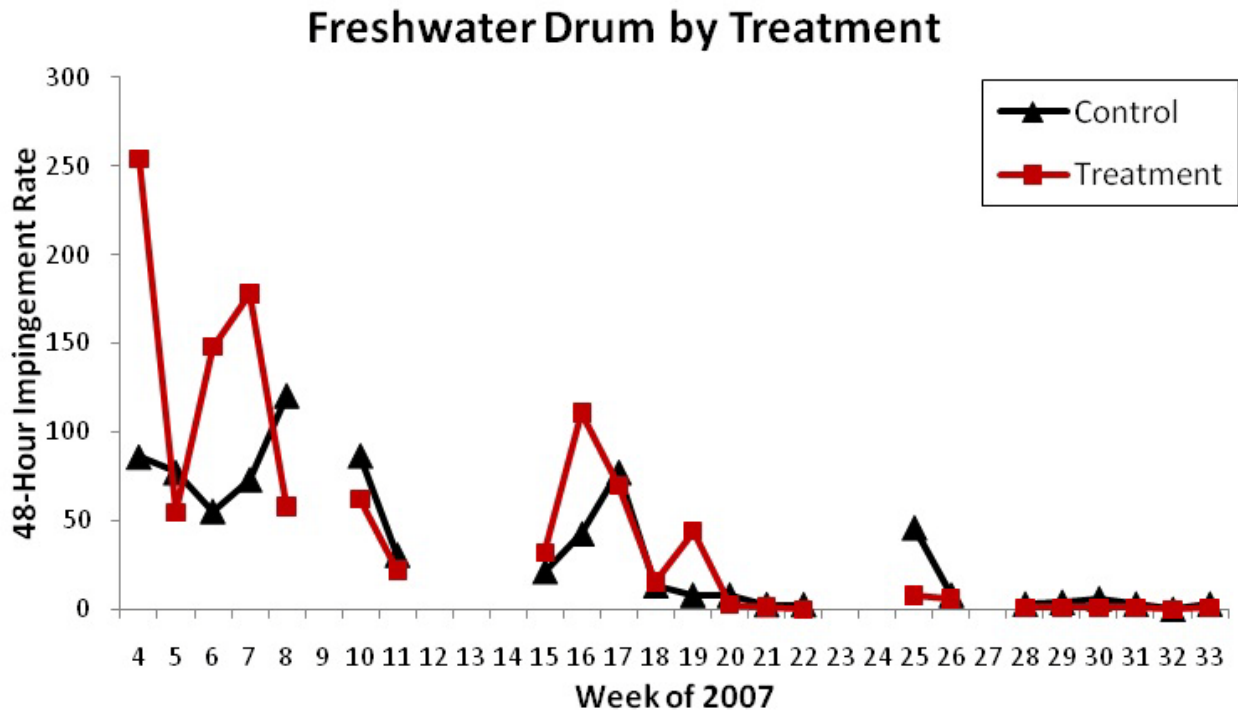
Figure 14. Impingement of freshwater drum by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
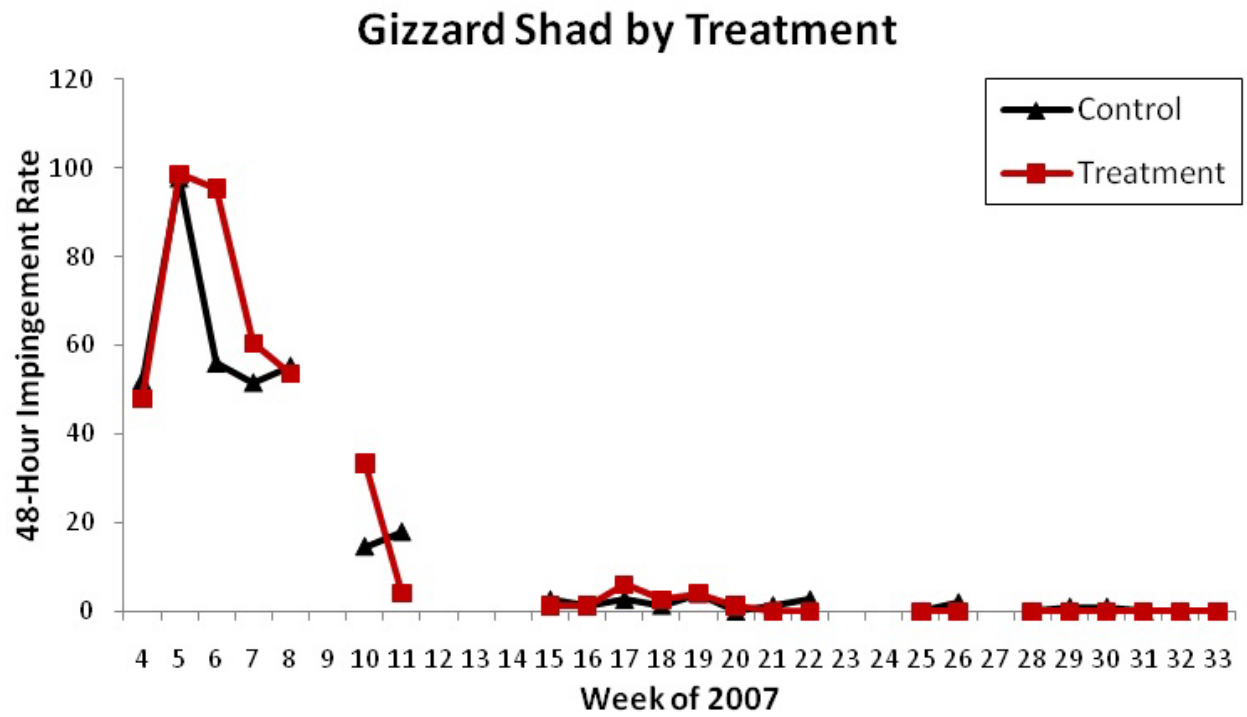
