EFFECTS OF ANTHROPOGENIC STREAM ALTERATION ON BROWN TROUT HABITAT, MOVEMENT AND PHYSIOLOGY

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ABSTRACT

Anthropogenic stream alterations are caused by various factors, such as dams and water diversions that often change the flow, temperature and turbidity regimes of the aquatic ecosystems they occur in. Altered, unfavorable stream conditions are known to affect fishes at multiple levels of biological organization. These effects range from primary (e.g., blood cortisol concentrations) to secondary (e.g., serum chemistry) to tertiary levels (e.g., growth and condition) and can be manifested at any and all biological levels. Knowledge of fish responses to altered stream conditions at these levels of organization provides insight into population-level effects of anthropogenic alterations on brown trout *Salmo trutta*, which can be used to inform management decisions and mitigation actions.

The response of brown trout to altered temperature, turbidity and flow regimes in upper Esopus Creek, New York were evaluated at multiple biological levels during summers 2010 and 2011. Secondary and tertiary-level effects were compared between trout in a stream segment receiving cold, turbid water releases from an aqueduct originating in a nearby reservoir and trout in a stream segment above the confluence of the aqueduct (i.e., unaffected by releases from the aqueduct). A fish health assessment was conducted to evaluate secondary and tertiary-level responses in summer 2010, and radio-telemetry, capture-recapture and intensive habitat surveys were used to evaluate additional tertiary-level responses in summer 2011. Results from summers 2010 and 2011 indicated that trout in all reaches of upper Esopus Creek were stressed at the secondary (i.e., serum chemistry) and tertiary-level (i.e., body condition, growth rates and movement rates), and that trout immediately downstream from the aqueduct were less stressed. In addition, summer 2011 habitat data indicated that greater amounts of habitat that was optimal

for adult trout existed downstream from the aqueduct, and adults preferred these types of habitats.

Stream conditions upstream from the aqueduct were warm and low-flow (i.e., stressful for trout), and the water was clear and not turbid during both summers. Conversely stream conditions immediately downstream from the aqueduct were turbid (i.e., stressful for trout), fast-flowing and cold. Therefore, stream conditions throughout upper Esopus Creek during both summers were stressful for trout to some extent, and this was reflected in serum chemistry, body condition, movement and growth. Stream temperature is a dominant factor affecting fish growth and performance, which may explain why trout immediately downstream from the aqueduct were less stressed.

Results from the present study have local and broad implications. They could be used locally to inform future management decisions of the water and fishery resources of upper Esopus Creek, as well as to educate the various stakeholder groups of the stream and its resources. They could be used broadly to inform management decisions and mitigation actions to benefit fishery resources in other aquatic ecosystems affected by humans (e.g., tailwater trout fisheries).

BIOGRAPHICAL SKETCH

Tyler Jeffrey (T.J.) Ross was born in Oklahoma City, Oklahoma on September 10, 1985 to Mike and Debbie Ross. He grew up in Oklahoma City and, as a child, spent much time exploring creeks and ponds near his home. He graduated from Westmoore High School in May 2004 and received a Bachelor of Science Degree in Kinesiology from Oklahoma Baptist University (OBU) in May 2008. Early into his tenure at OBU, T.J. discovered his passion for the field of fisheries during a summer internship at the H.B. Parsons Fish Hatchery in Oklahoma City. He supplemented his OBU studies with ecology and environmental conservation courses, and then he attended Oklahoma State University after graduating from OBU in order to complete additional fisheries ecology and management coursework. He began a Master's program at Cornell University in the Department of Natural Resources in August 2009. He worked for the New York Cooperative Fish and Wildlife Research unit under the guidance of Dr. William L. Fisher.

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INTRODUCTION

Anthropogenic alteration of streams impairs aquatic ecosystems and affects fish habitats and populations (Poff and Allan 1995; Marchetti and Moyle 2001; Bunn and Arthington 2002; Kershner et al. 2004). Stream alterations are caused by various factors, such as human and industrial effluents, improper land-use and dams and diversions. Dams and diversions affect nearly all important ecological processes of a river or stream, ranging from altering the flow of water, sediment and nutrients to changing the composition of biota (Ligon et al. 1995). Diversions are used to supplement the energy, transportation, food production and water needs of humans (Kingsford 2000; Murchie et al. 2008), and they often impair the flow, temperature and turbidity regimes of the streams that receive their water (Nilsson et al. 2005). These impaired, often unfavorable conditions affect all stream biota (Bunn and Arthington 2002), ranging from aquatic invertebrates (Hax and Golladay 1998) to fish (Murchie et al. 2008), and the effects are usually negative (Poff and Zimmerman 2010).

Brown trout *Salmo trutta* are an economically and ecologically important sport-fish species (Elliott 1989; Van Winkle et al. 1998) that are affected by impaired stream conditions (Young et al. 2010). They respond to these conditions at all levels of biological organization, from the sub-organismal level (e.g., serum chemistry) to the population-level (e.g., mortality rates) (Triebskorn et al. 1997; Barton et al. 2002). As the biological complexity of fish responses increases, so does the ecological relevance (i.e., population-level responses are more ecologically relevant than sub-organismal level responses; Barton et al. 2002). Therefore, it is important to understand the effects of altered stream flow, temperature and turbidity on brown trout at all biological levels, because this understanding provides insight into the status and management of fish populations in streams that have been impacted by humans.

Brown trout and other salmonids respond to unfavorable stream flow, temperature and turbidity at all biological levels, from primary responses (e.g., blood plasma cortisol concentrations) to secondary responses (e.g., histopathology) to tertiary responses (e.g., growth and condition). For example, brown trout exposed to both high and low stream flows had elevated plasma cortisol concentrations (Flodmark et al. 2002), indicating that fish were stressed at the primary-level. Serum concentrations of alkaline phosphatase in rainbow trout *Onchorhynchus mykiss* decreased with increasing stream temperatures (Sauer and Haider 1977), indicating that fish were stressed at the secondary-level. Rainbow trout exposed to low summer flows had reduced growth rates (Harvey et al. 2006), indicating that fish were stressed at the tertiary-level. Additional responses of trout to unfavorable flow, temperature and turbidity include: increased movement and energy expenditure in elevated stream flow (Heggenes et al. 2007), reduced growth (Sigler at al. 1984) and survival (Harvey and Railsback 2009) in highly-turbid conditions, and reduced foraging and growth in thermally marginal habitats (Lee and Rinne 1980), with the general consensus that altered, unfavorable stream conditions are detrimental to trout. However, juvenile chinook salmon *Oncorhynchus tshawythsca* in the Snake River that were exposed to artificially increased flow and decreased temperature during summer had increased survival (Connor et al. 2003), which suggests that anthropogenic alterations of stream environments can sometimes have favorable effects on fish populations.

The present study evaluated the effects of altered stream flow, temperature and turbidity on the health, habitat, movement, growth and condition of brown trout in upper Esopus Creek, New York, with the underlying hypothesis that these stream conditions would affect trout at multiple levels of biological organization. Stream conditions in upper Esopus Creek are somewhat similar to those of the Snake River and other systems receiving hypolimnetic releases from dams. One segment of upper Esopus Creek receives flow augmentation from the hypolimnion of nearby Schoharie reservoir through an underground aqueduct, known as the Shandaken Tunnel. Stream conditions downstream from the confluence of the aqueduct are characterized by cold water temperatures and high flow and turbidity during summer; whereas, stream conditions upstream from the aqueduct are unaffected by the aqueduct releases and characterized by low flow and turbidity and higher temperatures during summer. Upper Esopus Creek is an ideal experimental stream because discharge from the Shandaken Tunnel influences habitat downstream from its confluence, but not upstream; thus, creating a tunnel-influence stream segment and a reference segment.

Knowledge of fish responses to altered stream conditions at multiple levels of organization can provide insight into potential population-level effects (Barton et al. 2002), which can then be used to inform fishery and water resource management decisions. Results from the present study provide important information regarding the effects of altered stream conditions on brown trout, and this information is relevant to ecological and sociopolitical issues regarding management of the fishery and water resources of upper Esopus Creek. Since the mid-1800s, Esopus Creek has been a renowned trout fishing destination that historically boasted reports of anglers catching up to 200 trout per day (CCES 2007b). In addition to excellent trout fisheries, upper Esopus Creek has been a component of the Catskill District Water Supply System for New York City since the construction of the Shandaken Tunnel in 1924 (CCES 2007a). Upper Esopus Creek angler groups, such as Trout Unlimited, have reported decreased catch rates of trout, as well as concerns regarding effects of the tunnel releases, specifically the chronically elevated turbidity, on resident trout populations (Catskill Mountains Chapter of Trout Unlimited v. City of New York 2001). In response to these concerns, fisheries managers and some anglers have proposed that the cool, hypolimnetic releases from the tunnel provide added thermal refuge habitat for trout in summer (CCES 2007a). In 2001, the Catskill Mountains Chapter of Trout Unlimited filed (and won) a civil suit against the New York City Department of Environmental Protection for the effects of the tunnel releases on resident trout (Catskill Mountains Chapter of Trout Unlimited v. City of New York 2001). The suit led to the requirement that New York City obtain a State Pollutant Discharge Elimination System permit to regulate the allowable flow, temperature and turbidity from the tunnel releases in order to protect trout and other biota (CCES 2007a). Although regulations and permitting are in place, uncertainty still remains regarding effects of the releases on resident trout populations. Cooperating agencies have undertaken extensive efforts to conserve and manage trout populations in upper Esopus Creek in response to the dispute over the effects of the tunnel releases. These efforts include research, extensive surveys, brown trout stocking programs and attempts to mitigate the effects of stream alteration on resident populations (CCES 2007b). The present study aimed to evaluate the response of brown trout to releases from the Shandaken Tunnel, which will provide information relevant to the stakeholder groups involved in the fisheries and water resource issues in upper Esopus Creek.

CHAPTER 1

EFFECTS OF ALTERED TEMPERATURE, TURBIDITY AND FLOW REGIMES ON BIOMARKERS OF BROWN TROUT HEALTH

Abstract

The sub-organismal and individual-level health of brown trout *Salmo trutta* in response to altered temperature, turbidity and flow regimes was evaluated in upper Esopus Creek, New York during summer 2010. Serum chemistry, parasite occurrence, gill histopathology, water content and diet data were compared between trout in a stream segment receiving cold, turbid water releases from an aqueduct originating in a nearby reservoir and trout in a stream segment located upstream from the confluence of the aqueduct (i.e., unaffected by the aqueduct releases). Resident trout populations and the New York City drinking water supply rely on water resources of upper Esopus Creek which are supplemented by releases from the aqueduct; therefore, information regarding potential effects of aqueduct releases on the health of resident trout is needed to better manage both the water and fisheries resources. Serum chemistry indicated that trout in all study reaches were stressed, but that trout immediately downstream from the aqueduct were less stressed. Parasite occurrence was low (3 - 9%; range) and similar among reaches. Gill histopathology scores were highest (2.27 ± 1.27; mean ± SD) in fish from the reach farthest downstream from the aqueduct, but were generally low (1.13 - 2.27) which indicated mild stress for trout in all reaches. Water content levels in trout from all reaches were high (72.6 - 81.5%) which indicated that trout from all reaches were approaching nutritional and physiological stress. The composition and relative mass of gut contents were similar in trout from all reaches of the stream. Stream conditions throughout upper Esopus Creek during summer were stressful for trout at some level, and this was reflected in serum chemistry and

water content. There was some evidence of a stress-refuge immediately downstream from the aqueduct portal; however, it is unknown whether or not this refuge was manifested at the population level.

Introduction

Anthropogenic alteration of stream environments can affect fish health and the condition of their populations (Sauer and Haider 1977; Marchetti and Moyle 2001). Stream conditions can be affected by a variety of anthropogenic sources (e.g., human and industrial effluents, improper land-use, and dams and diversions), and fish responses to impaired conditions vary, occurring at both the sub-organismal (e.g., impairment of gill function) and population-levels (e.g., fish communities) (Triebskorn et al. 1997). Although fisheries biologists traditionally use morphometric measures of fish condition to obtain a generalized indication of fish health in response to unfavorable stream conditions, a more holistic approach is to assess fish health using a series of biological markers, or biomarkers [as defined by Triebskorn et al. (1997)], that progressively increases in complexity (i.e., primary, secondary and tertiary-level biomarkers). The ecological relevance of biomarkers increases with increasing biological complexity (i.e., tertiary responses are more informative than primary; Adams et al. 1989; Adams 1990); thus, a hierarchical approach evaluates primary, secondary and tertiary-level effects of stream alteration on fishes in order to gain insight into potential population-level effects (Sanchez and Porcher 2009). Biomarkers detect responses ranging from changes in serum chemistry due to organ tissue damage (Manera and Britti 2006) to histopathological lesions due to altered gill and organ function (Bernet et al. 1999) to parasitic infections due to compromised immune system function (Khan and Thulin, 1991; Poulin 1992) to poor body condition due to impaired physiological function (Peters et al. 2007; Hartman and Brandt 1995). Each of these responses provides valuable insight into the effects of stream alteration on fish populations (Schmidt-Posthaus et al. 2001; Burkhardt-Holm and Scheurer 2007).

Serum chemistry, histopathology, parasite evaluation and water content are four biomarkers commonly used in fish health assessments (Folmar et al. 1993; Adams et al. 1996; Bernet et al. 2000). Serum chemistry is a secondary-level biomarker used to compare serum concentrations of fish between impaired and unimpaired stream reaches (Bernet et al. 2001) and can also be used to compare serum concentrations of fish to literature-derived, baseline (i.e., normal) concentrations (Escher et al. 1999; Bernet et al. 2001). Serum concentrations in fish that differ from normal baseline concentrations indicate stress responses (Manera and Britti 2006), which can then be manifested at higher levels of organization such as histopathological lesions, parasites and body condition. Histopathological lesions are also secondary-level biomarkers that are early-indicators of the effects of environmental changes and contaminants on fish (Schlenk et al. 2008). Lesions can be detected using histopathology, and they often degrade internal organs which can cause enzyme leakage into the bloodstream and ultimately induce changes in serum chemistry (Hille 1982). In addition, histopathological lesions on the gill tissues of fish are the direct, physical manifestation of exposure to elevated suspended sediment levels (Adams 2001; Sutherland and Meyer 2007). Parasite evaluations are tertiary-level biomarkers that assess fish health in response to their physical and chemical environments (Landsberg et al. 1998). Extensive parasitic infections are typically greater in stressed, unhealthy or immuno-compromised fishes (Conte 2004), but parasites occur on and in healthy and unhealthy fish in all environments (Barber et al. 2000). Assessment of fish water content (i.e., tertiary-level biomarker) provides insight into the physiological and nutritional condition of fish (Peters et al. 2007), is a whole-animal measure of performance (Barton et al. 2002) and is more precise than traditional condition measures (Trudel et al. 2005). Peters et al. (2007) found chinook salmon *Oncorhynchus tshawytscha* with water content levels in excess of 78% to be in poor condition and at risk of severe nutritional and physiological stress, suggesting that 78% was a stress-threshold for salmonids. A hierarchical fish biomarker assessment using serum chemistry, histopathology, parasite evaluation and water content provides a better understanding of the effects of stream alteration on the status and health of fish populations.

Biomarkers have been used historically to evaluate the effects of chemical pollutants on fish; however, they also have been used by numerous investigators to evaluate the effects of unfavorable environmental conditions, such as atypical temperature, turbidity and flow on individual fish and their populations (Barton et al. 2002). The response of rainbow trout *Oncorhynchus mykiss* to upper and lower temperature extremes was evaluated using a serum chemistry assessment (Wagner et al. 1997). More parasites were found on eels *Anguilla rostrata* under increased flow and acidity conditions when compared to eels in unimpaired conditions (Barker and Cone 2000). Histopathological examination revealed that low levels of suspended sediment did not affect the gill tissue of coho salmon *Oncorhynchus kisutch* and steelhead (Redding et al. 1987). Biomarkers have not, however, been used to evaluate the effects of altered stream temperature, turbidity and flow regimes on brown trout populations, and it is not known whether or not these types of assessments would be informative.

Upper Esopus Creek, in the Catskill Mountains of southeastern New York, supports a renowned brown trout fishery and is one of several streams that contributes to New York City's drinking water supply through a system of aqueducts and reservoirs. The stream receives additional water from the nearby Schoharie Reservoir through an underground aqueduct, the Shandaken Tunnel, which is confluent with upper Esopus Creek at the Shandaken portal. Water entering upper Esopus Creek through the portal originates in the hypolimnion of the reservoir and is cool in summer, sometimes turbid, and it substantially increases the volume of flow in the stream. Summer base flows and turbidity are general lower, and water temperatures typically higher in reaches upstream from the portal than they are downstream. Stream angler groups, such as Trout Unlimited, have expressed concerns about the effects of the portal releases on the health of local trout populations; however, fishery managers and some anglers have asserted that the cool-water releases provide added thermal refuge habitat for trout during the warmer summer months (CCES 2007a). These different perspectives led to the present effort to assess the effects of the portal releases on brown trout populations in upper Esopus Creek using a hierarchical biomarker assessment. The main objective of the present study was to generate fish health data that could be used to better inform management decisions that could impact local fishery and water supply resources.   Differences in serum chemistry, gill histopathology, parasite occurrence and water content were investigated in resident brown trout that were sampled both upstream and downstream from the portal.

Methods

Study Area. – Upper Esopus Creek is located in the south-central Catskill Mountain Region of southeastern New York (Figure 1.1). The stream begins at Winnisook Lake on Slide Mountain (1,274 m above sea level), travels 41.8 kilometers around the base of Panther Mountain where it enters the Ashokan Reservoir (193 m above sea level). The upper Esopus Creek watershed is approximately 309 square kilometers, 95% forested, and entirely contained within the Catskill State Park; the remaining 5% of the watershed is comprised of commercial and residential development. The Shandaken Tunnel is confluent with upper Esopus Creek east of Shandaken, New York and west of Phoenicia, New York (Figure 1.1). Water discharged from the portal has the highest median turbidity (8.8 nephelometric turbidity units [NTU]) of streams in the watershed; however, its contribution to the total annual sediment load of upper Esopus

Creek is considered negligible (CCES 2007a). Tributaries to upper Esopus Creek are comprised of clay-rich, glacial till channels and contribute a significant portion of the total annual sediment load during heavy rain and snow-melt events (CCES 2007a). The portal can discharge 2.5 million cubic meters of water per day to upper Esopus Creek when operated at maximum output. Water releases are regulated by a State Pollutant Discharge Elimination System permit that places limits on flow, turbidity and temperature to preserve stream biota and habitat.

Eight sites were sampled in upper Esopus Creek, three located upstream and five downstream from the portal (Figure 1.1). Releases from the Shandaken Tunnel have the greatest impact on stream conditions within the first 3.2 km downstream from the portal (CCES 2007a); hence, the three upstream sites were grouped into the upstream reach, the first two downstream sites into the tunnel-influence reach and the three remaining downstream sites into the downstream reach. Ten brown trout averaging 196.5 ± 47.5 mm (mean total length ± SD) were collected from each site using back pack electrofishing (Smith-Root LR-24, Smith-Root, Vancouver, Washington, USA). Trout were then immediately euthanized with an aqueous solution of 500 mg/L of tricaine methanesulfonate (Western Chemical Inc., Ferndale, Washington, USA), buffered 1:1 (weight:weight) with sodium bicarbonate (Sigma-Aldrich, St. Louis, Missouri, USA) and immediately bled. Blood was collected from the caudal artery and vein using the Vacutainer system (Becton-Dickinson, Rutherford, New Jersey, USA) with lithium heparin as anticoagulant.

