

## 1. Contestant Profile

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## 2. Project Overview

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## ABSTRACT

Ecological restoration projects are often assessed by the extent of habitat or the species richness of taxa at a site. Functional measures of restoration effectiveness, on the other hand, have the goal of assessing habitat quality with respect to organismal activity, health, and abundance. In this project, we developed and tested a portable framework for HeidelbergCement (the Company) to partner with colleges and universities to augment restoration and biodiversity efforts. Our pilot study engaged professors and students in three undergraduate courses at McDaniel College -- Conservation Biology, Ecology, and Animal Physiology -- in authentic research on ecosystem function at Haines Branch, a stream and wetland site that was restored as part of the New Windsor Quarry expansion in Maryland, USA. In doing so, we deployed camera traps to evaluate mammalian species richness and habitat use, we investigated how within-site terrestrial and aquatic variables influenced tree health, crayfish abundance, and terrestrial invertebrate species richness, and we recorded fish species richness, assessed water quality, and piloted experiments assessing fish health. In this way, students in each course took ownership of their research questions and contributed to a robust assessment of ecosystem function at Haines Branch to provide recommendations to the Company. Some key findings include that mammalian species avoided the agricultural areas and forest cover was limited, so we recommend increasing both forested area and the planting of bushes at the edge of the forest. Also, the health of trees planted as part of the reconstruction did not vary based on distance to the stream, but juniper health was compromised by fungal galls and localized mortality of white oak warrants follow-up studies that have already been planned by students. Another important finding was that there was a significant difference in water quality in different locations along the stream but fish health did not vary based on site location. What did vary was fish diversity, which was greatest in constructed anastomosed channels. Our collaborative approach to assessing ecosystem function is flexible and can be modified based on Company goals for restoration, specific site variables, and expertise of professors at partnering institutions. The benefits of this framework for the Company, its community partners, and to science are many. Our results contribute to scientific knowledge by describing the health of regional wildlife and our framework has the potential to scale the number of scientific studies investigating the effectiveness of restoration efforts, leading to long-term improvements to the practice of restoration ecology. Professors and students gain increased opportunities, both to engage in scientific research and to form valuable partnerships with a corporation committed to sound environmental stewardship. The Company benefits from applying our framework at its restoration sites around the world because of the added value of research results pertaining to (1) the effectiveness of its restoration efforts for the promotion of biodiversity and (2) the generation of science-based restoration and management recommendations.

## ***A Framework to Assess Restoration Outcomes in Partnership with Educational Institutions***

### **INTRODUCTION**

This research project aims to expand on HeidelbergCement's "Indicators for the Representation of Successful Reconstruction Measures" (Rademacher et al. 2010) through the development of a framework to assess restoration effectiveness in partnership with institutions of higher education. Our research team developed and piloted this portable framework to engage college students and their professors in evaluating the ecological function and effectiveness of restoration and reconstruction activities conducted by HeidelbergCement (hereafter, the Company).

Assessing the efficacy of restoration projects often takes the form of measuring the extent of restored habitats or the species richness of particular taxa at restored sites (e.g., Roni et al. 2002; Harris et al. 2005; Arkle et al. 2014), including the evaluation method that has been developed by the Company (Rademacher et al. 2010). Functional measures of the effectiveness of ecological restoration are under development and have the aim of assessing the quality of the restored habitat with respect to the activity patterns, health, and abundance of taxa of interest (e.g., Ritchie et al. 2012; Jeffery et al. 2015). Whereas assessments of the presence and extent of different types of habitat at a restoration site are a necessary first step, understanding how organisms use habitat and how habitat quality influences their health and abundance provides a more holistic approach to restoration assessment and informs local land management practices.

Our framework for assessing restoration effectiveness in partnership with educational institutions (hereafter, the framework; Appendix A) enables the integration of multiple research disciplines in restoration assessment through the engagement of college students and faculty in authentic, course-based research at the Company's restoration sites. The workflow of the framework is as follows: In consultation with the Company and its subsidiaries, educational partners can begin by considering the educational resources available and the goals of the Company for that site. After evaluating the habitat types, biota, and environmental conditions, faculty can develop and propose work that is specific to their research expertise and that aligns with their goals for their classes and the goals of the Company for the restoration site. The framework then allows faculty to easily identify functional measures of restoration effectiveness. Thus, we offer a portable approach to assess the effectiveness of the Company's restoration projects, educate the next generation, achieve research goals, and generate useful management advice for the promotion of biodiversity.