Figure 15. Impingement of gizzard shad by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
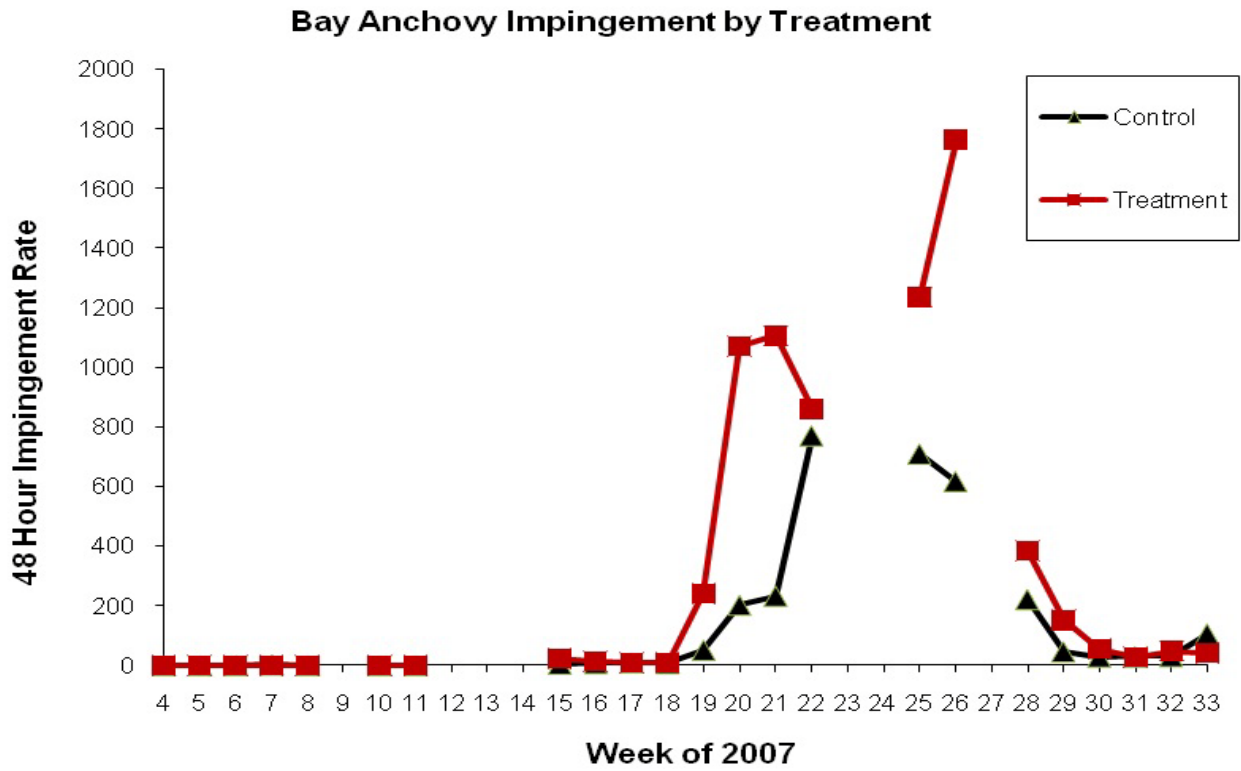
Figure 16. Impingement of bay anchovy by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.