Blood samples and the trout were transported to the nearby Cornell Cooperative Extension of Ulster County office in Phoenicia, NY for processing within an hour of collection. Wet-mounts of skin scrapes and gill clips were prepared and examined with a compound microscope for the presence or absence of parasites. Gill arches were collected, fixed with 10% neutral buffered formalin and transported to the College of Veterinary Medicine at Cornell University for preparation of histopathological slides and subsequent evaluation of gill histopathology. Each sample was thoroughly examined and scored for the presence of gill lesions. Scoring was qualitative and on a scale from one to five, with one indicating absence of lesions and five indicating severe lesions. Muscle chunks (1.8 ± 1.4 grams) were collected from the dorsal surface of each trout using a scalpel and beginning immediately posterior to the operculum, extending to the lateral line and ending anterior to the dorsal fin. Muscle chunks were blot dried, weighed to the nearest hundredth of a gram, placed into a drying oven at 55°C for ten days and then re-weighed to calculate water content of each sample. Gastrointestinal tracts were removed from all trout, and the composition and relative wet mass of gut contents were assessed. Gastrointestinal tracts were blot dried, and the mass of all tracts was measured prior to removing, identifying to order and counting all gut contents. The mass of the empty gastrointestinal tracts was measured after removing all gut contents in order to calculate the wet mass of the contents. The calculated wet mass of the gut contents was divided by the wet mass of the fish to calculate the relative wet mass of the gut contents. Vacutainer tubes containing the blood samples were centrifuged at 1500 x gravity for two minutes, and a minimum of 600 µL of plasma were collected. The plasma samples were submitted to the Clinical Pathology Laboratory, Animal Health Diagnostic Center, College of Veterinary Medicine, Cornell University for automatic blood chemistry analysis (routine small animal panel) immediately after collection. All plasma samples were analyzed for alkaline phosphatase, alanine aminotransferase (ALT), aspartate aminotransferase (AST), amylase, creatine kinase, creatinine, gamma-glutamyl transferase, bicarbonate, total, direct and indirect bilirubin, blood urea nitrogen, cholesterol, glucose, total protein, albumin, globulin, calcium, iron, sodium, potassium, chloride, magnesium, phosphate, and total iron binding capacity using an automated analyzer (Boehringer Mannheim/Hitachi Modular P System, Roche Diagnostics Corp., Indianapolis, Indiana, USA). Additionally, estimates of sodium/potassium and albumin/globulin ratios, percent saturation of iron binding, and anion gap were derived from the preceding analytes. Normal baseline serum concentrations used for comparison were for rainbow trout, and they were derived from Walker and Fromm (1976), Hille (1982) and Manera and Britti (2006).

Differences in serum chemistry, histopathology, parasite occurrence, water content and gut content data were evaluated with parametric and non-parametric statistical analyses. Prior to analysis, parametric data were tested for normality using Shapiro-Wilk tests (JMP, Version 7. SAS Institute Inc., Cary, North Carolina, USA) and transformed to obtain normality, as needed. Values for serum analytes that were normally distributed included: anion gap, sodium/potassium ratio, calcium, phosphate, magnesium, total protein, globulin, glucose, alkaline phosphatase, cholesterol, iron and percent saturation. Values for serum potassium, AST, amylase, creatine kinase and total iron binding capacity were log-transformed; sodium and chloride data were sine-transformed; albumin and albumin/globulin ratio data were cosine transformed; and bicarbonate, blood urea nitrogen, creatinine, ALT and total bilirubin data could not be transformed to obtain normality. Chi-square tests of independence were used to evaluate differences in parasite occurrence in trout among reaches; analysis of variance was used to test for differences in serum analytes, histopathological scores, water content levels and gut content wet mass and diet composition in trout among the reaches. If reaches were significantly different, we used Tukey-Cramer honestly significant difference (HSD) tests to evaluate differences among the mean values from individual reaches. Wilcoxon Ranked-Sums tests were used for all comparisons of non-normally distributed data.

Results

Throughout summer 2010, the tunnel-influence reach was cooler than the upstream and downstream reaches, and the tunnel-influence and downstream reaches were more turbid and faster flowing than the upstream reach (Table 1.1). Stream temperature was highest in the downstream reach, lowest in tunnel-influence reach and intermediate in upstream reach; turbidity was highest in tunnel-influence reach, lowest in the upstream reach, and intermediate in downstream reach; and discharge was highest in the downstream reach, lowest in the upstream reach, and intermediate in the tunnel-influence reach (Table 1.1).

Serum concentrations of sodium, chloride, calcium, albumin, alkaline phosphatase and total bilirubin were similar among reaches (Table 1.2). Trout in the upstream reach had significantly higher serum globulin and total protein concentrations than trout in the tunnel-influence reach, higher magnesium concentrations than trout in the downstream reach, and higher phosphate concentrations than trout in tunnel-influence and downstream reaches (Table 1.2). Trout in the downstream reach had significantly higher potassium concentrations than trout in the tunnel-influence reach (Table 1.2). Although not significant, trout in the upstream reach had higher iron and cholesterol concentrations than trout in tunnel-influence and downstream reaches and higher magnesium concentrations than trout in the tunnel-influence reach; trout in the downstream reach had higher glucose concentrations than trout in the upstream and tunnel-influence reaches and higher concentrations of potassium than trout in upstream reach (Table 1.2).

A significant pattern was observed in serum AST (F = 7.92, DF = 29, P = 0.002), ALT (X^2 = 10.05, DF = 2, P = 0.007) and creatine kinase (F = 8.18, DF = 29, P = 0.002) in trout among the three reaches (Figures 1.2 and 1.3). Trout in the tunnel-influence reach had the lowest AST, ALT and creatine kinase concentrations, followed by trout in the downstream and upstream reaches. Although not significant, the same pattern was observed in serum globulin (Figure 1.3), potassium, total protein, glucose and total bilirubin (Table 1.2). The inverse pattern was observed in serum creatinine (X^2 = 7.44, DF = 2, P = 0.02), with the highest concentrations in trout from the tunnel-influence reach, followed by trout in the downstream and upstream reaches (Figure 1.4). The same pattern, though not significant, was observed in bicarbonate and amylase (Table 1.2).

Concentrations of potassium, phosphate, magnesium, alkaline phosphatase, creatinine and ALT were below normal baseline at all reaches, and total protein, total bilirubin, albumin and cholesterol were above normal baseline at all reaches (Table 1.2). Concentrations of iron were above normal baseline in trout from the upstream reach and below normal baseline in trout from the tunnel-influence and downstream reaches (Table 1.2). Concentrations of AST, globulin and creatine kinase were above normal baseline in trout from the upstream and downstream reaches and below normal baseline in trout from the tunnel-influence reach (Figures 1.2 and 1.3).

Histopathology scores were significantly higher (i.e., more severe lesions) in trout from the downstream reach than in trout from upstream and tunnel-influences reaches; however, the scores were low in fish from all reaches (Table 1.3). Parasite occurrence was similar in trout among the three reaches (X^2 = 3.06, DF = 2, P = 0.22). Parasites occurred in 3 of 32 (9.4%) trout from the upstream reach, 0 of 19 (0.0%) trout from the tunnel-influence reach and 1 of 26 (3.8%) trout from the downstream reach. Water content levels did not differ significantly in trout sampled from the three reaches (F = 2.63, DF = 77, P = 0.08), and most measures were close the 78% threshold for stress (Figure 1.5). Although they did not differ significantly, water content levels were lower in trout from the downstream reach than in trout from the upstream and tunnel-influence reaches (Figure 1.5). The wet mass of gut contents ($F = 1.28$, $DF = 74$, $P = 0.28$; Figure 1.6) and diet composition (Table 1.4) were similar in trout among the reaches, with the exception of greater numbers of true bugs *Hemiptera* present in diets from trout in the upstream reach.

Discussion

The hierarchical biomarker assessment presented herein indicated that unfavorable stream temperature, turbidity and flow regimes affected the health of individual brown trout in upper Esopus Creek during summer 2010. Such effects on fish behavior (e.g., movement and habitat preferences) (Armstrong et al. 1998; Vehanen et al. 2000), physiology (e.g., growth rates) (Weisberg and Burton 1993) and populations (e.g., mortality rates) (Annear et al. 2002) are well known. Concentrations of many serum analytes in brown trout from all study reaches of upper Esopus Creek were found to differ from normal baseline, but only a few revealed significant differences among the three reaches. Trout in all reaches had low numbers of parasites, mild severity gill lesions, water content levels that were only slightly below the threshold for stress, and the composition and wet mass of gut contents were similar in trout from all reaches. These results were generally consistent with those from several other studies evaluating the response of salmonids to impaired stream conditions. Serum chemistry (i.e., plasma cortisol) in brown trout within an artificial stream changed in response to fluctuating flows that mimicked conditions often encountered below dams and diversions; the stream conditions were similar to those downstream from the portal in upper Esopus Creek during summer (Flodmark et al. 2002). Serum chemistry (i.e., plasma glucose, chloride and cortisol) was affected in rainbow trout when exposed to upper or lower temperature extremes (Wagner et al. 1997); the upper extremes were comparable to water temperatures observed in upper Esopus Creek upstream from the portal during summer. Gill structure and function were not affected in coho salmon and steelhead that were exposed to moderately elevated turbidity levels (1 - 11 NTU; Redding et al. 1987) which were similar to the mean turbidity levels measured in upper Esopus Creek downstream from the portal during summer.

Serum chemistry and water content biomarkers indicated that brown trout in all reaches of upper Esopus Creek were stressed during summer 2010. When interpreting serum chemistry data, it is important to consider the potential causes and health implications of different analyte concentrations (Triebskorn et al. 1997). Alkaline phosphatase, an enzyme that removes phosphate groups from various molecules, was measured in decreased concentrations (i.e., less than 179.2 ± 19.3 $U \cdot L^{-1}$) in rainbow trout that had been exposed to elevated thermal regimes (19 - 20°C) (Sauer and Haider 1977). Stream temperatures were generally high in the upstream (19.5 ± 2.5°C) and downstream (19.7 ± 2.6°C) reaches of upper Esopus Creek where trout were found to have abnormally low alkaline phosphatase concentrations (61.3 ± 22.5 $U \cdot L^{-1}$, US and 54.7 ± 21.4 $U \cdot L^{-1}$, DS). Total protein, the sum of serum albumin and globulin, often reaches elevated concentrations (i.e., greater than 3.6 ± 0.1 $g \cdot dL^{-1}$) in fish due to stress when environmental conditions deviate from their normal ranges (Hille 1982; Wendelaar et al. 2008). Elevated concentrations of total protein were observed in trout from the tunnel-influence (3.7 ± 0.45 $g \cdot dL^{-1}$) and downstream reaches (4.1 ± 0.3 $g \cdot dL^{-1}$) where turbidity and discharge were consistently high and in trout from the upstream reach (4.5 ± 0.6 $g \cdot dL^{-1}$) where stream temperatures were consistently high. Albumin, a serum protein that maintains fluid volume within the vascular space, often reaches elevated concentrations (i.e., greater than 1.4 ± 0.1 $g \cdot dL^{-1}$) in fish that have exercised strenuously, such as from increased movement (Heath 1995; Bernet et al. 2001). Trout actively displace and increase rates of activity in turbid, high-flow conditions (Clapp et al. 1990), like those present in the tunnel-influence and downstream reaches of upper Esopus Creek, and albumin concentrations exceeded normal baseline concentrations in trout from these reaches (1.6 ± 0.2 $g \cdot dL^{-1}$, TI and 1.6 ± 0.2 $g \cdot dL^{-1}$, DS). Water content is a more precise proxy for body condition (Trudel et al. 2005; Peters et al. 2007), and body condition data are useful for understanding the effects of chronic environmental stressors on fish populations (Barton et al. 2002). Trout in all study reaches of upper Esopus Creek had water content levels only slightly below the 78% stress threshold proposed by Peters et al. (2007), indicating that trout in all reaches were on the verge of expressing a higher-level stress response. The body condition of fish is correlated to diet (i.e. consumption; Bowen 1996), but the gut content composition and relative wet mass were similar in trout from all study reaches, which suggests that diet did not affect the body condition of trout. Elevated stream temperature (Elliott 2000) and turbidity (Redding et al. 1987) and reduced summertime flow (Xu et al. 2010a) have been found to stress trout. The reach upstream from the portal was characterized elevated stream temperature and reduced summertime flow, the tunnel-influence reach was characterized by elevated turbidity and the downstream reach was characterized by elevated stream temperature and turbidity; therefore, the serum chemistry responses and poor body condition of trout throughout upper Esopus Creek could reasonably be attributed to the stressful summertime conditions present at all reaches.

Several serum analytes from the present study provided evidence that brown trout within the tunnel-influence reach were less stressed than those from the upstream or downstream reaches. Aspartate aminotransferase, a key enzyme in protein to carbohydrate metabolism, has been observed in elevated concentrations (i.e., greater than 461.2 ± 27.6 $U \cdot L^{-1}$) in fish after being exposed to elevated temperature regimes (Sauer and Haider 1977). The upstream (19.5 ± 2.5°C) and downstream reaches (19.7 ± 2.6°C) had significantly higher stream temperatures than the tunnel-influence reach (18.1 ± 3.0°C), and concentrations of AST were highest (i.e., above normal baseline) in trout from the upstream (717.7 ± 242.5 $U \cdot L^{-1}$) and downstream reaches (714.6 ± 332.6 $U \cdot L^{-1}$) and lowest (i.e., below normal baseline) in trout from the tunnel-influence reach (374.1 ± 118.4 $U \cdot L^{-1}$). Creatine kinase, a catalyst for the conversion of creatine, has been observed in elevated concentrations (i.e., greater than 1265.1 ± 161.7 $U \cdot L^{-1}$) in stressed fish (Shieh 1978; Shahsavani et al. 2010). Stream temperatures in the upstream (19.5 ± 2.5°C) and downstream reaches (19.7 ± 2.6°C) were near levels known to stress trout (i.e., above 20°C; Elliott 1994; Ebersole et al. 2001) and in the tunnel-influence reach they were not (18.1 ± 3.0°C). Concentrations of creatine kinase were well-above normal baseline in trout from the upstream (7132.7 ± 6467.4 $U \cdot L^{-1}$) and downstream reaches (5893.5 ± 9644.8 $U \cdot L^{-1}$) and near normal baseline in trout from the tunnel-influence reach (1306.1 ± 2018.9 $U \cdot L^{-1}$). Alanine aminotransferase, a key component of the alanine cycle, often reaches elevated concentrations (12.9 ± 1.2 $U \cdot L^{-1}$) in fish that have been exposed to elevated thermal regimes (Manera and Britti 2006). Although trout from all reaches had ALT concentrations well below normal baseline concentrations (12.9 ± 1.2 $U \cdot L^{-1}$), trout from the tunnel influence reach (4.1 ± 0.4 $U \cdot L^{-1}$) had lower concentrations than those from the upstream (8.4 ± 4.3 $U \cdot L^{-1}$) and downstream reaches (5.8 ± 1.8 $U \cdot L^{-1}$), which suggests that trout from the tunnel influence reach were less stressed than those from the upstream and downstream reaches. Stream conditions (i.e., temperature, turbidity and flow) in all reaches of upper Esopus Creek during summer were stressful at some level for trout; however, stream conditions were least stressful (i.e., most suitable) for trout in the tunnel-influence reach. Stream temperatures were coolest and within optimal levels for trout growth and survival (i.e., 12 - 19°C; Elliott 1994) within this reach, and stream flows were supplemented, not reduced (which can be stressful in summer for trout; Xu et al. 2010a), in the tunnel-influence reach.

The parasite occurrence and gill histopathology data indicated that brown trout in all study reaches of upper Esopus Creek experienced low to mild stress during summer 2010, which was contradictory to the serum chemistry and water content data. Higher-level stress responses sometimes occur if lower-level responses are manifested (Morgan and Iwama 1997); however this is not always the case (Barton et al. 2002). A study on brown bullhead *Ameriurus nebulosus* used biomarkers at multiple levels of biological organization (including histopathology and parasite prevalence) to assess the effects of impaired stream conditions caused by channelization, high residential runoff and urban outflow (Steyermark et al. 1999). Similar to the present study, they found that the occurrence of lower-level stress responses did not always result in higher-level responses. Turbidity in upper Esopus Creek may not have reached levels high enough to cause severe gill lesions, and the fine, glacial clay-composition of the turbidity may not have been abrasive to gill tissues. Redding et al. (1987) found that turbidity treatments ranging from 1 - 11 NTU and comprised of topsoil, kaolin clay and volcanic ash had no effect on the gill tissue of coho salmon and steelhead. Turbidity levels in upper Esopus Creek during summer 2010 (0.5 – 14.6 NTU) were similar to those observed by Redding et al. (1987), and topsoil, kaolin clay and volcanic ash are larger, sharper and more abrasive than glacial clay; hence, the absence of gill lesions observed by Redding et al. (1987) supports the low gill histopathology scores observed in trout from upper Esopus Creek. Low parasite occurrence in trout from all reaches can likely be attributed to natural variability in parasite occurrence. Parasites occur in all natural systems (Barber et al. 2000) and in both healthy and unhealthy organisms, but parasitic infections are typically greater in highly stressed, unhealthy or immuno-compromised fishes

(Conte 2004). The few parasites observed (four parasites total) in trout may have represented the normal burden for trout in upper Esopus Creek.

The biomarker responses observed in the present study suggest that releases from the portal have minimal, biologically meaningful effects on the health of brown trout in upper Esopus Creek. Fish stress is first detectable in primary responses (e.g., circulating cortisol concentrations), next in secondary responses (e.g., serum chemistry or histopathology) and lastly in tertiary, whole-animal responses (e.g., condition) (Barton et al. 2002). Brown trout in upper Esopus Creek experienced stress at the secondary-level and approached it at the tertiary-level in all study reaches during summer 2010. Summertime stream conditions were most favorable for trout in the tunnel-influence reach (i.e., cool temperature and supplemented streamflow), so a lower stress response from trout from in this reach was expected to be observed. The pattern observed in serum AST, creatine kinase and alanine aminotransferase suggested that trout from the tunnel-influence reach were less stressed than those in the upstream and downstream reaches; however, this response was not manifested at higher organizational levels. Therefore, releases from the Shandaken Tunnel appear to have little effect on trout populations during summertime, with some evidence of providing a refuge from stressful, unfavorable stream conditions. Sub-organismal and individual-level stress responses have been identified, and these lower-level stress responses are known to provide insight into potential population-level effects. Population-level effects, however, were not investigated in the present study, and further research is needed to assess tertiary (e.g., growth and behavior) and population-level (e.g., mortality rates) responses to determine if they correlate with the sub-organismal and individual-level fish health assessment reported herein.

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Table 1.1. Upper Esopus Creek stream temperature, turbidity and discharge for reaches upstream and downstream from the Shandaken Tunnel (mean ± SD) during June, July and August 2010. Values with different letters denote significantly different means among the reaches (Tukey-Cramer HSD test; $P < 0.05$).

Stream Reach	Temperature (°C)	Turbidity (NTU)	Discharge ($m^3 \cdot s^{-1}$)
Upstream	19.5 ± 2.5^{Z}	0.5 ± 0.9^{Z}	9.4 ± 5.1^{Z}
Tunnel-influence	18.1 ± 3.0^{Y}	14.6 ± 5.5^{Y}	57.5 ± 27.4^{Y}
Downstream	19.7 ± 2.6^{Z}	12.0 ± 5.3^{X}	92.1 ± 54.0^{X}
F, DF, P-value	5.80, 183, 0.0036	124.73, 133, < 0.0001	129.52, 275, < 0.0001

Table 1.2. Serum analyte concentrations for brown trout from the upstream, tunnel-influence and downstream reaches (mean ± SD). Values with different letters denote significantly different means among the reaches (P < 0.05), and (n) denotes sample size. Row one contains normal baseline serum concentrations for rainbow trout (derived from Walker and Fromm 1976; Hille 1982; Manera and Britti 2006).