Here, we report on our pilot test of this framework in three undergraduate courses (Conservation Biology, Ecology, and Animal Physiology) at McDaniel College in Maryland, USA. In this project, we assessed functional measures of restoration effectiveness at Haines Branch, a stream and wetland restoration site in New Windsor, Maryland. As with many streams in this heavily agricultural region, Haines Branch had been severely impacted by agricultural use for over 100 years. Haines Branch was restored as part of the expansion of the New Windsor Quarry, which is operated by the Company's subsidiary Lehigh Hanson. Since 2016, Lehigh Hanson's restoration project at Haines Branch has resulted in the restoration of 13,061 linear feet of stream, 1.22 acres of wetland areas, and 18,000 trees and shrubs (see Appendix B for site map and descriptions).

In piloting our framework, as described below, we investigated restoration success at Haines Branch via complementary approaches that evaluate aspects of organismal health, behavior, and abundance.

## Application of Framework

**Conservation Biology.** Terrestrial wildlife are especially valuable for the range of ecosystem services they provide, such as increasing ecosystem productivity and maintaining the ecosystem processes of seed dispersal and nutrient redistribution (Lavelle et al. 2006; Dirzo et al. 2014). Thus, the presence, abundance, and species richness of wildlife can be used to assess the condition and function of natural ecosystems, as well as the level of human influence (Audino et al. 2013; Wortley et al. 2013). Camera trapping is a relatively new research approach that provides a low cost means to evaluate the status of wildlife, and thus ecosystem function. The overarching goal of this wildlife study was to evaluate the ecological condition of the Haines Branch restoration site using wildlife presence and diversity as a proxy of ecosystem function. The specific research objectives of the wildlife study were to: (1) establish a baseline of mammal species diversity present at the site relative to potential species; (2) evaluate the differences in habitat use of wildlife at the site; and (3) evaluate the level of human influence at the site by measuring the activity patterns of red foxes.

**Ecology.** Riparian zones are regions of high biodiversity due to the juxtaposition of terrestrial and aquatic habitats and the heterogeneity of the terrestrial vegetation (Naiman et al. 1993). Plants provision resources, serve as habitat for animals, control erosion, and influence nutrient cycling (Costanza et al. 1997; Lindenmayer et al. 2010); thus plant health can impact the capacity of the ecosystem to support biodiversity and stream health. The approach in the ecology course was to investigate how within-site terrestrial and aquatic variables influence organismal health, abundance, and species richness. This was achieved through three projects, with the following objectives: (1) assess whether water stress has impacted the health of trees planted in the riparian zone; (2) evaluate whether major terrestrial habitat types (wetlands, upland fields, and the grassland adjacent to the stream) support different levels of biodiversity, focusing on invertebrates as indicator taxa (e.g., Jansen 1997; Audino et al. 2013); and (3) investigate whether the abundance and size of crayfish, a group of moderately sensitive aquatic indicator species, varies with water quality.

**Animal Physiology.** Water quality can also affect fish health, and physiological responses to within site variables can be detected before changes in abundance might be apparent. Such an approach is taken in conservation physiology, a burgeoning field that establishes mechanistic links between environmental health and animal health (Cooke et al. 2013). Conservation physiology provides a proactive and pragmatic approach to balancing the needs of society and preservation of the natural world (Madliger et al. 2017). Physiological approaches also contribute to ecological restoration (Cooke and Suski, 2008), and this is especially useful for conserving fish in freshwater streams that are vulnerable to changes in the terrestrial areas bordering the stream (Jeffrey et al. 2015). Fish health can be assessed using a number of biochemical and physiological indicators that can act as an early-warning screening tool (Adams et al. 2011), measuring primary (e.g., plasma cortisol levels), secondary (e.g., blood glucose levels or metabolic rate), or tertiary (whole-organism, e.g., swimming/escape) responses to stress (Sopinka et al. 2016). The objectives of the animal physiology component of this study were to (1) establish approximate fish species richness and abundance at different locations of the stream; (2) measure abiotic water quality variables at different locations to define parameters for future experiments on physiological response to changes in water chemistry; and (3) to pilot, optimize, and validate physiological experimental techniques.