Figure 17. Impingement of hogchoker by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.

Figure 18. Impingement of shrimp spp. by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.

Figure 19. Impingement of blue crab by treatment during the Screen Rotation Study at Plant Barry, Alabama in 2007.
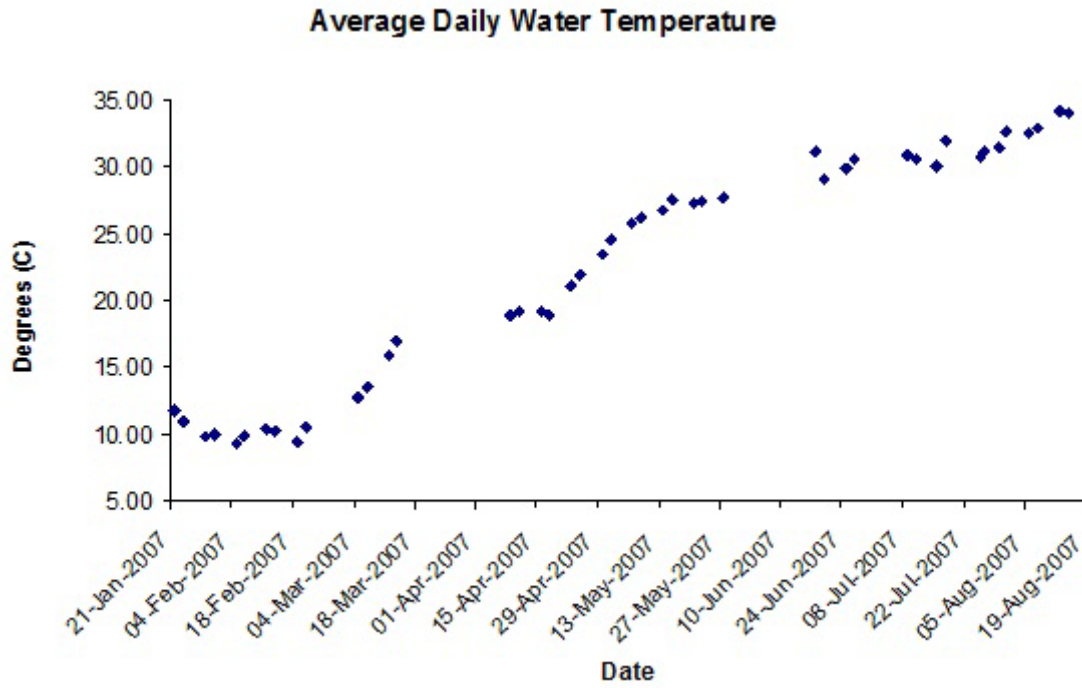