Stream Reach	Sodium ($mEq \cdot L^{-1}$)	Potassium ($mEq \cdot L^{-1}$)	Chloride ($mEq \cdot L^{-1}$)	Bicarbonate ($mEq \cdot L^{-1}$)	Calcium ($mg \cdot dL^{-1}$)	Phosphate ($mg \cdot dL^{-1}$)	Magnesium ($mg \cdot dL^{-1}$)	Iron ($ug \cdot dL^{-}1$)
Baseline Concentration	154.07 ± 0.85	3.45 ± 0.29	128.09 ± 1.13	-----	12.52 ± 0.20	22.66 ± 1.19	3.85 ± 0.11	55.00 ± 34.00
Upstream	156.00 ± 5.6 (4)	1.42 ± 0.45 (5)^{ZY}	127.80 ± 3.03 (5)	4.88 ± 2.53 (8)	12.52 ± 1.22 (11)	15.99 ± 2.92 (11)^{Z}	3.24 ± 0.48 (9)^{Z}	62.44 ± 22.34 (9)
Tunnel-influence	151.67 ± 8.57 (6)	1.13 ± 0.61 (7)^{Z}	128.14 ± 8.71 (7)	5.50 ± 1.85 (8)	11.95 ± 0.67 (8)	12.40 ± 1.38 (8)^{Y}	2.86 ± 0.31 (8)^{ZY}	47.38 ± 12.68 (8)
Downstream	152.88 ± 6.17 (8)	3.03 ± 1.40 (8)^{Y}	130.00 ± 7.78 (8)	4.67 ± 1.22 (9)	12.12 ± 1.06 (11)	12.38 ± 2.39 (11)^{Y}	2.79 ± 0.18 (11)^{Y}	48.55 ± 22.75 (11)
Test statistic, DF, P-value	0.04, 17, 0.96	6.14, 19, 0.008	0.68, 19, 0.52	0.43, 24, 0.66	0.77, 29, 0.47	7.83, 29, 0.002	4.91, 27, 0.02	1.54, 27, 0.23

Stream Reach	Total Prot. ($g \cdot dL^{-1}$)	Albumin ($g \cdot dL^{-1}$)	Globulin ($g \cdot dL^{-1}$)	Glucose ($mg \cdot dL^{-1}$)	Total Bili. ($mg \cdot dL^{-1}$)	Alk. Phosph. ($U \cdot L^{-1}$)	Amylase ($U \cdot L^{-1}$)	Cholesterol ($mg \cdot dL^{-1}$)
Baseline Concentration	3.59 ± 0.13	1.38 ± 0.05	2.21	108.11 ± 9.98	0.04 ± 0.00	179.22 ± 19.26	-----	247.38 ± 10.32
Upstream	4.50 ± 0.60 (11)^{Z}	1.62 ± 0.36 (9)	2.71 ± 0.37 (9)^{Z}	107.00 ± 18.54 (11)	0.24 ± 0.41 (10)	61.27 ± 22.52 (11)	403.64 ± 209.59 (11)	495.00 ± 174.25 (10)
Tunnel-influence	3.71 ± 0.45 (8)^{Y}	1.58 ± 0.22 (8)	2.14 ± 0.27 (8)^{Y}	101.00 ± 22.18 (8)	0.08 ± 0.05 (8)	55.25 ± 16.69 (8)	502.25 ± 161.80 (8)	405.63 ± 161.80 (8)
Downstream	4.06 ± 0.33 (11)^{ZY}	1.63 ± 0.20 (11)	2.44 ± 0.25 (11)^{ZY}	118.18 ± 20.54 (11)	0.12 ± 0.10 (11)	54.73 ± 21.41 (11)	336.27 ± 123.07 (11)	427.18 ± 101.08 (11)
Test statistic, DF, P-value	6.58, 29, 0.005	0.11, 27, 0.89	7.94, 27, 0.002	1.80, 29, 0.19	1.85, 2, 0.40	0.33, 29, 0.72	2.41, 29, 0.11	1.02, 28, 0.37

Table 1.3. Gill histopathology scores for trout in the upstream, tunnel-influence and downstream reaches (mean ± SD). Values with different letters denote significantly different means among the reaches (Tukey-Cramer HSD test; *P* < 0.05), and (n) denotes sample size.

Stream Reach	Histopathology Score (1-5)
Upstream	1.2 ± 1.0 (11)^{Z}
Tunnel-influence	1.1 ± 0.4 (8)^{Z}
Downstream	2.3 ± 1.3 (11)^{Y}
F, DF, P-value	4.39, 29, 0.02

Table 1.4. Diet composition for trout in the upstream, tunnel-influence and downstream reaches. Values indicate the number of diet items observed per trout stomach (mean ± SD). Values with different letters denote significantly different means among the reaches (Tukey-Cramer HSD test; $P < 0.05$).

Stream Reach	Plecoptera	Trichoptera	Ephemeroptera	Coleoptera	Hemiptera
Upstream	0.6 ± 1.6	0.8 ± 1.2	0.5 ± 1.1	0.1 ± 0.2	0.7 ± 0.1^{Z}
Tunnel-influence	0.3 ± 0.4	1.0 ± 1.1	0.6 ± 0.9	0.0 ± 0.0	0.3 ± 0.2^{Y}
Downstream	0.3 ± 0.5	0.3 ± 0.6	0.8 ± 0.3	0.1 ± 0.4	0.2 ± 0.1^{Y}
F, DF, P-value	0.69, 79, 0.51	3.00, 79, 0.06	0.37, 79, 0.69	0.48, 79, 0.62	4.09, 79, 0.02

Stream Reach	Diptera	Odonata	Decapoda	Fish
Upstream	13.1 ± 35.9	0.0 ± 0.0	0.1 ± 0.3	0.35 ± 0.69
Tunnel-influence	6.2 ± 19.1	0.1 ± 0.2	0.1 ± 0.2	0.15 ± 0.49
Downstream	0.9 ± 2.7	0.0 ± 0.0	0.0 ± 0.0	0.58 ± 0.81
F, DF, P-value	1.7, 79, 0.18	1.52, 79, 0.23	1.79, 79, 0.17	2.19, 79, 0.12

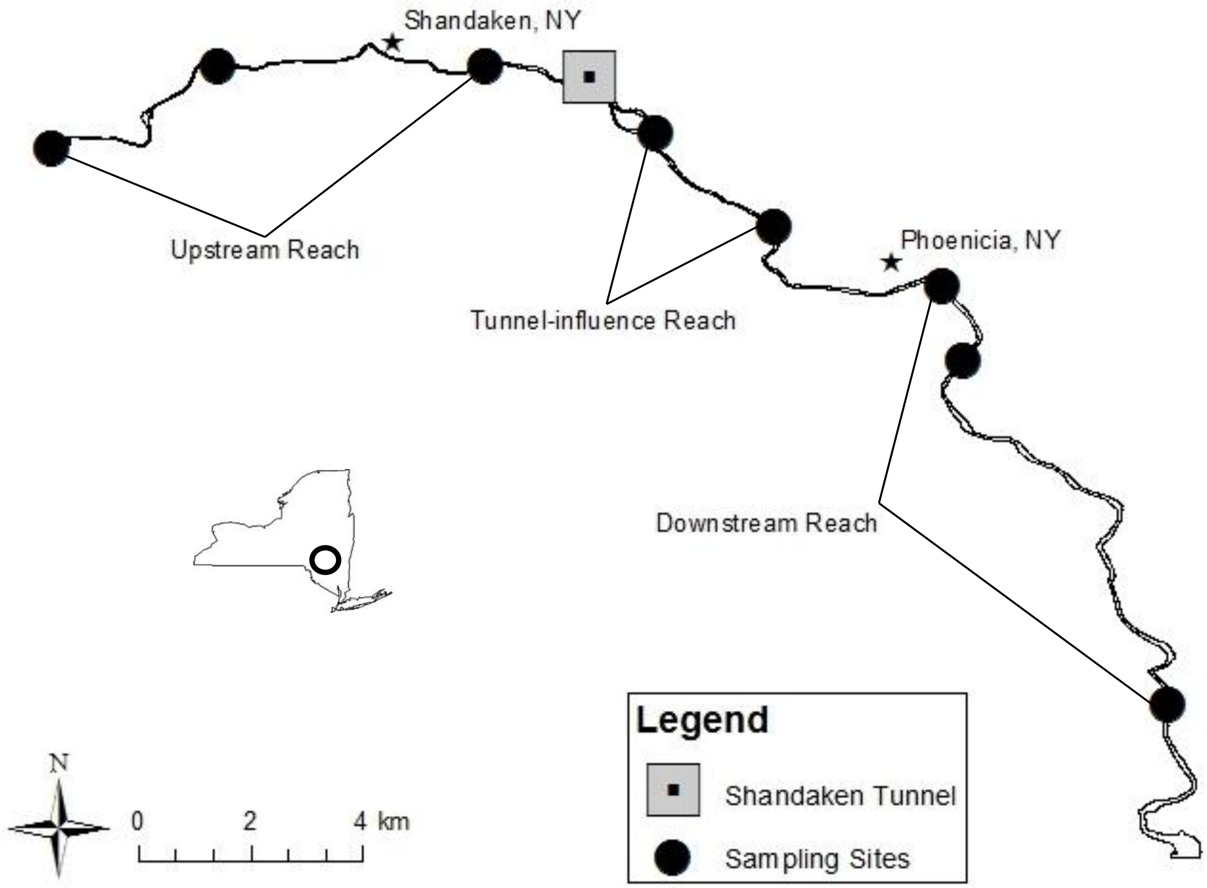

Figure 1.1. Map of upper Esopus Creek study area.

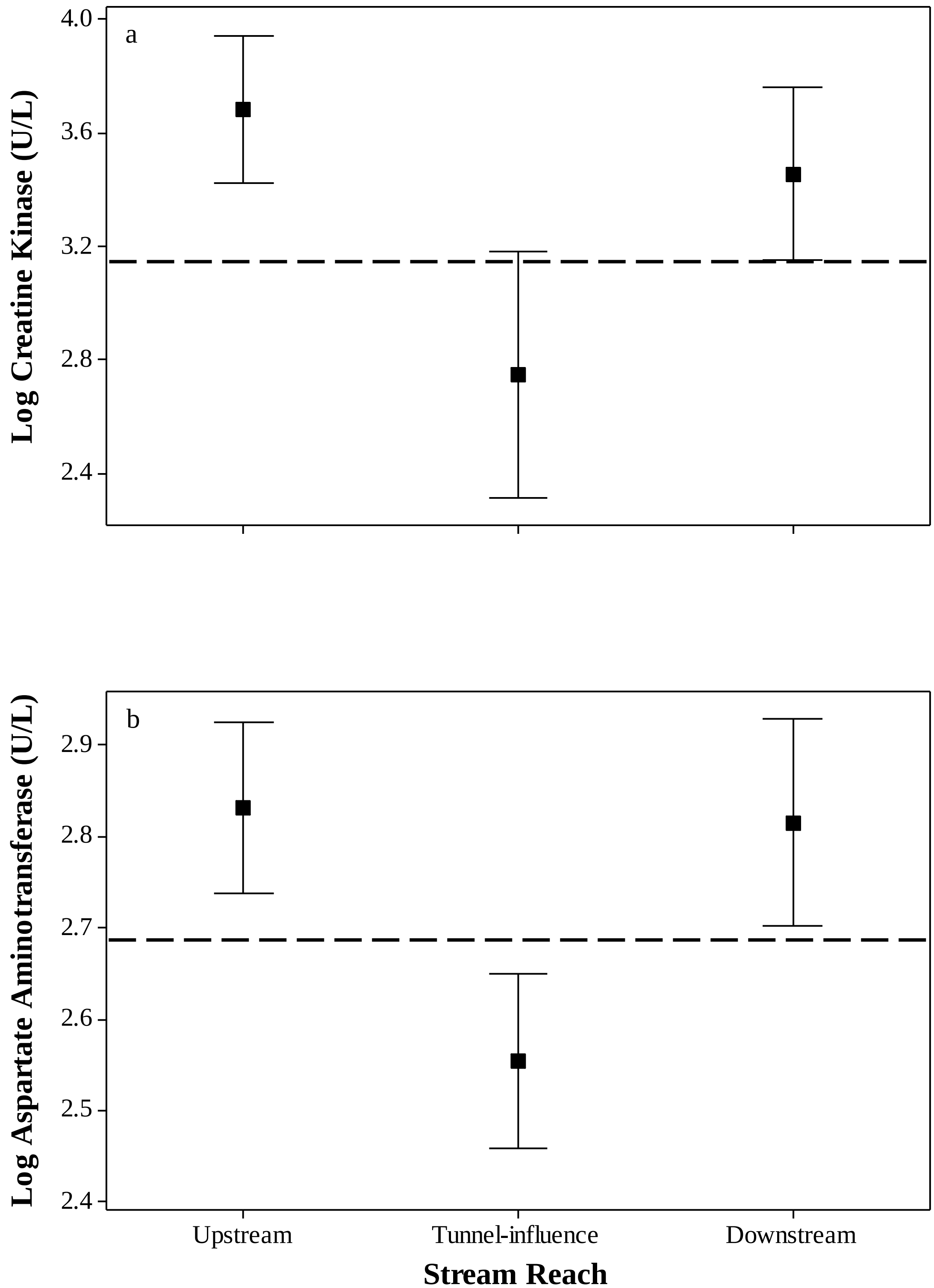

Figure 1.2a,b. Concentrations of (a) Log creatine kinase and (b) log AST in trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means, solid lines denote ± 1 standard error and dashed lines denote normal baseline concentrations.

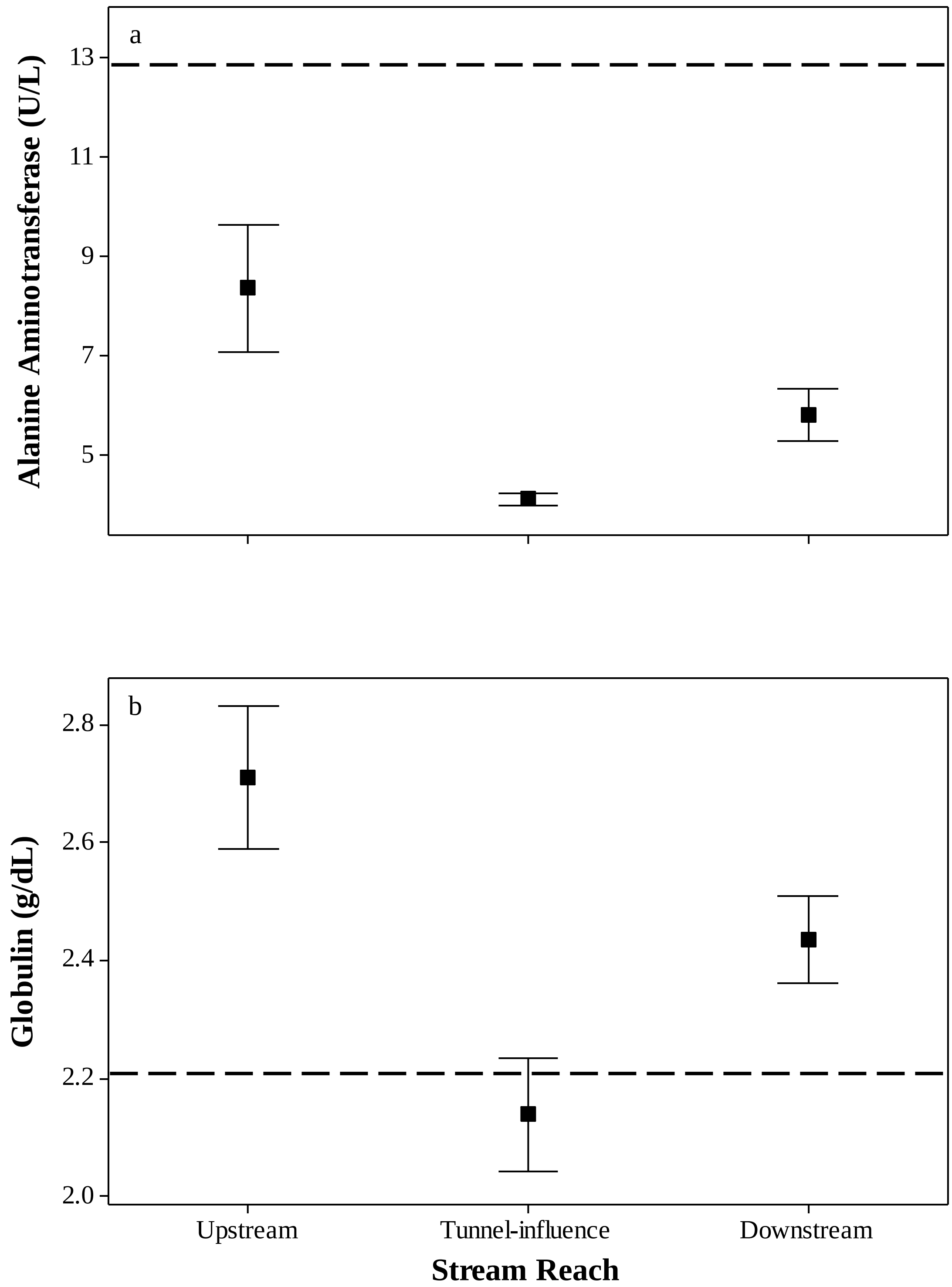

Figure 1.3a,b. Concentrations of (a) ALT and (b) globulin in trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means, solid lines denote ± 1 standard error and dashed lines denote normal baseline concentrations.

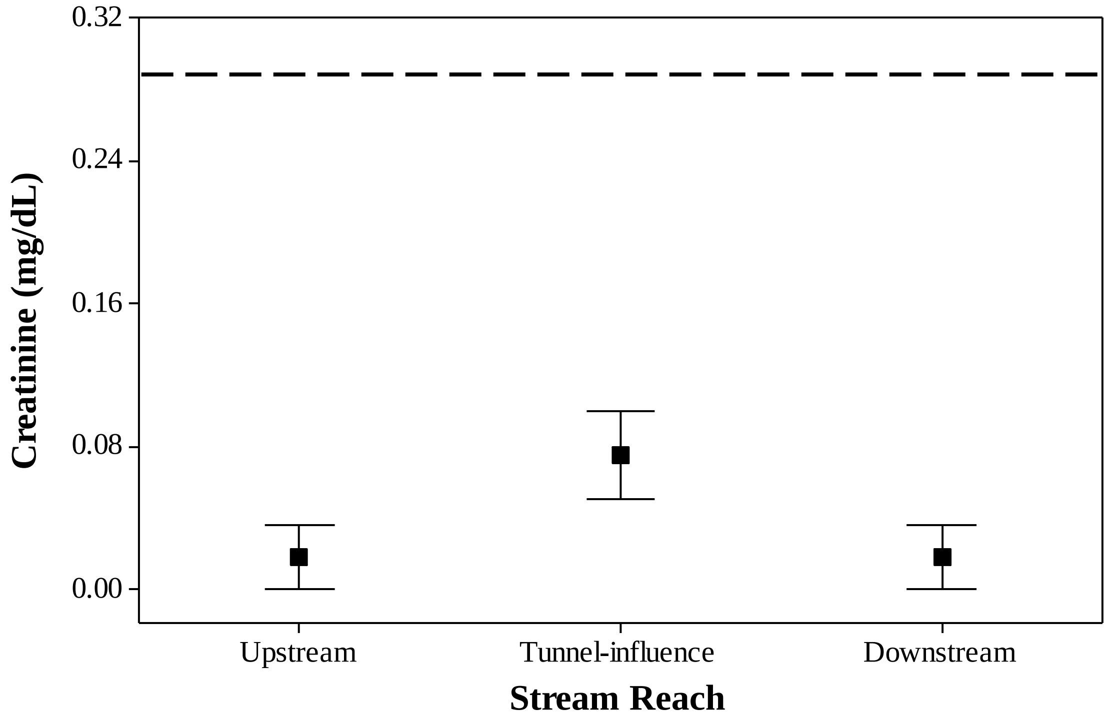

Figure 1.4. Concentrations of creatinine in trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means, solid lines denote ± 1 standard error and dashed line denotes normal baseline concentrations.

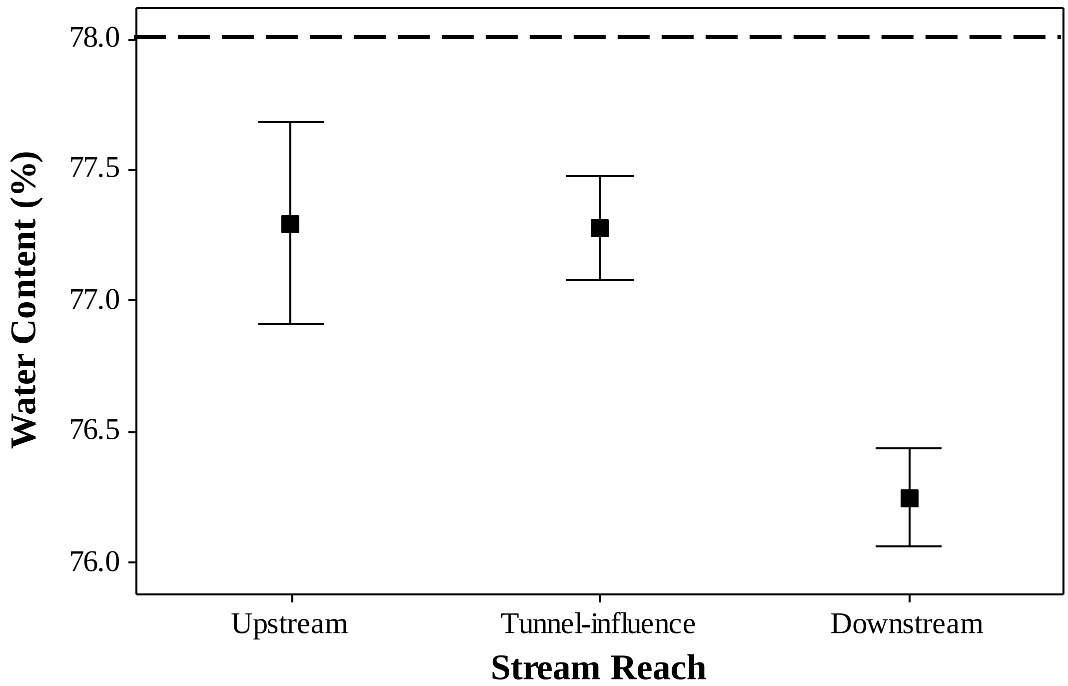

Figure 1.5. Water content of trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means, solid lines denote ± 1 standard error and dashed line denotes the 78% stress threshold.

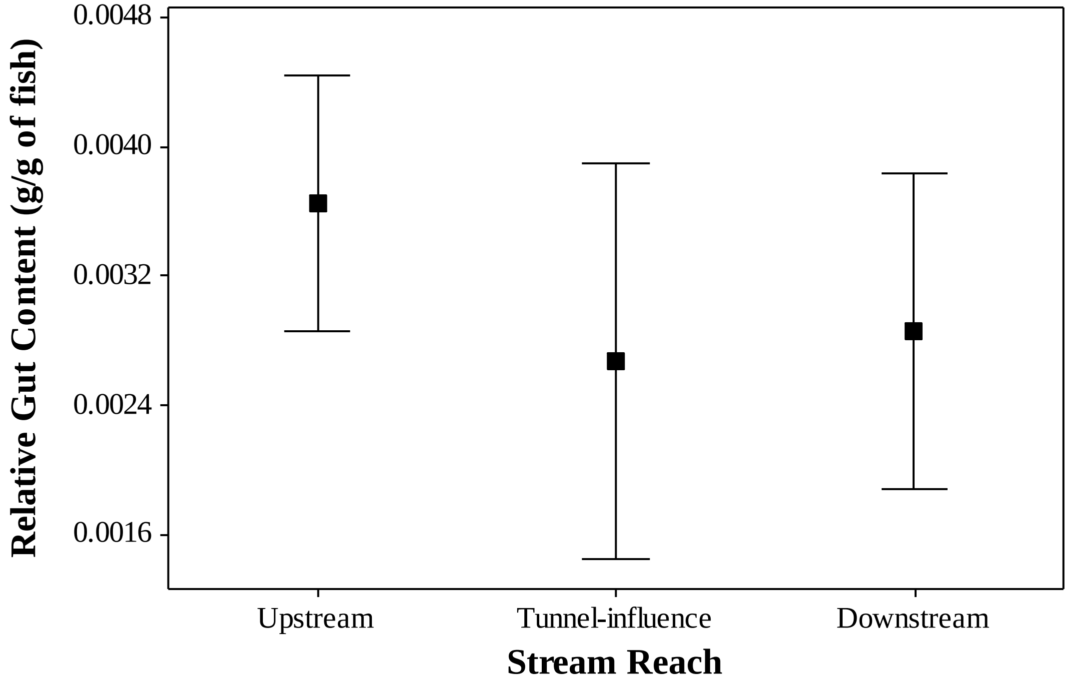

Figure 1.6. Relative gut content mass of trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means and solid lines denote ± 1 standard error.

CHAPTER 2

EFFECTS OF ALTERED STREAM FLOW, TEMPERATURE AND TURBIDITY ON BROWN TROUT HABITAT MOVEMENTS, GROWTH AND CONDITION

Abstract

Habitat, movement, growth and condition of brown trout *Salmo trutta* in response to turbid, cold-water releases from an underground aqueduct were evaluated in upper Esopus Creek, New York. Radio-telemetry, capture-recapture and intensive habitat surveys were conducted in reaches upstream, immediately downstream and far downstream from the confluence of the aqueduct during summer 2011. Stream conditions were cooler, deeper, faster-flowing and more turbid in reaches downstream from the aqueduct than they were in the reach upstream from it. The reaches downstream from the confluence of the aqueduct contained greater amounts of habitat that were optimal for adult trout compared to the reach upstream from it, and adults preferred these habitats. Conversely, the reach upstream from the aqueduct contained greater amounts of habitat that were optimal for juvenile trout compared to the reaches downstream from it, and juveniles preferred these habitats. Movement rates (38.5 - 72.2 $m \cdot day^{-1}$; range) and patterns were similar in trout among the reaches. Mean growth rates of trout were similar and negative in all reaches. Growth rates of trout immediately downstream (-0.0002 ± 0.0030 $g \cdot day^{-1} \cdot$g of $fish^{-1}$; mean ± SD) from the aqueduct were less negative than those upstream (-0.0012 ± 0.0062 $g \cdot day^{-1} \cdot$g of $fish^{-1}$) and farther downstream (-0.0029 ± 0.0047 $g \cdot day^{-1} \cdot$g of $fish^{-1}$). Water content levels of trout were higher in reaches downstream (74.1-74.8%) from the aqueduct than in the reach upstream (72.9 ± 2.5%) from it; however, water content levels of trout from all study reaches were below the 78% threshold that indicates severe physiological and nutritional stress when surpassed. Summer 2011 stream conditions in all reaches of upper

Esopus Creek were stressful to trout, and trout immediately downstream from the aqueduct's confluence appeared to be less stressed than those in other parts of the stream; these results are consistent with the findings of Ross et al. (Chapter 1). The results of the present study have implications for future management of upper Esopus Creek brown trout populations and water resources, including conserving cold water reserves available for release from the aqueduct, considering potential structural improvements of the aqueduct intake, improving brown trout stocking locations and densities and educating stakeholders of upper Esopus Creek.

Introduction

Water diversions supplement the energy, transportation, food production and water needs of humans (Kingsford 2000; Murchie et al. 2008), and they also alter the habitats and environmental conditions of the aquatic ecosystems that receive their water (Poff and Allan 1995; Bunn and Arthington 2002; Kershner et al. 2004). Conditions downstream from a diversion are often characterized by altered flow, temperature and turbidity regimes (Nilsson et al. 2005). These impaired conditions affect all stream organisms (Bunn and Arthington 2002), ranging from aquatic invertebrates (Hax and Golladay 1998) to fish (Murchie et al. 2008), and the effects are generally negative (Poff and Zimmerman 2010). Brown trout *Salmo trutta* are an economically and ecologically important sport-fish species (Elliott 1989; Van Winkle et al. 1998) whose habitat (Bunt et al. 1999), movements (Clapp et al. 1990), growth (Sweka and Hartman 2001b) and condition (Redding et al. 1987) are affected by impaired stream flow, temperature and turbidity regimes.

Brown trout and other salmonids are affected by variation in stream flow caused by human alterations to stream ecosystems. More habitat is available to trout in high flow conditions (Bunt et al. 1999), but the habitat that is available can be of low quality (Wang et al. 2001) which affects fish distribution (Jowett and Duncan 1990). Trout move more (Heggenes et al. 2007; Young et al. 2010), seek near-shore refuges (Bunt et al. 1999) and expend more energy (Fausch 1984) when exposed to high flow conditions; however, trout in these conditions can prey upon greater amounts of drifting invertebrates that provide increased opportunities for foraging and growth (Murchie and Smokorowski 2004). Trout move less (Young et al. 2010), rely more heavily on pools and cover (Bunt et al. 1999) and ultimately expend less energy (Fausch 1984) when stream flow is low. Trout stress in low-flow conditions during summer (Xu et al. 2010a) which can result in reduced prey-consumption rates (Sotiropoulos et al. 2006) and reduced growth rates (Harvey et al. 2006; Carlson et al. 2007). Ultimately, the effects of flow alterations on trout depend on their ability to respond behaviorally and find adequate refuges in order to minimize energetic and physiological costs (Valentin et al. 1996).

The effects of stream temperature on trout habitat, and the movement, growth and condition of trout are generally well defined. Optimal temperatures for brown trout growth and survival are 12 - 19˚C (Elliott 1994), with a lower thermal tolerance limit of 0.0 ˚C (Elliott 1994) and an upper thermal tolerance limit of 27.2˚C (Raleigh et al. 1986). Trout activity, foraging and growth rates are usually reduced when exposed to temperatures below the optimal range (Ojanguren 2001). Similarly, trout forage and grow less (Lee and Rinne 1980; Xu et al. 2010b), experience reduced availability of habitat and actively seek thermal refuge habitat (Kaya et al. 1977; Elliott 2000; Ebersole et al. 2001) when exposed to temperatures above the optimal range. The response of trout to temperatures above the optimal range, however, is largely dependent upon the duration of exposure (Dickerson and Vinyard 1999). Although trout streams in many regions of the U.S. are known to exceed optimal temperatures during summertime (Bunnell et al. 1998; Elliott 2000), many are characterized by diel temperature cycles that can limit the duration of exposure to elevated temperatures (Hokanson et al. 1977).

Numerous studies have concluded that elevated turbidity levels can degrade habitat and affect the movements, growth and condition of brown trout and other salmonids. Rainbow trout *Oncorhynchus mykiss* exposed to high turbidity levels (53 nephelometric turbidity units [NTU]) occupied shallower and slower habitats, actively foraged for prey (as opposed to passively waiting for drifting prey from stationary positions), experienced no net energy gain and were in poor condition; whereas, trout exposed to low turbidity levels (6 NTU) drift-foraged, experienced a positive energy gain and were in good condition (Harvey and Railsback 2009). The gill tissue and physiological condition of coho salmon *Oncorhynchus kisutch* and steelhead were not affected by low turbidity levels (1-11 NTU; Redding et al. 1987). Brook trout *Salvelinus fontinalis* were better able to detect prey items in clear water than in turbid water (43.0 NTU; Sweka and Hartman 2001a), and brook trout had significantly higher specific growth rates in clear water than in turbid water (44.8 NTU; Sweka and Hartman 2001b). Similarly, brown trout had fuller stomachs in water with lower turbidity levels (26 NTU) than in water with higher turbidity levels (141 NTU), which suggests that foraging success of trout was impaired more at high turbidity levels (Stuart-Smith et al. 2004).

A better understanding of the behavioral and physiological responses of trout to altered stream flow, turbidity and temperature is needed to provide insight into potential population-level effects of anthropogenic stream alteration. Trout adapt behaviorally to impaired stream conditions in ways that allow them to persist (Gowan and Fausch 2002; Ebersole et al. 2003; Schwartz and Herricks 2005). These behavioral adaptations range from active-search foraging strategies in turbid water (Sweka and Hartman 2001b) to increased movement rates in high-flow conditions (Young et al. 2010) to behavioral thermoregulation in water temperatures that exceed optimal levels (Ebersole et al. 2001). These behavioral adaptations, however, are often energetically and physiologically taxing and might reduce survival and fitness.

Upper Esopus Creek, in the Catskill Mountains of southeastern New York, supports viable populations of brown trout, brook trout and rainbow trout that could possibly be adversely affected by current water-management practices. Upper Esopus Creek is a key component in the water supply system for New York City. It receives supplemental flows from the nearby Schoharie Reservoir through an underground aqueduct, the Shandaken Tunnel, which joins the upper Esopus Creek at the Shandaken portal. Releases from the portal affect the discharge, turbidity and temperature of the system midway between its headwaters and the Ashokan Reservoir. Trout populations experience higher turbidity and streamflows downstream from the portal; however, water from the portal originates in the hypolimnion of Schoharie Reservoir, which provides cool water for trout during the warmer summer months. In contrast, stream conditions upstream from the portal are unaffected by releases from the aqueduct, and trout experience lower flows and turbidity and warmer water temperatures during summer in this segment of the stream. Stream angler groups, such as Trout Unlimited, have expressed concerns about the effects of the portal releases on the health of local trout populations. Stream managers and some anglers have speculated that the cool-water releases from the portal provide thermal refuge for trout populations during warmer summer months (CCES 2007a). A fish health assessment by Ross et al. (Chapter 1) revealed that brown trout populations throughout all study reaches of upper Esopus Creek were stressed and in relatively poor condition. They found that resident trout immediately downstream from the portal were slightly less stressed than those individuals residing upstream and farther downstream, based on concentrations of several serum analytes. They speculated that stream conditions in all reaches of upper Esopus Creek were stressful, but that the cooler temperatures immediately downstream from the portal might have reduced stress for the trout. Assessment of behavioral (e.g., movement and habitat selection) and physiological (e.g., growth and condition) metrics described herein will provide further insight into population-level responses of brown trout to altered stream conditions caused by releases from the Shandaken Tunnel.

Results from the present study extend the fish health findings of Ross et al. (Chapter 1), they further inform upper Esopus Creek stakeholders of the effects of Shandaken Tunnel releases on brown trout populations and they inform future fishery and water resource management decisions. The effects of altered stream flow, turbidity and temperature caused by releases from the Shandaken Tunnel on the habitat, movement, growth, and condition of brown trout were evaluated in upper Esopus Creek. The main objectives of this investigation were to determine (1) if the ranges in stress noted for individual trout, and differences in habitat upstream and downstream from the portal, could account for variations in trout behavior, growth and survival and (2) whether these differences were large enough to impact trout populations located downstream from the portal.

Methods

Study area. –Esopus Creek is located in the south central Catskill Mountain Region of southeastern New York (Figure 2.1). It begins at Winnisook Lake on Slide Mountain [1,274 m above sea level (asl)], travels 41.8 km around the base of Panther Mountain, and flows into Ashokan Reservoir (193 m asl) before draining into the Hudson River. The upper Esopus Creek watershed above Ashokan Reservoir is approximately 309 km^2, 95% forested, and entirely contained within the Catskill State Park; the remaining 5% of the watershed is comprised of commercial and residential development (CCES 2007a). Water entering upper Esopus Creek from the Shandaken Tunnel has the highest median turbidity (8.8 NTU) within the watershed; however, its contribution to the total, annual sediment load of upper Esopus Creek is considered negligible (CCES 2007a). Tributaries to upper Esopus Creek are comprised of clay-rich, glacial till channels and contribute a significant portion of the total annual sediment load during heavy rain and snow-melt events (CCES 2007a). The Shandaken Tunnel can discharge 2.5 million cubic meters of water per day into upper Esopus Creek when operated at maximum output. Discharge from the portal is regulated by a State Pollutant Discharge Elimination System permit that establishes criteria for stream flow, turbidity and temperature intended to preserve stream biota and habitat (CCES 2007a).

Habitat availability, use and selection. – Physical stream habitat available to and used by brown trout was measured in three stream reaches of upper Esopus Creek during June, July and August 2011 (Figure 2.1): an upstream reach (located 1.37 km upstream from the portal); a tunnel-influence reach (located 2.09 km downstream from the portal); and a downstream reach (located 13.23 km downstream from the portal). These data were used to evaluate habitat selection of trout and to calculate and compare the area of habitat that was optimal for juvenile (less than 200 mm) and adult brown trout (greater than 200 mm) both upstream and downstream from the portal. Stream temperature, turbidity and discharge data were continuously logged throughout summer 2011 using temperature loggers (HOBO ProV2, Onset Computer Corporation, Bourne, Massachusetts, USA), turbidity loggers (DTS-12 SDI, Forest Technology Systems Ltd., Victoria, British Columbia) and United States Geological Survey (USGS) gauging stations within each reach. Stream-channel boundaries were mapped once per month within each reach using a Trimble GeoExplorer GeoXH Global Positioning System [GPS] (Trimble Navigation Limited, Sunnyvale, California, USA). Each stream reach was 20 mean-stream-widths in length to ensure that all mesohabitat types (e.g., pool, riffle, run) were represented (Fitzpatrick et al. 1998). Available habitat was measured at five equidistant points (Fore et al. 2007) along each of 20 transects spaced one mean-stream-width apart (Simonson et al. 1994) and placed perpendicular to stream flow. Stream habitat parameters measured included: turbidity and temperature (Yellow Springs Instruments model 6920V2 meter; YSI, Yellow Springs, Ohio, USA), mean column velocity (Marsh-McBirney Flo-Mate 2000; Hach Company, Loveland, Colorado, USA), depth using a wading rod and dominant substrate using the modified

Wentworth scale (Cummins 1962). Spline interpolation in GIS (ArcGIS 10, Environmental Science Research Institute. Redlands, California, USA) was used to create raster maps (0.37-0.62 meter grid cell size; Laymosn and Reid 1986) for each measured habitat parameter during June, July and August 2011.

Habitat use of brown trout was evaluated by electrofishing in each stream reach, marking locations where trout were captured and extracting habitat data from the marked locations on the interpolated layers using ArcGIS 10. All mesohabitat types were sampled with effort equal to their proportion of occurrence within each reach. Every time a trout was captured using backpack electrofishing (Smith-Root LR-24, Smith-Root, Vancouver, Washington, USA), the fish's location within the stream was marked with a weighted float, and the trout was placed into a partially submerged wire cage along the stream bank until sampling was completed. Care was taken to ensure that trout were not being displaced while electrofishing. If a trout was displaced prior to collection, the location where it was first observed was marked. GPS coordinates of the marked trout locations were collected after an entire reach was sampled. Habitat use data were extracted from the interpolated temperature, turbidity, mean column velocity, depth and dominant substrate maps using ArcGIS 10.

Habitat use data from all reaches were combined and compared to habitat availability data from all reaches in order to evaluate habitat selection of all trout in the three study reaches. This was done for adult and juvenile trout during June, July and August using the two-sample Kolmogorov-Smirnov test (Kolmorgorov-Smirnov test statistic, D; Newcomb et al. 2007). The total area of optimal habitat was calculated by first developing habitat suitability criteria (HSC) for juvenile and adult brown trout during June, July and August. Habitat suitability criteria were developed following methods described by Baltz (1990) and Newcomb et al. (2007). In brief, optimal habitat was defined by a normalized suitability index greater than 0.4 (Freeman et al. 1997); index values less than 0.4 were defined as suboptimal. Raster calculator in ArcGIS 10 was used to map and calculate the total are of optimal habitat based on the 0.4 threshold of the HSC.

Movements.–Brown trout movement, apparent survival and thermal refuge use were quantified using radio telemetry. Thirty brown trout (354.0 ± 24.7 mm; mean total length ± SD) weighing at least 300 grams (2% fish-to-transmitter weight; Winter 1996) were obtained from the New York State Department of Environmental Conservation (NYSDEC) Catskill Fish Hatchery. Trout were surgically implanted with Advanced Telemetry Systems (ATS) F1810T radio-telemetry transmitters with internally coiled antennas (ATS, Isanti, Minnesota, USA). Ten additional trout (357.7 ± 19.1 mm) were surgically implanted with ATS F1810T dummy transmitters to serve as controls; however, the control transmitters could not be implanted using identical surgical techniques because the dummy transmitters were a different design. Surgeries were performed by Cornell Center for Animal Resources and Education veterinarians as described in Bridger and Booth (2003). Trout were placed into concrete raceways for 21 days after the surgeries to allow for full healing and to meet the 21-day withdrawal period for the anesthetic (tricaine methanesulfonate). Trout were monitored during recovery for infection, tag expulsion, abnormal behavior or swimming, and mortality. Equal numbers of trout with transmitters were randomly released into either the upstream, tunnel-influence or downstream reach. Tracking of tagged trout commenced three days following stocking (Bridger and Booth 2003) using an ATS R4500S radio-frequency receiver equipped with a Yagi antenna. Attempts were made to manually locate each trout once every day throughout the months of June, July and August. Internal fish body temperature, ambient stream temperature, and the surveyor's location, bearing angle (degrees), and distance (m) from the fish location were recorded each time a trout was located. The location of the surveyor was determined using a GPS and differentially corrected real-time to an accuracy of 30 cm using a base-station located in Kingston, New York. GPS locations (of the surveyor) were plotted in ArcGIS 10 and then corrected by bearing angle and distance to indicate true, in-stream fish locations.