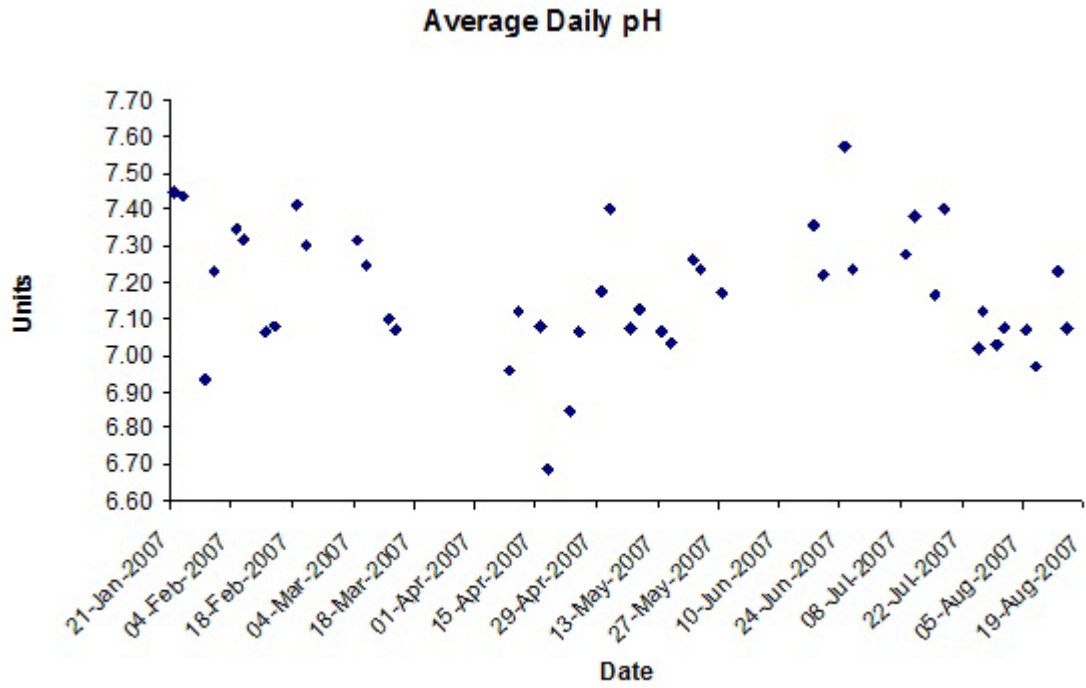Figure 20. Daily water temperature (average of all daily samples at both units) during the Screen Rotation Study at Plant Barry, Alabama in 2007.
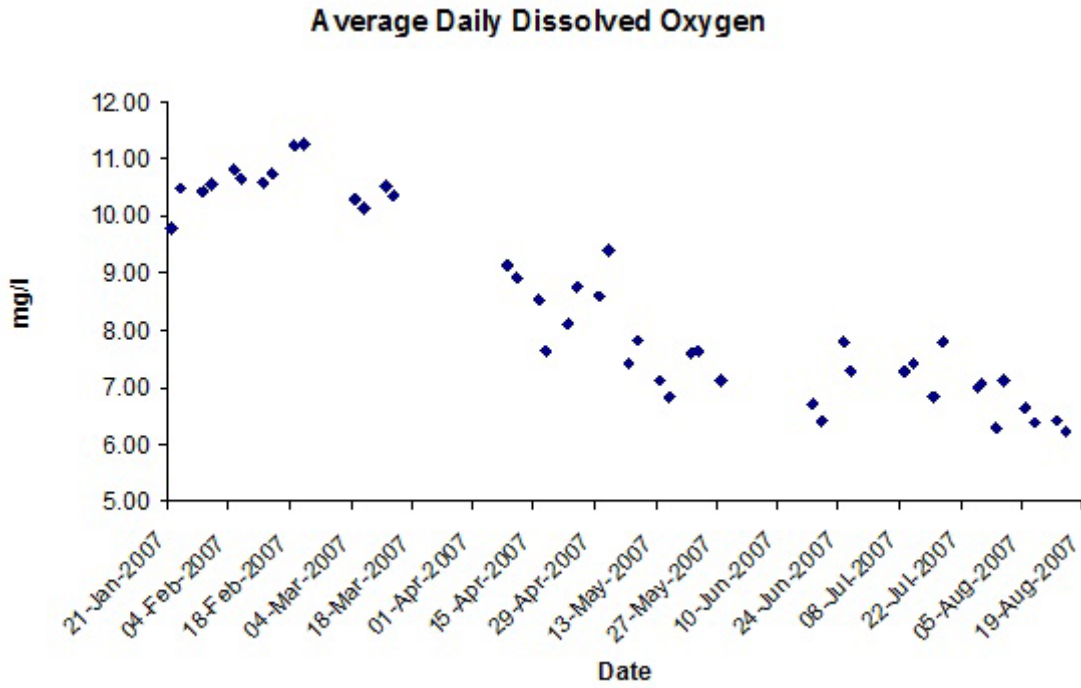
Figure 21. Daily dissolved oxygen (average of all daily samples at both units) during the Screen Rotation Study at Plant Barry, Alabama in 2007.