Telemetry-derived spatial data on brown trout locations were used to calculate average daily movement rate ($m \cdot day^{-1}$), total movement (m moved throughout the summer), home range size (m^2; using minimum convex polygon), site fidelity (maximum number of days spent at a single site), and dispersal (m traveled from stocking site) for all tagged trout. Apparent survival (the number of days a tagged trout was alive or could be located within the stream) was determined by the loss of tagged trout due to mortality or emigration (Mitro and Zale 2002) and was quantified by a mortality signal from the radio transmitter or the complete loss of signal (Boisvert 2008). Temperature differentials were calculated by subtracting the ambient stream temperature from the internal body temperature of the fish. Use of thermal refuge habitat was defined as a temperature differential greater than or equal to 1˚C (Boisvert 2008).

Distributions of all metrics were tested for normality using the Shapiro-Wilk test (JMP, Version 7. SAS Institute Inc., Cary, North Carolina, USA) prior to statistical analyses. The following data were non-normal and transformed as indicated: home range and dispersal data were log-transformed, daily movement data were cosine-transformed, total movement data were sine-transformed and temperature differentials data were arcsine-transformed to obtain normality. Analysis of variance (ANOVA) was used to test for differences among trout in the reaches in transformed variables. Apparent survival, site fidelity and occurrence in thermal refuge habitat data were non-parametric, and the Wilcoxon Rank-Sum test was used to test for differences in trout among the study reaches. Correlation analysis was used to evaluate associations between movement rates, site fidelity, dispersal, home range size, apparent survival and occurrence in thermal refuge habitat within each reach.

Growth and condition.–Growth rates and condition of brown trout in the three study reaches were quantified and compared by capturing and recapturing trout during June, July and August 2011. Three sampling events per month were conducted in each stream reach. A six-person crew comprised of two members with backpack electrofishers and four members with dip nets sampled each reach beginning at the downstream end and proceeding upstream. Every trout that was collected was placed into a partially submerged wire cage located near the stream bank, where it remained until being processed. All trout were removed from the cage and anaesthetized, and then the total length (mm), fork length (mm) and mass (g) of trout were measured. Trout were then identified as either hatchery-reared or of wild origin based on pre-existing adipose and/or pelvic fin clippings (by the NYSDEC) and then the bioelectrical impedance analysis (BIA; following methods of Cox and Hartman 2005) and internal body temperature of trout were measured. All trout were then tagged on the dorsum with a Floy tag (FD-94B; Floy Tag and Manufacturing Incorporated; Seattle, Washington, USA) and tagged with a caudal fin clip to assess tag retention (described in Guy et al. 1996). Trout were placed into a holding cage and then released to the stream after they were fully recovered.

Water content levels of juvenile and adult brown trout were used to assess body condition. Chinook salmon *Oncorhynchus tshawytscha* were considered to be in poor condition and at risk of severe nutritional and physiological stress when their water content levels exceeded 78% (Peters et al. (2007), which suggests that 78% is a threshold that is indicative of stress for salmonids. Specific BIA models developed using the methods described by Cox and Hartman

(2005) were used to estimate water content for all brown trout. Relative growth rates of recaptured trout were calculated using the equation:

$$\text{Relative Growth Rate} = \frac{Y_2 - Y_1}{Y_1(t_2 - t_1)};$$

where *Y* denotes trout mass (g) and *t* denotes time (days). Distributions of growth rate and water content estimate data were tested for normality using a Shapiro-Wilk test. Mixed-effects ANOVA models were used to evaluate the effects of origin (hatchery or wild), life-stage (juvenile or adult), stream reach and interactions among these variables on brown trout growth rates and water content. A randomly generated, unique fish identification number was assigned to each data point and then used as a random effect in the models so that all data points could be utilized in the analysis without violating the assumption of independence among data points collected from the same trout.

Results

Habitat availability, use and selection.–The proportion of adult to juvenile trout collected throughout summer 2011 varied among study reaches. In the upstream reach, 33.7% of all trout collected were adults, and 66.3% were juveniles. Trout collections in the tunnel-influence reach were comprised of 82.7% adults, and 17.3% juveniles. In the downstream reach, 58.7% of all trout collected were adults, and 41.3% were juveniles.

Available habitat differed among the three study reaches in summer 2011. Stream conditions in the tunnel-influence reach were colder, more turbid, and faster flowing than those in upstream and downstream reaches during June, and conditions in the downstream reach were deeper and comprised of larger substrate than those in the tunnel-influence and upstream reaches in June (Table 2.1). Stream conditions in the tunnel-influence reach were more turbid and faster flowing than those in the upstream and downstream reaches and cooler than those in the upstream reach during July; conditions in the downstream reach were deeper than those in the upstream and tunnel-influence reaches and comprised of larger substrate than those in the upstream reach during July (Table 2.1). Stream conditions in the tunnel-influence reach were cooler and more turbid than those in the upstream and downstream reaches and faster flowing than those in the upstream reach during August; conditions in the downstream reach were deeper than those in the upstream and tunnel-influence reaches and comprised of larger substrate than those in the upstream reach during August (Table 2.1). Stream conditions in the tunnel-influence reach were cooler, more turbid and faster flowing than those in the upstream and downstream reaches throughout the entire summer (i.e., combining data for all months), and conditions in the downstream reach were deeper and comprised of larger substrate than those in the tunnel-influence and upstream reaches (Table 2.1).

Adult and juvenile brown trout exhibited significant selection for specific habitats throughout summer 2011 (Table 2.2). Adult trout in all study reaches preferred cool ($D = 0.247$ and $P = 0.022$), deep ($D = 0.295$ and $P = 0.004$) and fast flowing ($D = 0.240$ and $P = 0.034$) habitats in June; cool ($D = 0.227$ and $P = 0.035$) and deep ($D = 0.308$ and $P = 0.001$) habitats with large substrate ($D = 0.229$ and $P = 0.033$) in July; and deep ($D = 0.435$ and $P < 0.0001$), turbid ($D = 0.301$ and $P = 0.007$) and fast flowing ($D = 0.370$ and $P < 0.0001$) habitats with large substrate ($D = 0.265$ and $P = 0.030$) in August. Juvenile trout in all study reaches preferred less turbid ($D = 0.300$ and $P = 0.015$) habitats in June and shallow ($D = 0.301$ and $P = 0.026$) habitats in July. Juvenile trout did not exhibit significant selection for specific habitats during August.

The amount of optimal habitat for adult and juvenile brown trout in upper Esopus Creek varied by month and study reach (Table 2.3). A greater amount of optimal habitat for adult trout was present in the tunnel-influence and downstream reaches than in the upstream reach during June (Figure 2.2). A greater amount of optimal habitat for adults was present in the tunnel-influence reach than in the upstream and downstream reaches during July (Figure 2.3) and August (Figure 2.4). A greater amount of optimal habitat for juvenile trout was present in the upstream reach than in the tunnel-influence and downstream reaches during June (Figure 2.5), July (Figure 2.6) and August (Figure 2.7).

Movements.–Brown trout movements, apparent survival and use of thermal refuge habitat were similar at all study reaches (Table 2.4). Trout in all reaches exhibited dispersal after stocking, primarily in a downstream direction. During this initial dispersal, one trout moved from the upstream reach, past the portal and into the tunnel-influence reach where it remained until death from predation by common mergansers *Mergus merganser*; there were no other occurrences of trout moving past the portal. Trout settled into well-defined habitat areas within 3-5 days of being stocked, and they typically remained in these areas for the duration of the summer. Trout in the upstream reach occurred primarily in deep, pool habitats; whereas, trout in the tunnel-influence and downstream reaches occurred primarily in deep, run habitats. Although temperature differentials and occurrence in thermal refuge habitat were similar in trout at all reaches, trout in the upstream reach utilized thermal refuge habitats more often and had lower temperature differentials than trout in the tunnel-influence and downstream reaches (Table 2.4). Two trout from the upstream reach behaviorally thermoregulated when stream temperatures peaked in July 2011. These two trout moved upstream to occupy groundwater seeps when ambient stream temperatures exceeded 24°C. This behavior enabled them to maintain body temperatures 3 - 5°C lower than ambient stream temperatures, and it was not observed in any other trout during the study. Apparent survival was positively correlated with site fidelity at the upstream (R^2 = 0.68, *F* = 16.13, *DF* = 7, *P* = 0.007), tunnel-influence (R^2 = 0.90, *F* = 21.80, *DF* = 7, *P* = 0.003) and downstream reaches (R^2 = 0.75, *F* = 87.55, *DF* = 10, *P* < 0.0001). Apparent survival was negatively correlated with log dispersal at the tunnel influence reach (R^2 = 0.30, *F* = 5.21, *DF* = 10, *P* = 0.05). Although not significantly different, daily movement rates and dispersal of trout were higher at the downstream reach than at the tunnel-influence and upstream reaches, and site fidelity, apparent survival and home range size were higher at the upstream reach than at the tunnel-influence and downstream reaches (Table 2.4).

Growth and condition.–Growth rates of trout collected from the three study reaches were similar (Figure 2.8), and trout origin had a significant effect on growth rates. For all stream reaches and both life stages combined, hatchery trout growth rates were lower than those of wild trout (*F* = 5.05, *DF* = 1, *P* = 0.0283). Growth rates were not significantly related to study reach (*F* = 2.62, *DF* = 2, *P* = 0.0801), trout life-stage or factor interactions. Growth rates of trout from the tunnel-influence reach were least negative, followed by growth rates of trout from the upstream and then downstream reach (Figure 2.8).

Water content levels of trout were significantly related to stream reach (*F* = 6.72, *DF* = 2, *P* = 0.0014), life-stage (*F* = 13.59, *DF* = 1, *P* = 0.0003) and the interaction between stream reach and life-stage (*F* = 4.62, *DF* = 2, *P* = 0.0104). Water content levels of adult trout from the tunnel-influence and downstream reaches were significantly higher than those of adults from the upstream reach; they did not differ in juveniles across all study reaches (Table 2.5). The average water content levels of juvenile and adult trout were below the 78% threshold for stress at all study reaches (Table 2.5).

Discussion

The differences in stream conditions among the three study reaches of upper Esopus Creek affected habitat availability and selection by trout, as well as movement, growth and condition of hatchery and wild trout. The tunnel-influence and downstream reaches were cooler (12.0 – 17.4°C), deeper (0.5 – 0.7 m), more turbid (5.3 – 11.3 NTU) and faster-flowing (0.5 – 0.9 $m \cdot s^{-1}$) than the upstream reach during summer 2011, and adult brown trout appeared to prefer these stream conditions. The relative abundance of adult trout in the tunnel-influence (82.7%) and downstream reaches (58.7%) and the large amount of habitat that was optimal for adult trout in the tunnel-influence (59.2 ± 33.6%) and downstream reaches (38.2 ± 49.5%) supported this. Conversely, the upstream reach was warmer (14.8 – 18.0°C), shallower (0.3 m), less turbid (2.5 – 5.6 NTU) and slower-flowing (0.3 – 0.5 $m \cdot s^{-1}$) than the tunnel-influence and downstream reaches, and juvenile brown trout appeared to prefer these stream conditions. The relative abundance of juveniles in the upstream reach (66.25%) and the large amount of habitat that was optimal for juvenile trout in the upstream reach (63.8 ± 51.8%) supported this. These results are generally consistent with those of others who found that adult trout prefer deep, cool (Ayllon et al. 2010) and low-moderate flow habitats (Elliott 1994; Heggenes 1996), and that juveniles prefer shallow (Bohlin 1977; Hermansen and Krog 1984), cool (Elliott 1994) and moderate flow habitats (Heggenes and Saltveit 1990). Adult trout are not known to prefer turbid, fast-flowing habitats, as they did in the tunnel-influence reach; however, adults are less tolerant than juveniles of warm stream temperatures (Bohlin 1977), which may explain why the relative abundance of adults was higher in the tunnel-influence and downstream reaches than in the upstream reach. Juveniles are known to occupy warmer habitats to avoid larger conspecifics (Bohlin, 1977;

Elliott 2000), which may explain why the relative abundance of juvenile trout was higher in the upstream reach than in the tunnel-influence and downstream reaches.

The movement rates of brown trout in all study reaches of upper Esopus Creek were higher than those reported in the literature, which suggests that stream conditions at all reaches stressed trout and affected their behavior during summer 2011. Habitat quality affects trout behavior (e.g., movements and habitat use), and trout often exhibit innate responses to increase the chance of survival when habitat quality is impaired (Pickering 1981; Heggenes 1996). Several studies have identified temperature and turbidity thresholds that create stream conditions known to affect trout behaviors and movements once they are exceeded. Trout stress and move more in pursuit (Ebersole et al. 2001) and use of thermal refuge habitats (Kaya et al. 1977) when exposed to stream temperatures in excess of 20°C, and they move less after locating a thermal refuge habitat (Young et al. 2010). Trout activity and movement often increases when turbidity levels exceed 7.1 NTU (Gradall and Swenson 1982) and little or no effect on trout activity has been observed when turbidity levels are at or below 6 NTU (Harvey and Railsback 2009). During the present study, mean daily stream temperatures in the upstream, tunnel-influence and downstream reaches exceeded 20°C for 40%, 0% and 12% of the days, respectively, and mean daily turbidity levels exceeded 7.1 NTU for 49%, 76% and 40% of the days, respectively. Therefore, temperature and/or turbidity exceeded levels known to stress trout and affect their movements for a portion of the summer and in all study reaches, which may explain why the movement rates observed in trout from all reaches were similar. Movement rates of stream-dwelling brown trout vary widely (Young et al. 2010), but trout are relatively sedentary during summer months (Meyers et al. 1992; Burrell et al. 2000). Brown trout in an unaltered Pennsylvania stream moved an average 6 $m \cdot day^{-1}$ (Knouft and Spotila 2002). Brown trout have been found to move an average of 5-16 m·day^{-1} in streams characterized by thermally stressful summer conditions (Burrell et al. 2000; Popoff and Neumann 2005). Brown trout moved an average of 12.5 m·day^{-1} in an urbanized stream in South Dakota that was characterized by increased suspended sediment levels and degraded water quality and habitat (James et al. 2007). Therefore, the movement rates of brown trout in upper Esopus Creek during summer 2011 (38.5 – 72.2 m·day^{-1}) were higher than those reported in the literature for trout (5-16 m·day^{-1}).

Summer stream conditions in all reaches of upper Esopus Creek appeared to impair growth rates of wild and hatchery brown trout to some extent but possibly to a lesser extent for trout located in the tunnel-influence reach. Food consumption and environmental disturbances are two of the many factors known to affect the feeding and growth of fish (Jobling 1994). With the exception of wild brown trout in the tunnel-influence reach, resident trout across upper Esopus Creek actually lost mass during summer 2011. Growth rates are highly related to the consumption of food (Railsback and Rose 1999), but the composition and relative wet mass of gut contents were generally similar for trout across upper Esopus Creek during summer 2010 (Ross et al. Chapter 1), which suggests that consumption likely didn't explain the negative growth rates of trout. Increased turbidity (Sweka and Hartman 2001b), elevated summer stream temperatures (McCormick et al. 1972), exposure to increased flows (Fausch 1984) and exposure to decreased flows in summer (Xu et al. 2010a) can cause declines in the growth rates of trout. Trout in all study reaches were exposed to stream conditions known to impair growth, which may explain why most growth rates of trout were negative in upper Esopus Creek; however, it does not explain why growth rates of wild trout were positive in the tunnel-influence reach and why growth rates of trout were higher in the tunnel-influence reach than in the upstream and downstream reaches. The optimum temperature for brown trout growth is between 12-19°C

(Elliott 1994), and temperature is one of the most influential environmental factors affecting the growth of trout (Jensen 1990). Stream temperatures in the tunnel-influence reach were within the optimal range throughout the entire summer; whereas, temperatures in the upstream and downstream reaches were within the optimal range for 57% and 67% of the days in summer, respectively. This may explain the slightly higher (i.e., less negative) growth rates of trout in the tunnel-influence reach and partially explain the positive growth rates of wild trout in this reach. Wild trout are better-suited to the natural environment than hatchery trout (Vehanen et al. 2009), which enables them to adapt and respond to unfavorable stream conditions. This may further explain the positive growth rates of wild trout in the tunnel-influence reach.

Water content levels for all brown trout throughout upper Esopus Creek were below the 78% threshold for stress, which suggests that trout in all reaches were not severely stressed; this result was not expected. Fish condition typically declines in response to environmental perturbations and provides insight into the effects of chronic environmental stress on fish (Barton et al. 2002); therefore, the body condition of trout was expected to be poor in all study reaches of upper Esopus Creek in response to summer 2011 stream conditions. Trout condition is not always affected when other metrics (e.g., growth) indicate stress (Barton et al. 2002). Steyermark et al. (1999) used histopathology, fish condition, health assessment index, parasites and population age structure (all stress-indicating metrics) to evaluate the health of brown bullheads *Ameiurus nebulosus* in an industrialized, Pennsylvania stream affected by channelization, high residential runoff and urban outflow; stream conditions more heavily impaired than those of upper Esopus Creek. Similar to the findings of the present study, these authors found several metrics that suggested fish were stressed; however, fish condition and parasites did not reflect this. Growth rates of fish are generally consistent with fish condition

(Pope and Kruse 2007) (i.e., positive growth rates correlate to good condition and negative growth rates correlate to poor condition). Although growth rates of trout in upper Esopus Creek were negative, they were all very close zero, which suggests that trout in all study reaches should have been in fair to poor condition. This may explain why water contents of trout in upper Esopus Creek were below the 78% threshold that indicates stress.

Results from the present study confirm the findings from Ross et al. (Chapter 1), that indicated that brown trout in all study reaches of upper Esopus Creek were stressed, but that trout located immediately downstream from the Shandaken Tunnel were less stressed. Trout in all reaches lost mass (i.e., negative growth rates) and had higher movement rates than those reported by other studies, which indicates stress at the population-level. The tunnel-influence reach had the largest amount of habitat that was optimal for adult trout throughout the course of the summer, and trout lost less mass in this reach than in the upstream and downstream reaches, which suggests that these trout were less stressed. These findings have several implications for the management of brown trout populations in upper Esopus Creek. First, New York City residents receive water from upper Esopus Creek, so it would be useful to manage summer portal releases for drinking water to better conserve the cool, hypolimnetic reserve available in Schoharie Reservoir. The hypolimnetic volume (available for summertime diversion through the Shandaken Tunnel) is usually established when Schoharie Reservoir stratifies in late-spring, (CCES 2007b). If this cool, hypolimnetic reserve is depleted too rapidly, then warm, turbid water is discharged from the portal (U.S. Geological Survey gauge 01362230), which could eliminate the stress-reducing effects of the cold-water discharge. Second, the cold discharge from the portal appeared to create habitat conditions that adult trout preferred and that were conducive to better growth and reduced levels of stress. However, discharge from the portal also created turbid conditions downstream from its confluence that reached levels known to stress trout (i.e., greater than 7.1 NTU; Gradall and Swenson 1982). Theoretically, the turbidity that is transported through the Shandaken Tunnel and into upper Esopus Creek could be reduced by improving the intake structure at Schoharie Reservoir (CCES 2007b) and could possibly further decrease stress experienced by trout in the tunnel-influence reach. Anglers and other recreational groups have recommended that the New York City Department of Environmental Protection (NYCDEP) construct a multi-level intake at Schoharie Reservoir in order to transfer the highest quality water to upper Esopus Creek; such an option is being evaluated by the NYCDEP (CCES 2007b).

The results of this effort need to be qualified because the study was limited to an analysis of the portal effects (on brown trout) only during summer 2011. Stream ecosystems are dynamic, and stream habitat conditions vary among seasons and years. Additional research is needed to better define how the releases from the Shandaken Tunnel might affect (a) trout during other seasons and years, (b) the reproductive success (e.g., egg survival) and health of other life-stages of brown trout and (c) other fish species and prey resources that support brown trout populations.

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Table 2.1. Habitat availability in the upstream, tunnel-influence and downstream reaches during June, July and August (mean ± SD). Values with different letters denote significantly different means among the reaches (*P* < 0.05). Comparisons were not made among months.

Stream Reach	Temperature (°C)	Turbidity (NTU)	Velocity (m·s^{-1})	Depth (m)	Substrate (mm)
			June		
Upstream	14.8 ± 0.1^{Z}	2.6 ± 1.0^{Z}	0.5 ± 0.3^{Z}	0.3 ± 0.2^{Z}	115.9 ± 80.8^{Z}
Tunnel-influence	12.0 ± 0.2^{Y}	11.3 ± 1.29^{Y}	0.9 ± 0.5^{Y}	0.5 ± 0.2^{Y}	100.4 ± 79.5^{Z}
Downstream	14.0 ± 0.2^{X}	6.5 ± 0.74^{X}	0.7 ± 0.4^{X}	0.7 ± 0.3^{X}	148.6 ± 96.9^{Y}
F-statistic, DF, P-value	5547.51, 299, < 0.0001	1403.36, 299, < 0.0001	25.29, 299, < 0.0001	68.54, 299, < 0.0001	8.03, 299, 0.0004
			July		
Upstream	18.0 ± 0.3^{Z}	5.6 ± 0.6^{Z}	0.4 ± 0.2^{Z}	0.3 ± 0.2^{Z}	89.5 ± 71.0^{Z}
Tunnel-influence	16.6 ± 0.3^{Y}	8.0 ± 0.9^{Y}	0.7 ± 0.4^{Y}	0.5 ± 0.2^{Y}	130.9 ± 91.6^{Y}
Downstream	16.6 ± 0.3^{Y}	5.3 ± 0.5^{X}	0.5 ± 0.5^{X}	0.7 ± 0.3^{X}	146.3 ± 95.4^{Y}
F-statistic, DF, P-value	907.09, 299, < 0.0001	450.88, 299, < 0.0001	22.87, 299, < 0.0001	79.79, 299, < 0.0001	11.90, 299, < 0.0001
			August		
Upstream	17.3 ± 0.1^{Z}	4.8 ± 0.3^{Z}	0.3 ± 0.2^{Z}	0.3 ± 0.2^{Z}	125.7 ± 84.9^{Z}
Tunnel-influence	17.0 ± 0.3^{Y}	9.5 ± 0.7^{Y}	0.6 ± 0.4^{Y}	0.5 ± 0.2^{Y}	138.4 ± 126.6^{ZY}
Downstream	17.4 ± 0.1^{X}	8.4 ± 0.5^{X}	0.5 ± 0.3^{Y}	0.6 ± 0.2^{X}	173.1 ± 95.8^{Y}
F-statistic, DF, P-value	109.89, 299, < 0.0001	2560.04, 299, < 0.0001	30.74, 299, < 0.0001	75.20, 299, < 0.0001	5.72, 299, 0.0036
			All months		
Upstream	16.7 ± 1.4^{Z}	4.6 ± 1.1^{Z}	0.4 ± 0.3^{Z}	0.3 ± 0.2^{Z}	110.4 ± 80.4^{Z}
Tunnel-influence	15.2 ± 2.3^{Y}	9.6 ± 1.7^{Y}	0.7 ± 0.4^{Y}	0.5 ± 0.2^{Y}	123.3 ± 102.2^{Z}
Downstream	16.0 ± 1.5^{X}	6.7 ± 1.4^{X}	0.6 ± 0.4^{Y}	0.6 ± 0.3^{X}	156.0 ± 96.5^{Y}
F-statistic, DF, P-value	54.88, 899, < 0.0001	945.63, 899, < 0.0001	70.36, 899, < 0.0001	220.40, 899, < 0.0001	19.45, 899, < 0.0001

Table 2.2. Habitat availability and use of adult and juvenile brown trout throughout upper Esopus Creek during June, July and August 2011 (mean ± SD). ^{Z} indicates significant habitat selection (as determined by comparing monthly habitat availability to habitat use for adult and juvenile fish) (*P* < 0.05).

Month	Temperature (°C)	Turbidity (NTU)	Velocity ($m \cdot s^{-1}$)	Depth (m)	Substrate (mm)
			Available		
June	13.7 ± 1.2	7.0 ± 3.3	0.7 ± 0.4	0.5 ± 0.3	121.9 ± 88.2
July	17.1 ± 0.7	6.3 ± 1.4	0.5 ± 0.4	0.5 ± 0.3	121.5 ± 89.5
August	17.2 ± 0.3	7.5 ± 2.1	0.4 ± 0.3	0.4 ± 0.3	145.5 ± 104.9
			Adult		
June	13.2 ± 1.3^{Z}	8.1 ± 2.9	0.8 ± 0.3^{Z}	0.6 ± 0.2^{Z}	116.2 ± 79.1
July	16.8 ± 0.5	6.2 ± 1.4	0.5 ± 0.3	0.6 ± 0.3^{Z}	132.6 ± 75.3^{Z}
August	17.2 ± 0.2	8.7 ± 1.3^{Z}	0.6 ± 0.2^{Z}	0.6 ± 0.1^{Z}	153.3 ± 76.7^{Z}
			Juvenile		
June	14.1 ± 0.9	6.1 ± 2.7^{Z}	0.6 ± 0.3	0.5 ± 0.2	112.9 ± 60.5
July	17.4 ± 0.7	6.0 ± 1.0	0.5 ± 0.2	0.4 ± 0.2^{Z}	129.3 ± 72.7
August	17.3 ± 0.2	7.4 ± 2.2	0.4 ± 0.3	0.5 ± 0.2	146.0 ± 82.9

Table 2.3. Adult and juvenile brown trout optimal habitat in the upstream, tunnel-influence and downstream reaches during June, July and August. Percentages were calculated by: optimal area/total reach area.

Stream Reach	Optimal Habitat					
	June		July		August	
	(m^2)	(%)	(m^2)	(%)	(m^2)	(%)
			Adult			
Upstream	1707.1	29.5	8.5	0.2	664.1	11.9
Tunnel-influence	8427.5	89.7	2154.4	23.2	5928.8	64.8
Downstream	14295.0	95.2	912.2	5.9	1998.6	13.4
			Juvenile			
Upstream	258.2	4.5	5083.5	87.0	5596.6	99.9
Tunnel-influence	54.5	0.6	2268.7	24.4	4373.7	47.8
Downstream	124.3	0.8	1981.6	12.9	6867.9	46.2

Table 2.4. Movement rates, apparent survival and thermal refuge use of trout from the upstream, tunnel-influence and downstream reaches (mean ± SD). Occurrence in thermal refuge habitat is a proportion calculated from: days observed in thermal refuge habitat/total days observed.

Stream Reach	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Home Range (m^2)	Site Fidelity (days)	Dispersal (m)	Apparent Survival (days)	Occurrence in Thermal Refuge Habitat	Temperature Differentials (°C)
Upstream	38.5 ± 49.7	1391.1 ± 2255.2	12359.2 ± 21387.9	39.4 ± 21.8	1898.9 ± 1469.1	53.9 ± 18.6	0.12 ± 0.10	-0.43 ± 0.16
Tunnel-influence	41.4 ± 104.6	693.4 ± 1000.8	1559.5 ± 2891.9	34.4 ± 18.7	2356.4 ± 4034.2	45.1 ± 17.7	0.09 ± 0.08	-0.34 ± 0.19
Downstream	72.2 ± 161.9	501.8 ± 501.6	8903.3 ± 18065.4	18.4 ± 15.9	2685.7 ± 5530.4	32.4 ± 17.4	0.07 ± 0.09	-0.02 ± 0.65
F, DF, P-value	0.35, 26, 0.71	0.42, 26, 0.66	0.88, 24, 0.43	2.75, 26, 0.08	0.44, 26, 0.65	2.93, 26, 0.07	0.48, 26, 0.62	1.15, 25, 0.33

Table 2.5. Water content of brown trout from the upstream, tunnel-influence and downstream reaches (mean ± SD). Values with different letters denote significantly different means among the reaches ($P < 0.05$).

	Water content (%)	
Stream Reach	Adult	Juvenile
Upstream	72.9 ± 2.5^{Z}	73.0 ± 1.9
Tunnel-influence	74.1 ± 3.0^{Y}	72.6 ± 1.2
Downstream	74.8 ± 3.0^{Y}	73.3 ± 1.3

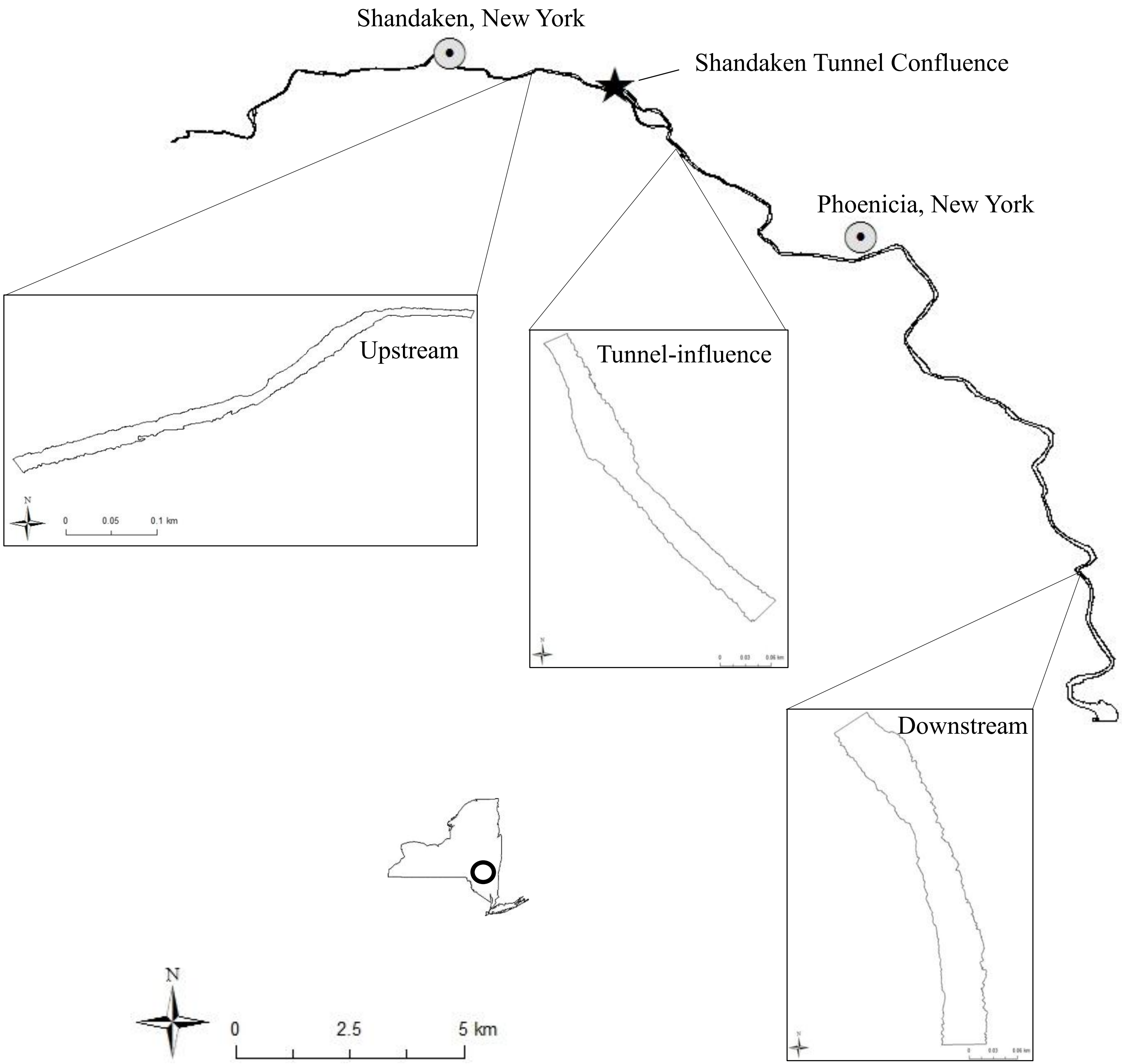

Figure 2.1. Map of upper Esopus Creek study area.

Figure 2.2. Map of optimal and suboptimal adult brown trout habitat in the upstream, tunnel-influence and downstream reaches during June 2011.

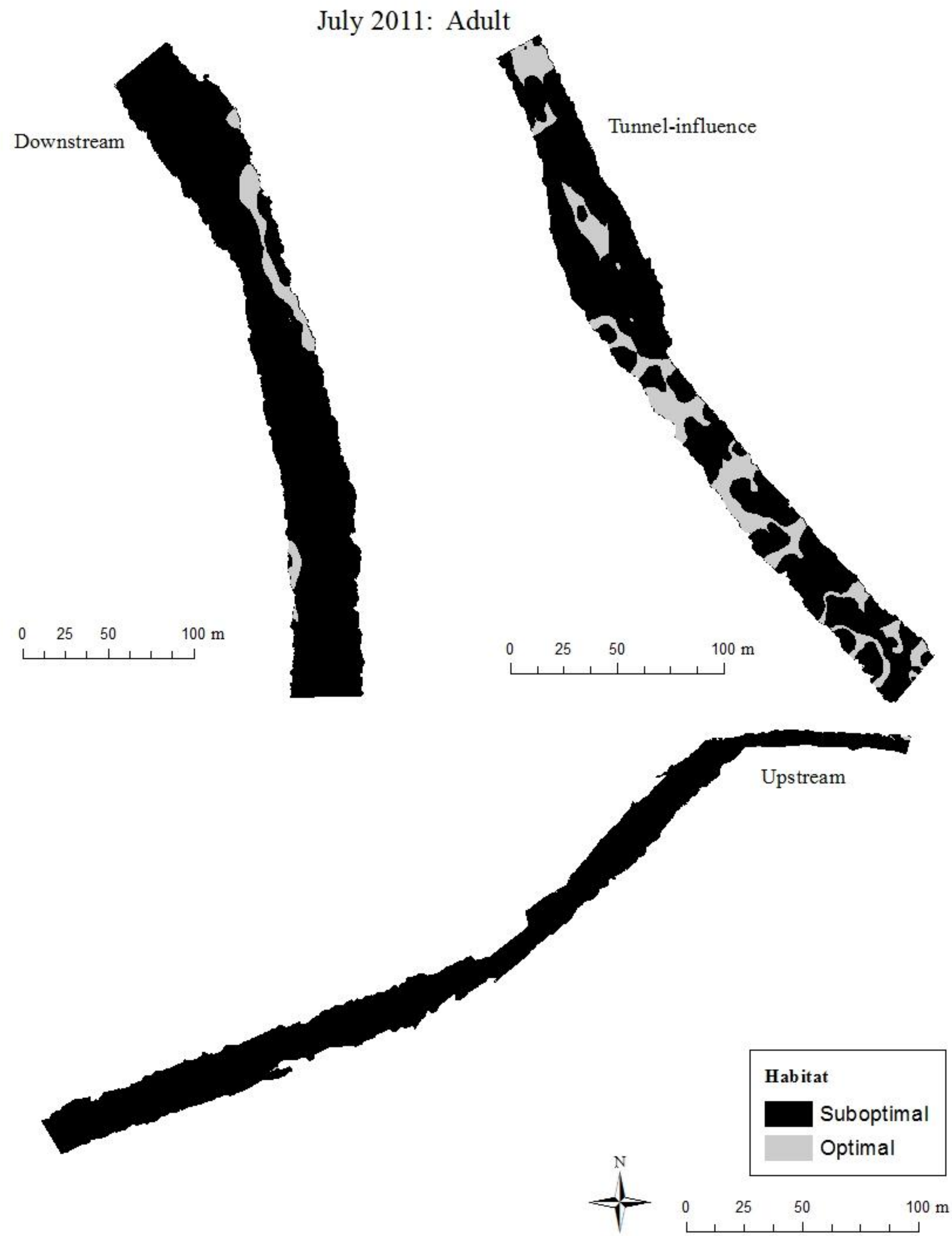

Figure 2.3. Map of optimal and suboptimal adult brown trout habitat in the upstream, tunnel-influence and downstream reaches during July 2011

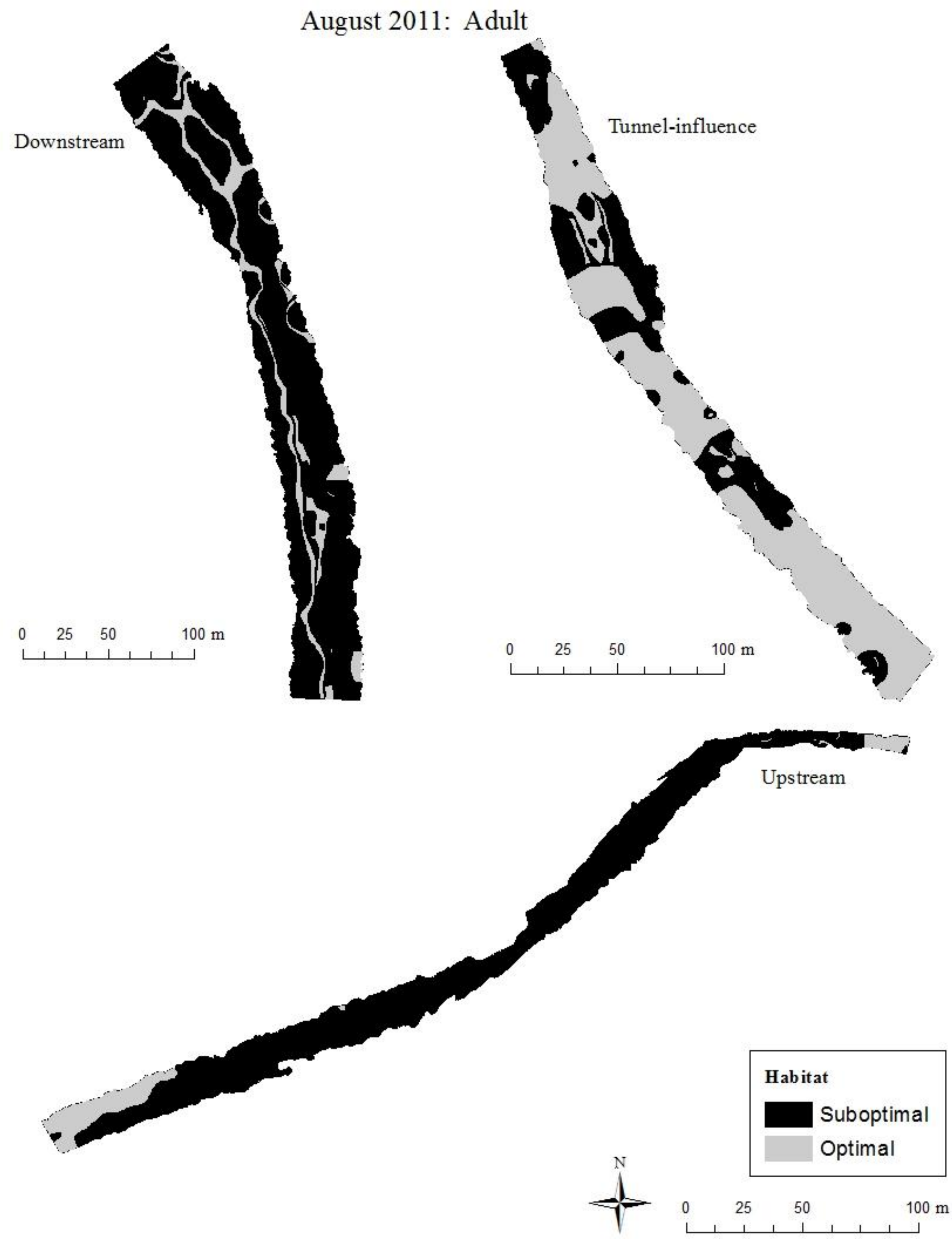

Figure 2.4. Map of optimal and suboptimal adult brown trout habitat in the upstream, tunnel-influence and downstream reaches during August 201

Figure 2.5. Map of optimal and suboptimal juvenile brown trout habitat in the upstream, tunnel-influence and downstream reaches during June 2011.