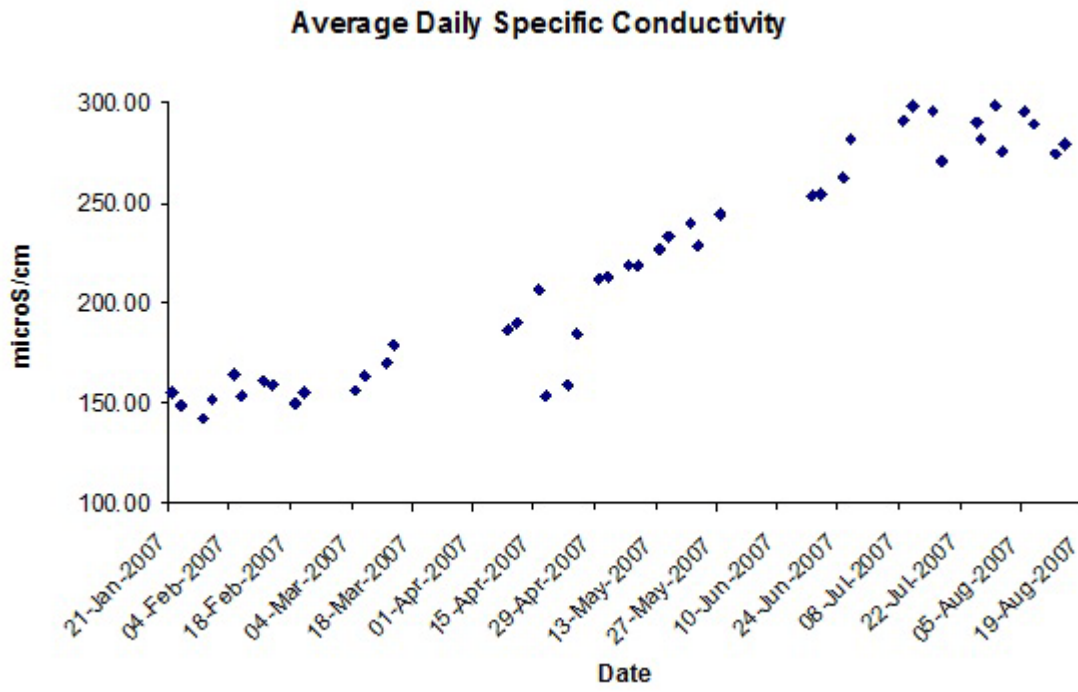
Figure 22. Daily specific conductivity (average of all daily samples at both units) during the Screen Rotation Study at Plant Barry, Alabama in 2007.
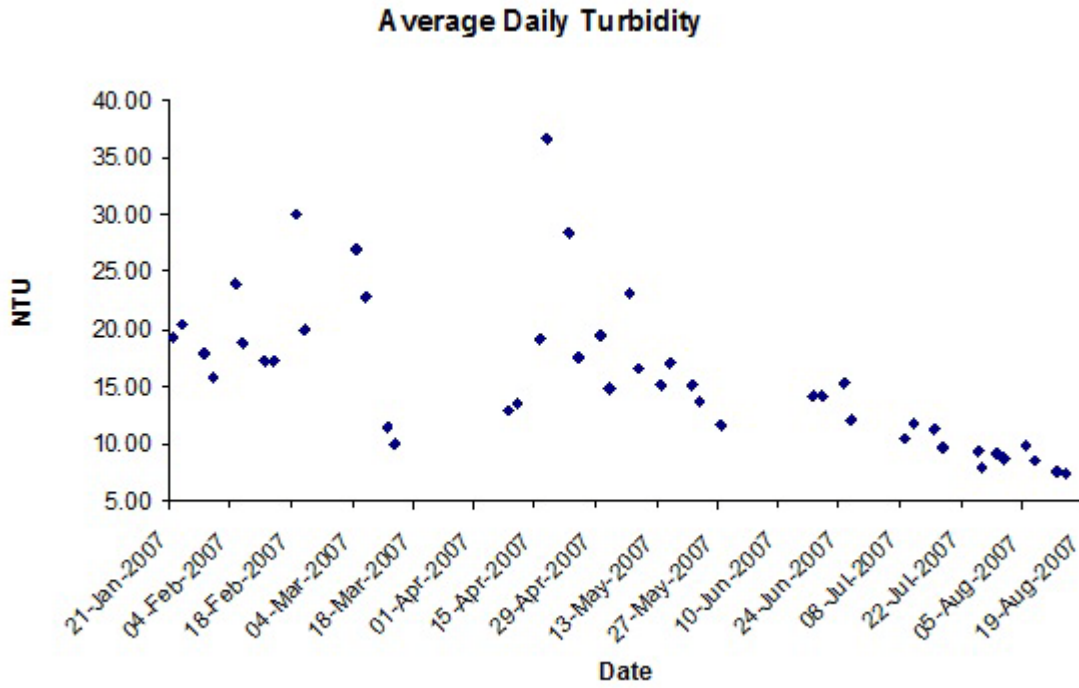
Figure 23. Daily turbidity indicated by NTU (Nephlometric Turbidity Units)(average of all daily samples at both units) during the Screen Rotation Study at Plant Barry, Alabama in 2007.

Figure 24. Daily pH (average of all daily samples at both units) during the Screen Rotation Study at Plant Barry, Alabama in 2007.
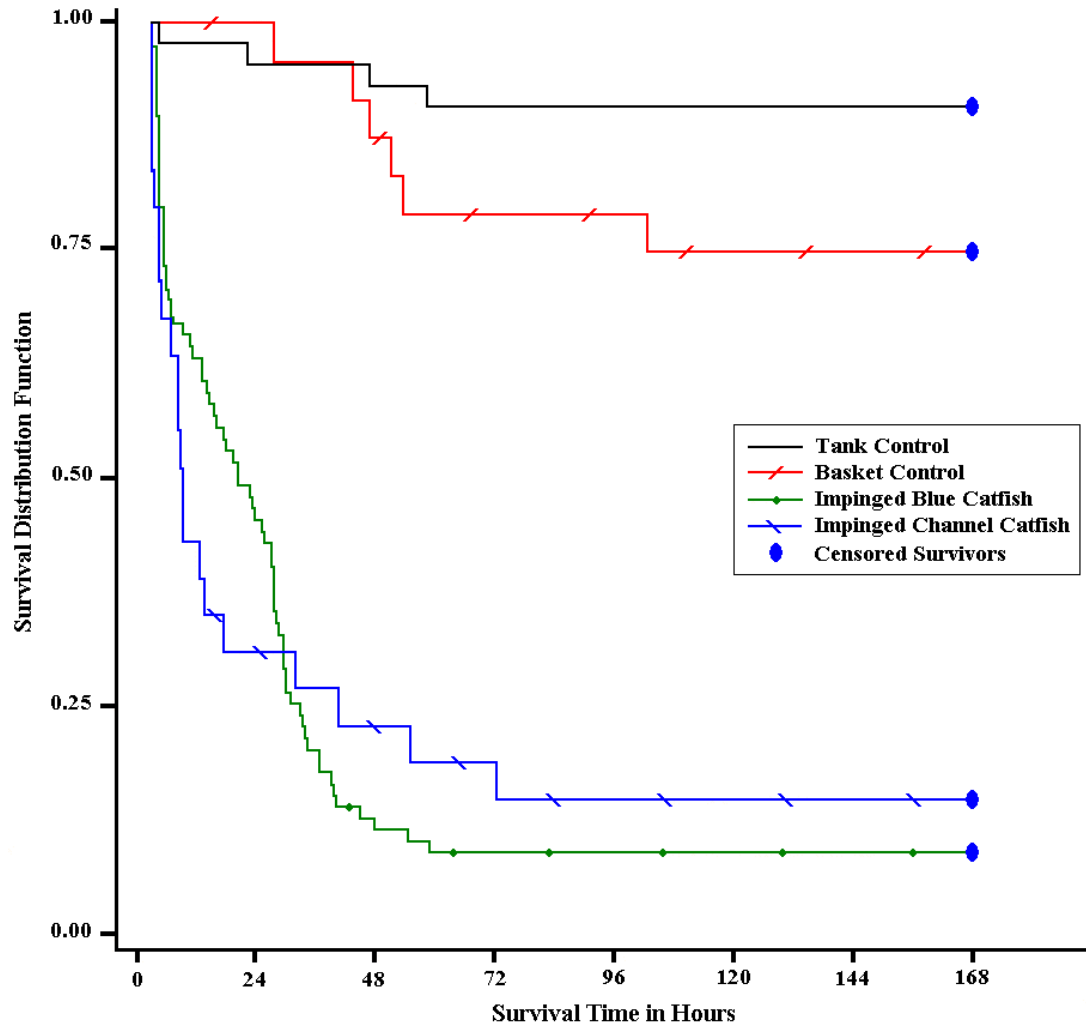
Figure 25. The Survival Distribution Functions of the four treatments in the Survival Study at Plant Barry, Alabama in 2007. Survival rates between the impinged catfish and the control groups were significantly different (p≤ 0.0001). There was no significant difference between the survival rates of impinged blue catfish and channel catfish (p≤ 0.9304) or between the aquaria controls and the basket controls (p≤ 0.0863).