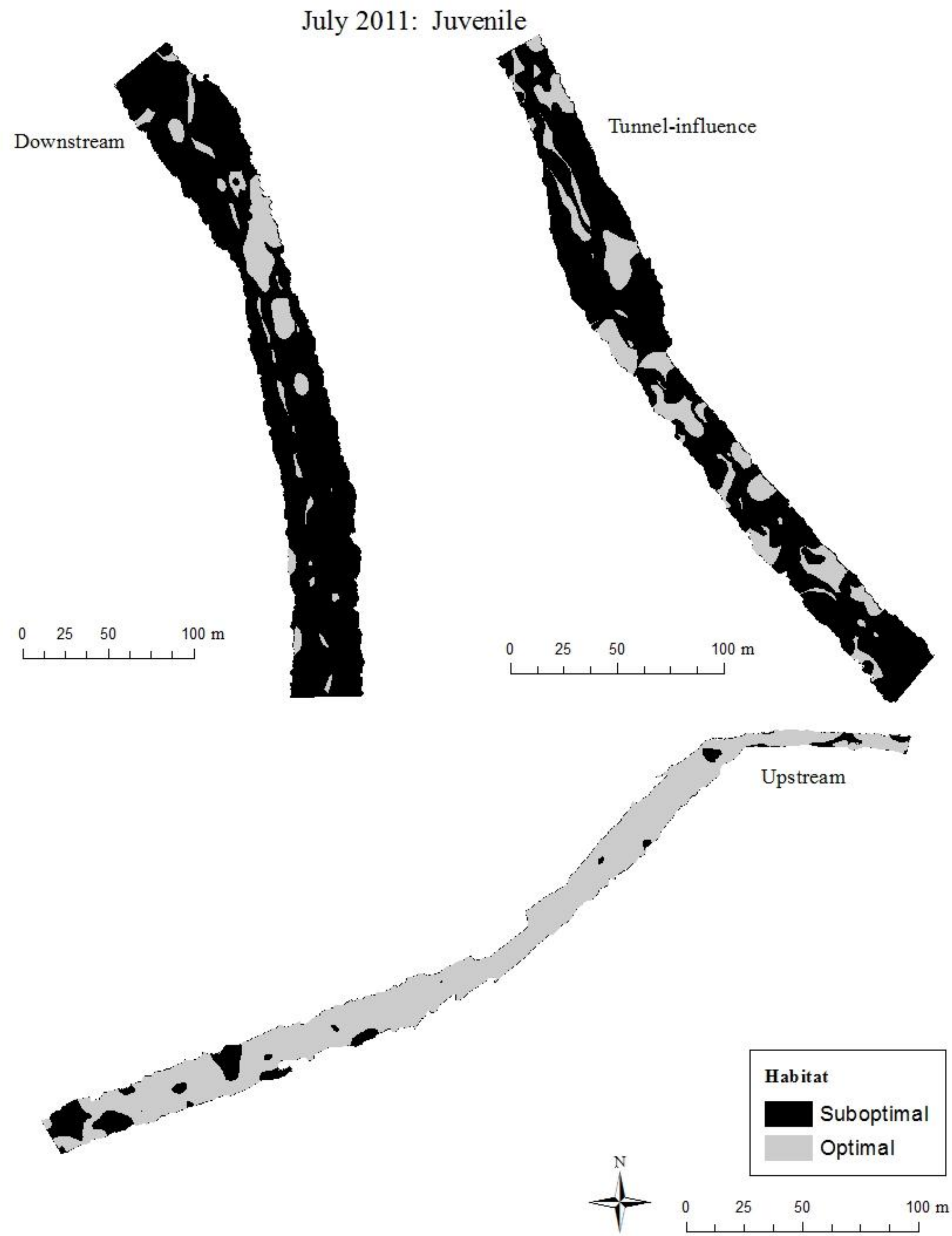

Figure 2.6. Map of optimal and suboptimal juvenile brown trout habitat in the upstream, tunnel-influence and downstream reaches during July 2011.

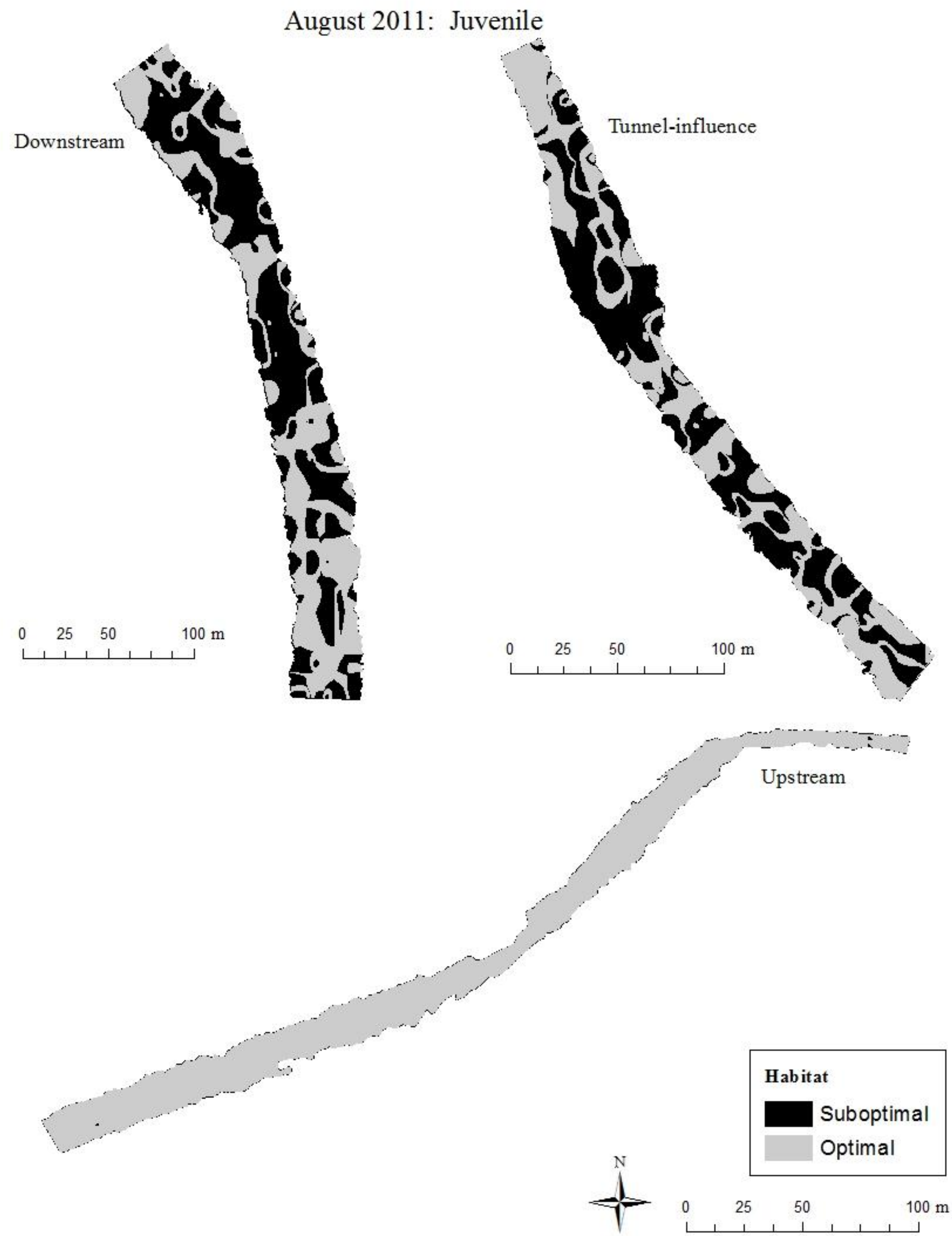

Figure 2.7. Map of optimal and suboptimal juvenile brown trout habitat in the upstream, tunnel-influence and downstream reaches during August 2011.

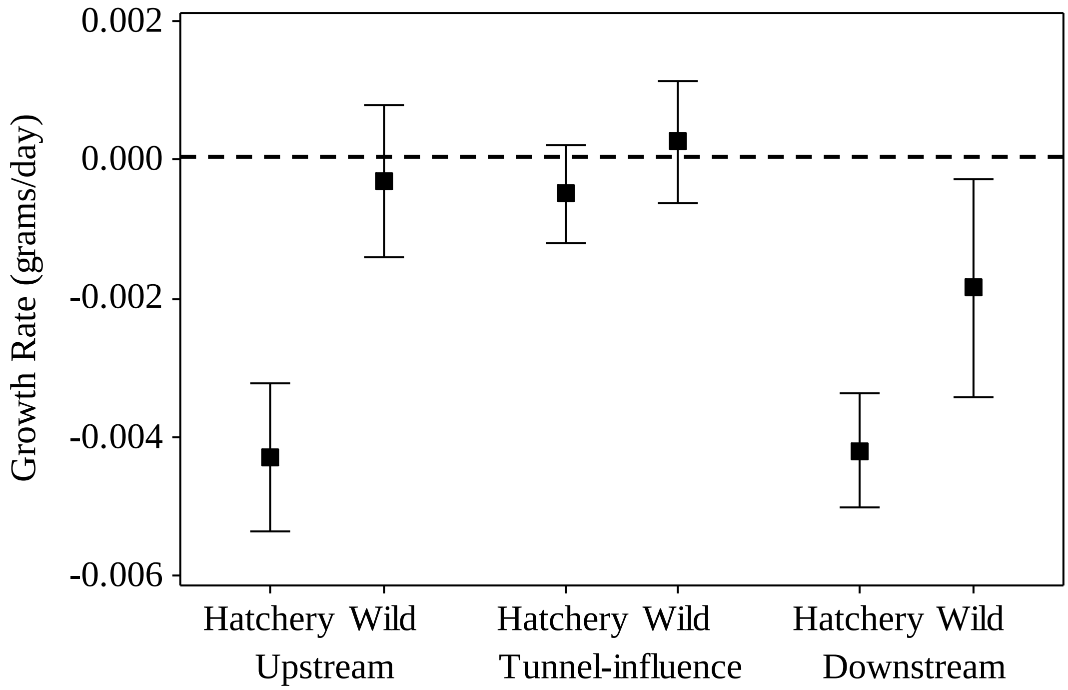

Figure 2.8. Growth rates (g·day^{-1}) of brown trout from the upstream, tunnel-influence and downstream reaches. Black squares denote means, solid lines denote ±1 standard error, and dashed line denotes no (i.e., zero) growth.

CONCLUSION

The effects of altered stream flow, temperature and turbidity on the habitat, movement and physiology of brown trout populations in upper Esopus Creek, New York were evaluated during summer 2010 and 2011. Differences in habitat conditions between reaches upstream and downstream from the Shandaken Tunnel evoked secondary and tertiary-level stress responses in trout throughout the stream during both years. Trout throughout upper Esopus Creek were stressed at the secondary-level (i.e., serum chemistry) and showed some indication of stress at the tertiary-level (i.e., condition) during summer 2010. Trout immediately downstream from the portal appeared less stressed than those upstream or farther downstream. Similarly, trout throughout the stream were stressed at the tertiary-level (i.e., growth and movement rates), with evidence of less stress immediately downstream from the portal during summer 2011. Collectively, these findings suggest that summer stream conditions throughout upper Esopus Creek were stressful for trout to some extent, which is common in many streams throughout North America where human activities have impaired natural stream conditions and degraded stream habitat (Roth et al. 1996; Allan 2004). This effect, however, was lessened immediately downstream from the portal. Stream conditions within each reach likely provided different costs and benefits for trout residing in them, which may explain why trout in all study reaches showed signs of stress based on findings from the present study. The reach upstream from the portal was characterized by low flows and warm temperatures during summer 2011, which are stressful to trout (Elliott 2000; Xu et al. 2010a,b); however, it was also characterized by turbidity levels below those known to stress trout. The reach immediately downstream from the portal was characterized by turbidity levels known to stress trout (Gradall and Swenson 1982) and high flows; however, they were also cold, which is not stressful to trout in summer. The reach farther downstream from the portal was characterized by high flows and water temperatures and turbidity levels between those in the reaches upstream and immediately downstream; hence, they were also stressful for trout to some extent. Stream temperature is a primary environmental factor affecting the growth and performance of all fish, (Jensen 1990); therefore, the cold water discharge from the portal likely explains why trout were less stressed in the tunnel-influence reach.

Results from the present study have management and ecological implications at the watershed-scale. First, discharge from the portal appeared to create habitat conditions that adult trout preferred and that were conducive to better growth and lower stress levels; this was likely due to the cold waters of the portal discharge. However, discharge from the portal also created turbid conditions that reached levels known to stress trout (i.e., greater than 7.1 NTU; Gradall and Swenson 1982) during both years. The turbidity transported through the Shandaken Tunnel and into upper Esopus Creek could be reduced by improving the intake structure at Schoharie Reservoir (CCES 2007b), and this could possibly further reduce stress experienced by trout immediately downstream from the portal. Anglers and other recreational groups have recommended that the NYCDEP construct a multi-level intake at Schoharie Reservoir in order to transfer the highest quality water to upper Esopus Creek; such an option is being considered by the NYCDEP as part of its Catskill Turbidity Control Study (CCES 2007b). Second, New York City residents receive water from upper Esopus Creek, so it would be useful to manage summer portal releases for drinking water to better conserve the cool, hypolimnetic reserve available in Schoharie Reservoir. The hypolimnetic volume (available for summertime diversion through the Shandaken Tunnel) is usually established when Schoharie Reservoir stratifies in late-spring, (CCES 2007b). If this cool, hypolimnetic reserve is depleted too rapidly, then warm, turbid water is discharged from the portal (U.S. Geological Survey gauge 01362230), which could eliminate the stress-reducing effects of the cold-water discharge. Third, the NYSDEC annually stocks catchable-size brown trout in upper Esopus Creek to increase angler success and satisfaction (CCES 2007b). Stocked trout lost less mass immediately downstream from the portal than in other portions of the stream, which suggests that stream conditions were most favorable for them in this reach. Increasing stocking densities within this reach and decreasing them in other parts of the stream could provide hatchery trout with greater potential for growth and survival, which could provide anglers with increased opportunities for successful harvest. Additionally, wild and stocked trout compete for resources (Weiss and Schmutz 1999); hence, although this type of stocking scheme may increase competition between wild and hatchery trout immediately downstream from the portal, it could also reduce competitive interactions between wild and hatchery trout in portions of the stream receiving lower stocking densities. Lastly, the stream conditions created by releases from the portal have been a contentious topic for several decades, and previous studies have not addressed potential effects on brown trout populations (CCES 2007a). Results from the present study address some of the ecological uncertainties posed in CCES upper Esopus Creek management plans (2007a,b) and should be used to educate and inform the public, special interest groups and management agencies in the watershed in order to facilitate agreeable management solutions and actions.

Dams and diversions are present in streams and rivers worldwide, and they affect fish habitats and populations in most systems they occur in (Nilsson et al. 2005). Anthropogenic stream alteration generally has deleterious effects on fish habitats and resources; however, the present study and others (e.g., McKinney et al. 2001; Connor et al. 2003) have indicated that human alterations to streams and rivers can be utilized in ways to benefit fisheries resources (e.g., tailwater trout fisheries; Krause et al. 2005) by considering the biology and habitat needs of target species. Lastly, results from the present study highlight the importance of evaluating stress responses of fish at multiple biological levels. Stress was detected at the secondary and tertiary-levels, which provided a more comprehensive understanding of the extent to which impaired stream conditions affected brown trout populations in upper Esopus Creek. Fisheries researchers and managers are often interested in population-level responses; thus, evaluating stress at multiple biological levels provides a better understanding of population-level effects of anthropogenic stream alteration on fish (Barton et al. 2002).

The response of brown trout to altered flow, temperature and turbidity regimes were evaluated at multiple levels of biological organization; however, potential effects on brown trout reproduction (e.g., spawning), recruitment and mortality rates were not investigated, and further research is needed in these areas. In addition, the present study was conducted only during summer for two years, which limited the temporal scale of the results. Additional research is needed during other years and seasons to better understand how releases from the portal might affect stream habitat and brown trout during these times.

APPENDIX A

APPARENT SURVIVAL, MOVEMENT AND THERMAL REFUGE USE OF BROWN TROUT IN UPPER ESOPUS CREEK, NEW YORK USING RADIO-TELEMETRY

Introduction and Methods

Apparent survival, movement and thermal refuge use of brown trout *Salmo trutta* were compared in reaches upstream and downstream from the Shandaken Tunnel in upper Esopus Creek, New York using radio telemetry in summers 2009 and 2010. The objective of the present study was to evaluate how cold, turbid releases from the Shandaken Tunnel were affecting resident trout populations.

Trout weighing at least 300 grams (2% fish-to-transmitter weight; Winter 1996) were surgically implanted with Advanced Telemetry Systems (ATS) F1810T whip-antenna, radio-telemetry transmitters (ATS, Isanti, Minnesota, USA) during summers 2009 and 2010. Forty-three trout were tagged in 2009 (356.2 ± 56.9 mm [mean total length ± SD]) and 47 in 2010 (360.0 ± 28.4 mm). Ten additional trout (349.9 ± 49.2 mm) were tagged in 2010 with ATS F1810T dummy transmitters to serve as controls. Both wild (collected using backpack electrofishing) and hatchery trout were tagged and then immediately released into upper Esopus Creek (i.e., no recovery period) in June 2009. Hatchery trout were tagged and held in four, 15 kiloliter holding tanks at the Pine Hill Waste Water Treatment Plant in Pine Hill, New York for seven days before being released into the stream in June and July 2010. Supply water in the holding tanks was from nearby Birch Creek. Trout were monitored during recovery for infection, tag expulsion, abnormal behavior or swimming, and mortality. Equal numbers of tagged trout were randomly released into reaches upstream and downstream from the portal during both years.

Tracking of tagged trout commenced three days following stocking (Bridger and Booth 2003) using an ATS R4500S radio-frequency receiver equipped with a Yagi antenna. Methods for tracking tagged trout differed between summers 2009 and 2010. Attempts were made to locate each trout three to five days per week during summer 2009, and attempts were made to locate each trout once every day in 2010. The internal fish body temperature, ambient stream temperature, and the surveyor's location (using a global positioning system) were recorded each time a trout was located in 2009; data on the precise, in-stream location of trout were not collected. Global positioning system locations (of the surveyor) were plotted in GIS (ArcGIS 10, Environmental Science Research Institute. Redlands, California, USA), and then buffered by 0.8 km to reflect the maximum transmitter detection distance (T.J. Ross, unpublished) since coordinates did not indicate true trout locations in 2009. Data were collected so that precise, in-stream trout locations could be determined in summer 2010. The internal fish body temperature, ambient stream temperature, and the surveyor's location, bearing angle (degrees), and distance (m) from the fish location were recorded. Global positioning system locations were plotted in GIS, corrected by bearing angle and distance (to indicate true trout location), and then buffered by 2.5 meters to reflect surveyor detection error (T.J. Ross, unpublished) in 2010.

Telemetry-derived spatial data on brown trout locations were used to calculate daily movement rate ($m \cdot day^{-1}$) and total movement (m moved throughout study) for all tagged trout. Apparent survival (the number of days a tagged trout was alive or could located within the stream) was determined by the loss of tagged trout due to mortality or emigration (Mitro and Zale 2002) and was quantified by a mortality signal from the radio transmitter or the complete loss of signal (Boisvert 2008). Temperature differentials were calculated by subtracting the ambient stream temperature from the internal body temperature of the fish. Use of thermal refuge habitat was defined as temperature differentials greater than or equal to 1˚C (Boisvert 2008).

Distributions of all metrics were tested for normality using the Shapiro-Wilk test (JMP, Version 7. SAS Institute Inc., Cary, North Carolina, USA). Daily movement, total movement, apparent survival and thermal refuge use data were non-normal and log-transformed to obtain normality in 2009. Daily movement and total movement data were non-normal and log-transformed in 2010. T-tests were used to test for differences in daily movement, total movement, apparent survival and thermal refuge use data between trout located in reaches upstream and downstream from the portal.