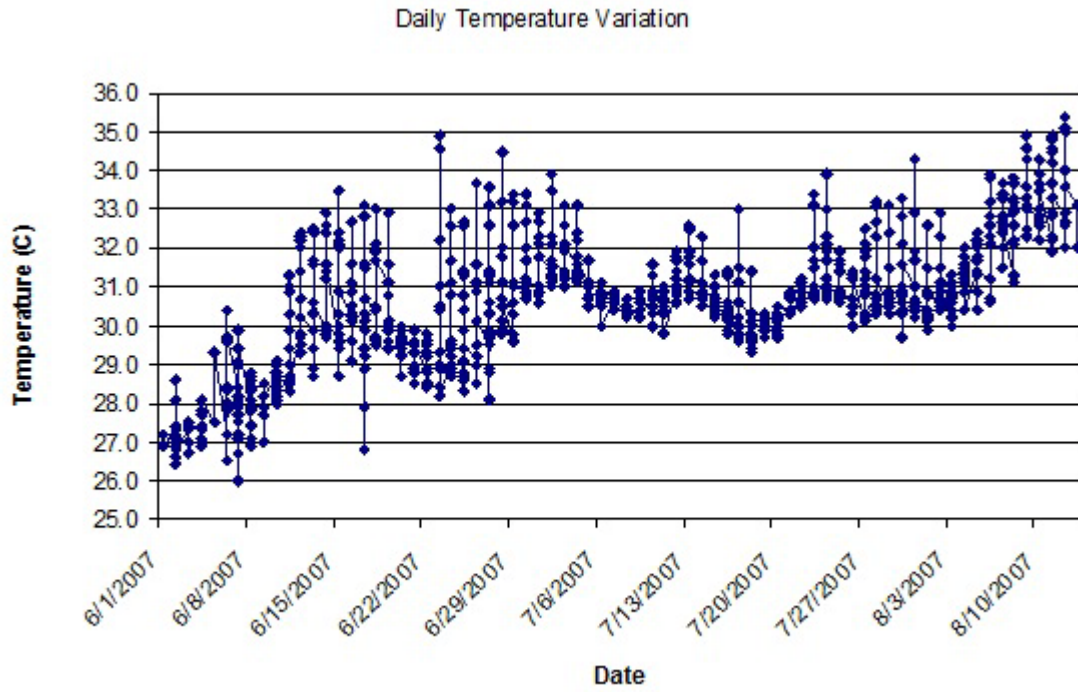
Figure 26. Daily fluctuations of aquaria water temperature during the Survival Study at Plant Barry, Alabama in 2007.
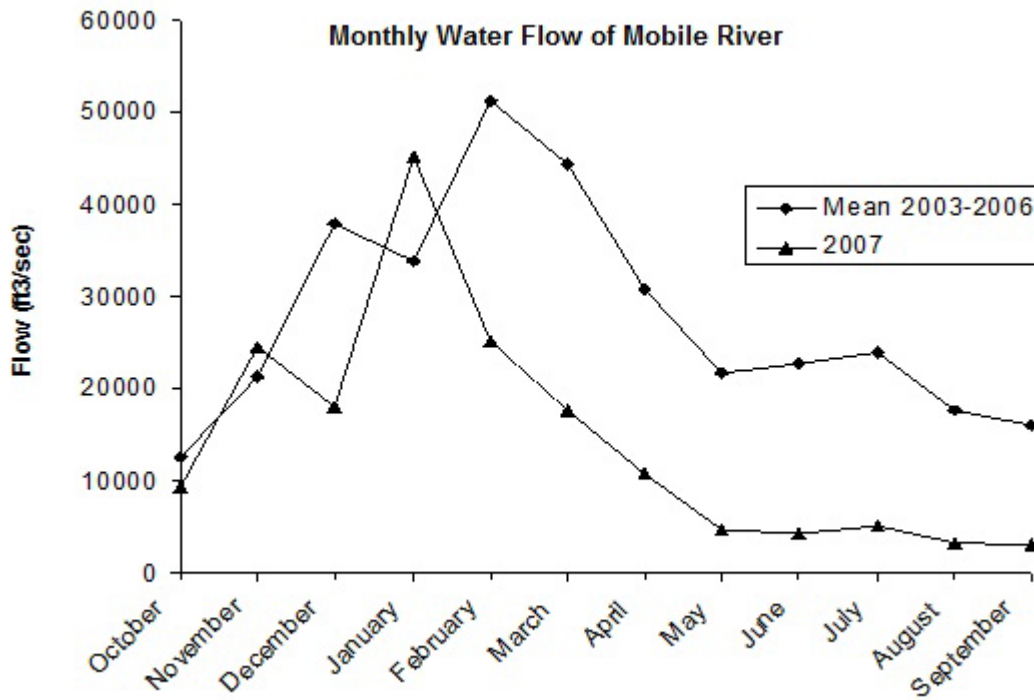
Figure 27. Comparison between the 2007 average monthly flow and the prior 4-year average flow at USGS Gage 02470629 River Mile 31.0 at Bucks, Alabama. This gage is located within the Plant Barry intake canal.
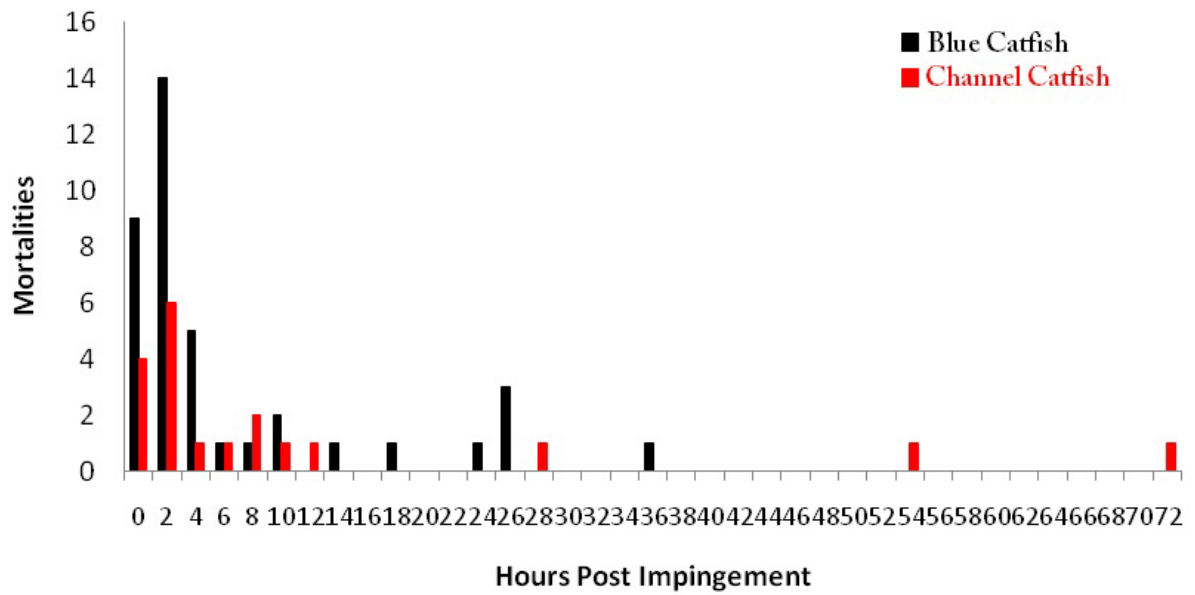
Figure 28. Three day mortality for Latent Impingement Mortalities from the Survival Study at Plant Barry, Alabama in 2007.