Results and Conclusions

Stream habitat conditions differed in reaches upstream and downstream from the Shandaken Tunnel during summers 2009 and 2010. Conditions downstream from the portal were cooler and higher flow than those upstream during summer 2009 (Table A.1). Similarly, conditions downstream from the portal were cooler, more turbid and higher flow than those upstream from the portal during summer 2010 (Table A.1).

Apparent survival, movements and thermal refuge use of brown trout were similar in reaches upstream and downstream from the portal during summer 2009 (Table A.2). Apparent survival of all trout was low, with many trout surviving less than one month after being tagged and released (Table A.2). Trout typically settled into defined habitat areas shortly after being tagged and released, and they remained in these areas throughout the summer. A few trout exhibited large-scale movements, most of which were in a downstream direction. Two trout were released upstream from the portal, moved downstream past the portal, and then remained downstream from the portal until death or tag loss. No other tagged trout moved past the portal during summer 2009. All but two trout maintained average body temperatures that were lower than ambient stream temperatures; however, only four trout behaviorally thermoregulated (i.e., body temperatures at least 1°C lower than ambient stream temperatures) (Table A.2).

Post-surgery recovery of all tagged trout was poor and survival was low during summer 2010, (4.1 ± 7.9 days; [mean ± SD]), which resulted in only ten trout being used for analyses (instead of 47). Apparent survival, daily movement and total movement were similar in trout located upstream and downstream from the portal (Table A.3); average apparent survival of trout was only slightly over two weeks (Table A.3). Trout settled into relatively defined habitat areas shortly after being stocked, and they typically remained in these areas for the duration of the summer, similar to 2009. Only one trout exhibited large-scale movement, and it was in a downstream direction; however; there were no trout that moved past the portal. Trout located downstream from the portal utilized thermal refuge habitat significantly more than trout located upstream from it (Table 3); however, there were no trout that behaviorally thermoregulated.

Stream habitat conditions differed upstream and downstream from the Shandaken Tunnel during summers 2009 and 2010; therefore, differences in apparent survival, movement and thermal refuge use of trout were expected to be observed. These differences, however, were not observed. A minimum of 24 hours of recovery is needed for fish to resume normal behavior (Bridger and Booth 2003); other studies suggested at least two weeks of recovery were needed (Paukert et al. 2001). Trout were not held for recovery during summer 2009, which may explain why apparent survival of tagged trout was low and why movement and thermal refuge use were similar in trout located in reaches upstream and downstream from the portal. Global positioning system coordinates were collected at a coarse-scale at trout locations in summer 2009, and this may also explain why movements of trout were similar in reaches upstream and downstream from the portal. Errors in telemetry locations can occur when attempting to determine the precise, in-stream location of a tagged trout (Roger and White 2007). Global positioning system coordinates were collected at streamside, surveyor locations (not at actual trout locations), and additional data (i.e., bearing angle and distance from trout) were not collected to allow for post-survey correction during summer 2009. This affected the accuracy of true trout locations. The more frequently fish are observed (i.e., multiple times in a 24-hour period) the better the estimates are of movement and behavior (Roger and White 2007). Trout were tracked 3-5 days per week during summer 2009, instead of daily, which may have affected the accuracy of the apparent survival, movement and thermal refuge use data.

Surgeries in 2010 were performed during June and July, when water temperatures were above those known to stress trout (i.e., 20°C; Ebersole et al. 2001); hence, the poor recovery and low survival of trout after surgery can likely be attributed to warm water temperatures in the holding tanks and in the stream. Most fish, especially salmonids, exhibit increased mortality and greater risk of infection when surgically implanted with telemetry transmitters and then released into warm water (Knights and Lasee 1996; Bunnell and Isely 1999; Walsh et al. 2000). In addition, fish at risk of mortality and infection may not behave normally, which affects movement, habitat use and behavior (Roger and White 2007). Therefore, movement and thermal refuge use of tagged trout in 2010 may have been affected by the poor recovery and low survival after surgery in response to warm water temperatures.

Table A.1. Temperature, turbidity and discharge upstream and downstream from the Shandaken Tunnel (mean ± SD) during summers 2009 and 2010. Values with different letters are significantly different (*P* < 0.05). Comparisons were not made among years.

Stream Reach	Temperature (°C)	Turbidity (NTU)	Discharge ($m^3 \cdot s^{-1}$)
		2009	
Upstream	16.2 ± 1.9^Z	-----	45.3 ± 61.3^Z
Downstream	14.6 ± 1.2^Y	-----	198.7 ± 193.7^Y
T-ratio, DF, P-value	6.03, 108.57, < 0.0001		-6.26, 81.84, < 0.0001
		2010	
Upstream	19.5 ± 2.5^Z	0.5 ± 0.9^Z	9.4 ± 5.1^Z
Downstream	18.1 ± 3.0^Y	14.6 ± 5.5^Y	92.1 ± 54.0^X
T-ratio, DF, P-value	-2.35, 86.77, 0.02	14.29, 46.41, < 0.0001	14.64, 92.59, < 0.0001

Table A.2. Average apparent survival, daily movement, total movement and thermal refuge use of brown trout located upstream and downstream from the Shandaken Tunnel during summer 2009 (mean ± SD).

Stream Reach	Apparent Survival (days)	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Temperature Difference (°C; stream-fish)
Upstream	18.3 ± 14.0	84.2 ± 104.4	1192.9 ± 1119.9	0.6 ± 0.4
Downstream	18.0 ± 20.5	130.4 ± 305.3	1596.2 ± 2788.9	0.6 ± 0.5
T-ratio, DF, P-value	0.27, 30.78, 0.39	-0.06, 30.88, 0.52	-0.07, 30.89, 0.53	0.52, 24.30, 0.30

Table A.3. Average apparent survival, daily movement, total movement and thermal refuge use of brown trout located upstream and downstream from the Shandaken Tunnel during summer 2010 (mean ± SD). Values with different letters are significantly different (*P* < 0.05).

Stream Reach	Apparent Survival (days)	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Temperature Difference (°C; stream-fish)
Upstream	17.0 ± 14.4	15.3 ± 22.1	61.3 ± 30.7	-0.2 ± 0.2[Z]
Downstream	14.6 ± 10.7	89.5 ± 154.4	448.8 ± 595.2	0.5 ± 0.3[Y]
T-ratio, DF, P-value	0.26, 3.00, 0.41	-1.22, 4.25, 0.86	-1.72, 7.91, 0.94	4.32, 4.39, 0.01

APPENDIX B

TABLE OF STATISTICS FOR BROWN TROUT IMPLANTED WITH RADIO-TELEMETRY TRANSMITTERS DURING SUMMERS 2009-2011 IN UPPER ESOPUS CREEK, NEW YORK

Table of statistics summarizing data from brown trout implanted with radio-telemetry transmitters during summers 2009-2011 in upper Esopus Creek, New York. The table contains the following data for each trout used in the study: study year; radio-frequency of implanted transmitter; stocking site; species; hatchery or wild (origin); total length; weight; the number of days alive or detectable within the stream (apparent survival); the average distance moved in one day (daily movement); the average distance moved throughout the study (total movement); the average difference between ambient stream temperature and internal fish body temperature (temperature difference).

Appendix B. Table of statistics for brown trout implanted with radio-telemetry transmitters during summer 2009 in upper Esopus Creek, New York. Blank spaces indicate no data, and (a) indicates that a transmitter was implanted in a second fish.

Year	Radio Frequency (Mhz)	Stock Site	Species	Origin	Length (mm)	Weight (g)	Apparent Survival (days)	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Temperature Difference (°C)
2009	150.212	Upstream		Wild	326	358	9	65.5	589.2	0.8
2009	150.212(a)	Upstream	Brown	Wild	437	959	33	55.9	1845.5	0.5
2009	150.233	Upstream	Brown	Hatchery	338	415	18	48.8	878.5	-0.1
2009	150.252	Upstream		Wild	344	421	11	319.9	3519.1	0.5
2009	150.252(a)	Upstream	Brown	Wild	297	297	37	11.6	417.4	0.7
2009	150.272	Upstream		Wild	434	1025	14	143.9	2014.4	0.6
2009	150.272(a)	Upstream	Rainbow	Wild	427	905	3	141.0	423.0	0.4
2009	150.292	Upstream		Wild	385	717	16	60.3	965.3	0.4
2009	150.292(a)	Upstream	Brown	Wild	410	745	8	20.4	163.5	0.7
2009	150.311	Upstream	Brown	Hatchery	335	453	3	7.8	15.7	0.5
2009	150.311(a)	Upstream	Brown	Wild	431	918	6	10.0	60.3	0.4
2009	150.333	Downstream	Brown	Hatchery	353	574	20	142.9	2857.7	0.4
2009	150.352	Downstream	Brown	Hatchery	352	481	23	153.2	3524.1	0.1
2009	150.352(a)	Upstream	Brown	Wild	300	290	3	152.9	458.8	-0.7
2009	150.372	Downstream	Brown	Hatchery	417	959	26	15.7	409.0	0.5
2009	150.393	Upstream	Brown	Wild	286	275	3	385.4	1156.2	0.3
2009	150.412	Upstream	Brown	Wild	179	241	3	102.3	307.0	0.4
2009	150.431	Downstream	Brown	Hatchery	315	379	12	88.3	1060.0	0.4
2009	150.452	Downstream	Brown	Hatchery	359	509	43	72.2	3105.7	0.9
2009	150.473	Downstream	Brown	Hatchery	371	589	93	18.0	1671.4	1.1
2009	150.492	Downstream	Brown	Hatchery	348	474	26	68.7	1784.9	0.4
2009	150.492(a)	Downstream	Brown	Wild	374	499	3	374.0	1122.1	0.7
2009	150.513	Downstream	Brown	Wild	296	332	9	56.6	509.0	-0.5
2009	150.531	Downstream		Wild	328	337	9	52.9	476.2	0.8
2009	150.552	Upstream	Brown	Hatchery	359	601	4	346.8	1387.0	0.5
2009	150.572	Downstream	Brown	Hatchery	331	416	7	16.1	112.9	0.5
2009	150.572(a)	Downstream	Brown	Wild	472	1090	9	1302.5	11722.1	1.8
2009	150.591	Downstream	Brown	Hatchery	371	557	15	15.0	225.3	0.5
2009	150.591(a)	Downstream	Brown	Wild	295	283	1	1042.3	1042.3	0.8
2009	150.612	Downstream		Wild	307	302	10	118.3	1183.3	1.3
2009	150.632	Downstream	Brown	Hatchery	385	705	5	31.5	157.4	0.4
2009	150.652	Downstream		Wild	356	476	11	30.1	330.7	0.1
2009	150.652(a)	Downstream		Wild	512	1395	18	19.7	353.6	0.9
2009	150.672	Upstream	Brown	Hatchery	327	385	7	28.9	202.3	0.6
2009	150.691	Upstream	Brown	Hatchery	346	501	1	11.1	11.1	.05
2009	150.691(a)	Downstream	Brown	Wild	358	469	3	21.6	64.9	0.4
2009	150.712	Downstream	Brown	Hatchery	314	341	6	54.5	326.8	0.3
2009	150.731	Upstream	Brown	Hatchery	351	568	48	55.6	2667.6	0.3
2009	150.753	Upstream	Brown	Hatchery	362	557	7	33.2	431.6	0.8
2009	150.753(a)	Downstream		Wild	334	413	13	21.6	151.3	1.7
2009	150.771	Upstream	Brown	Hatchery	320	401	16	49.3	788.3	0.6
2009	150.771(a)	Downstream	Rainbow	Wild	418	902	11	218.4	2402.1	0.9
2009	150.791	Upstream	Brown	Hatchery	356	504	9	36.9	332.5	0.3

Appendix B continued. Table of statistics for brown trout implanted with radio-telemetry transmitters during summer 2010 in upper Esopus Creek, New York. (a) indicates that a transmitter was implanted in a second fish.

Year	Radio Frequency (Mhz)	Stock Site	Species	Origin	Length (mm)	Weight (g)	Apparent Survival (days)	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Temperature Difference (°C)
2010	150.022	Upstream	Brown	Hatchery	393	752	0	0.0	0.0	0.0
2010	150.042	Downstream	Brown	Hatchery	367	649	6	430.7	1292.1	0.5
2010	150.042(a)	Downstream	Brown	Hatchery	382	830	0	0.0	0.0	0.0
2010	150.061	Upstream	Brown	Hatchery	368	608	0	0.0	0.0	0.0
2010	150.061(a)	Downstream	Brown	Hatchery	403	708	2	153.4	457.2	0.7
2010	150.082	Upstream	Brown	Hatchery	292	314	1	0.0	0.0	0.0
2010	150.082(a)	Downstream	Brown	Hatchery	386	898	2	208.7	626.1	0.5
2010	150.101	Downstream	Brown	Hatchery	375	680	22	70.1	1332.4	0.9
2010	150.122	Downstream	Brown	Hatchery	356	546	25	12.4	272.1	0.4
2010	150.142	Downstream	Brown	Hatchery	364	460	0	0.0	0.0	0.0
2010	150.142(a)	Downstream	Brown	Hatchery	335	979	8	7.4	36.9	0.5
2010	150.162	Downstream	Brown	Hatchery	353	594	7	12.0	48.1	0.8
2010	150.183	Downstream	Brown	Hatchery	383	744	1	0.0	0.0	0.0
2010	150.201	Downstream	Brown	Hatchery	378	714	2	8.6	17.1	1.0
2010	150.222	Downstream	Brown	Hatchery	353	570	1	0.0	0.0	0.0
2010	150.222(a)	Downstream	Brown	Hatchery	273	226	0	0.0	0.0	0.0
2010	150.242	Downstream	Brown	Hatchery	324	423	2	26.3	78.9	-2.1
2010	150.242(a)	Downstream	Brown	Hatchery	387	878	30	2.5	68.5	0.7
2010	150.261	Downstream	Brown	Hatchery	372	669	0	0.0	0.0	0.0
2010	150.282	Downstream	Brown	Hatchery	393	724	2	395.8	791.5	0.6
2010	150.282(a)	Downstream	Brown	Hatchery	390	898	0	0.0	0.0	0.0
2010	150.302	Downstream	Brown	Hatchery	325	438	3	75.8	227.4	0.5
2010	150.322	Downstream	Brown	Hatchery	366	606	2	47.6	95.3	-0.9
2010	150.342	Downstream	Brown	Hatchery	337	464	4	91.6	91.6	0.1
2010	150.342(a)	Upstream	Brown	Hatchery	334	424	1	0.0	0.0	0.0
2010	150.363	Downstream	Brown	Hatchery	345	457	2	193.7	581.0	-0.1
2010	150.363(a)	Upstream	Brown	Hatchery	363	518	0	0.0	0.0	0.0
2010	150.382	Upstream	Brown	Hatchery	376	698	5	40.8	81.6	-0.4
2010	150.382(a)	Upstream	Brown	Hatchery	371	632	0	0.0	0.0	0.0
2010	150.402	Upstream	Brown	Hatchery	385	658	1	0.0	0.0	0.0
2010	150.402(a)	Upstream	Brown	Hatchery	353	568	13	2.6	26.0	-0.2
2010	150.422	Upstream	Brown	Hatchery	359	548	3	6.2	24.6	0.2
2010	150.422(a)	Upstream	Brown	Hatchery	402	750	0	0.0	0.0	0.0
2010	150.441	Upstream	Brown	Hatchery	381	722	1	0.0	0.0	0.0
2010	150.463	Upstream	Brown	Hatchery	287	319	0	0.0	0.0	0.0
2010	150.463(a)	Upstream	Brown	Hatchery	362	618	0	0.0	0.0	0.0
2010	150.481	Upstream	Brown	Hatchery	362	597	2	40.9	122.6	-0.0
2010	150.502	Upstream	Brown	Hatchery	378	668	1	0.0	0.0	0.0
2010	150.502(a)	Upstream	Brown	Hatchery	353	548	0	0.0	0.0	0.0
2010	150.522	Upstream	Brown	Hatchery	366	596	0	0.0	0.0	0.0
2010	150.522(a)	Upstream	Brown	Hatchery	379	670	33	2.6	76.4	0.0
2010	150.543	Upstream	Brown	Hatchery	332	444	1	0.0	0.0	0.0
2010	150.543(a)	Upstream	Brown	Hatchery	342	504	1	0.0	0.0	0.0
2010	150.562	Upstream	Brown	Hatchery	370	613	3	59.6	178.8	0.3
2010	150.562(a)	Upstream	Brown	Hatchery	368	548	0	0.0	0.0	0.0
2010	150.582	Upstream	Brown	Hatchery	336	461	0	0.0	0.0	0.0
2010	150.601	Upstream	Brown	Hatchery	332	504	0	0.0	0.0	0.0

Appendix B continued. Table of statistics for brown trout implanted with radio-telemetry transmitters during summer 2011 in upper Esopus Creek, New York.

Year	Radio Frequency (Mhz)	Stock Site	Species	Origin	Length (mm)	Weight (g)	Apparent Survival (days)	Daily Movement ($m \cdot day^{-1}$)	Total Movement (m)	Temperature Difference (°C)
2011	150.032	Upstream	Brown	Hatchery	335	440	66	4.2	219.5	0.4
2011	150.050	Upstream	Brown	Hatchery	260	575	41	6.2	211.3	0.4
2011	150.072	Upstream	Brown	Hatchery	360	545	50	5.4	182.8	0.7
2011	150.091	Upstream	Brown	Hatchery	347	512	16	61.4	552.9	0.3
2011	150.111	Upstream	Brown	Hatchery	334	414	66	3.8	227.3	0.3
2011	150.130	Upstream	Brown	Hatchery	341	420	66	4.6	274.7	0.4
2011	150.151	Upstream	Brown	Hatchery	349	451	66	4.9	290.8	0.3
2011	150.172	Upstream	Brown	Hatchery	352	524	66	125.2	6512.0	0.6
2011	150.192	Upstream	Brown	Hatchery	349	503	1	0.0	0.0	0.0
2011	150.211	Upstream	Brown	Hatchery	357	566	44	97.9	2840.0	0.7
2011	150.230	Tunnel-influence	Brown	Hatchery	367	547	27	8.3	173.5	0.0
2011	150.251	Tunnel-influence	Brown	Hatchery	360	496	16	356.2	3561.5	0.3
2011	150.271	Tunnel-influence	Brown	Hatchery	365	676	66	6.3	380.2	0.2
2011	150.291	Tunnel-influence	Brown	Hatchery	334	484	66	11.2	579.8	0.5
2011	150.311	Tunnel-influence	Brown	Hatchery	383	684	40	27.3	929.2	0.4
2011	150.332	Tunnel-influence	Brown	Hatchery	355	590	65	3.6	213.6	0.1
2011	150.352	Tunnel-influence	Brown	Hatchery	391	682	45	8.4	325.6	0.4
2011	150.371	Tunnel-influence	Brown	Hatchery	332	401	59	19.0	1005.0	0.2
2011	150.392	Tunnel-influence	Brown	Hatchery	383	716	35	7.6	220.2	0.5
2011	150.412	Tunnel-influence	Brown	Hatchery	366	560	27	26.7	320.2	0.3
2011	150.430	Downstream	Brown	Hatchery	364	555	16	28.3	28.3	0.1
2011	150.450	Downstream	Brown	Hatchery	334	476	3	0.0	0.0	0.0
2011	150.472	Downstream	Brown	Hatchery	372	702	27	2.3	56.1	0.4
2011	150.490	Downstream	Brown	Hatchery	395	779	35	13.2	420.8	0.0
2011	150.512	Downstream	Brown	Hatchery	375	625	0	0.0	0.0	0.0
2011	150.530	Downstream	Brown	Hatchery	352	477	17	1.6	22.4	-0.1
2011	150.550	Downstream	Brown	Hatchery	345	482	65	17.4	1079.2	0.5
2011	150.571	Downstream	Brown	Hatchery	363	617	18	472.3	1416.8	-1.4
2011	150.591	Downstream	Brown	Hatchery	358	660	31	6.9	191.9	-0.1
2011	150.612	Downstream	Brown	Hatchery	341	463	50	11.4	534.5	0.8

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