Based on these combined approaches at Haines Branch, this report documents a robust terrestrial animal community and describes how organismal abundance and health vary, at least in part, with abiotic conditions. We also describe a set of recommendations for future restoration efforts at Haines Branch. We conclude this report by highlighting how our multidisciplinary research approach can be both flexibly implemented at the Company's restoration sites around the world and result in multifaceted benefits to the Company. These benefits include partnerships between industry and local institutions of

higher education that can generate not only positive press about the Company's environmental stewardship but also robust datasets that result in science-based management recommendations.

## METHODS

A total of 48 students from Conservation Biology, Ecology, and Animal Physiology courses designed studies to assess the functional health of the restored ecosystem at Haines Branch. We piloted the framework using approaches from our areas of expertise and describe the protocols we used here.

**Conservation Biology.** A total of 21 camera traps were deployed across the 379-acre study site. The cameras were deployed semi-randomly across the study site in areas likely to be used by mammals (e.g., watering holes and ridgelines). The cameras were deployed for 44 "trap nights" (Oliver et al. 2017) from 2/24/2018 - 4/7/2018. The cameras used were the brand Browning Strike Force with 'night vision' infrared flash and set to 3-shot rapid fire with a 2-minute delay following each photo set taken. Once the camera imagery was collected, it was evaluated and the image capture data collated. Information recorded for each image capture included camera station code, species code, number of individuals, date of capture, and time of capture. To establish a baseline of mammal species diversity at the site relative to species that were historically present in the region, the species list of Paradiso (1969) was used. Specifically, all mammal species listed in our local ecoregion (Piedmont forests) with ranges overlapping the study site, and with habitat preferences that include the habitats found at the study site were identified and compared to the actual species photographed at the site. To determine the habitat preferences of each species identified, we used the Audubon Mid-Atlantic Field Guide (Alden 1999). To evaluate the use of the four different habitats at the restoration site by mammals (i.e., grasslands, forests, wetlands, and agricultural areas), the habitat type of each camera location was determined and the capture of each species was recorded using the established occurrence measurement of Species Events / Trap Nights x 100 (Oliver et al. 2017). Chi-square tests were used to evaluate whether the frequency of captures among habitats was proportional to the percent cover of habitat. To evaluate the level of human influence at the site, the timing of the image capture of fox activity was recorded and analyzed. The assumption used is that high levels of human presence would push the foxes to behave unnaturally, specifically not being present during preferred dawn/dusk hunting periods and only present at night, which is a behavioral response detected in a variety of species under high levels of human pressure (Gaynor et al. 2018).

**Ecology. Tree Health.** In the tree health study, we set out to investigate whether the size and health of trees installed at Haines Branch depended on potential water stress, using distance from a water source as a proxy. This investigation was restricted to trees planted within 50 m of the stream or a wetland and focused on the two most common trees at this site, *Juniperus virginiana* and *Quercus alba*. Tree health was assessed using three measures: diameter at breast height (DBH), tree height, and average crown diameter (as in Schomaker et al. 2007). Initial observations noted fungal galls of cedar-apple-rust on individuals of *J. virginiana*; the presence and number of galls were therefore also recorded. The relationships between measures of tree health and distance to water source were assessed with regression (Analysis ToolPack in Excel). **Terrestrial invertebrates.** To investigate the extent to which terrestrial habitats support invertebrate abundance and taxonomic richness, pitfall traps were placed in three habitat types: adjacent to wetlands, in upland fields, and adjacent to the stream (see Appendix B for locations). Pitfall traps were constructed from two nested plastic cups partially filled with soapy, salty water as a preservative; funnels were installed in the top cup to prevent the catch of small vertebrates, and plates supported by 5 cm bolts covered the traps to exclude precipitation. Pitfall traps were deployed on 4/4/2018 and retrieved on 4/11/2018. Specimens were transferred to 70% ethanol for preservation, and taxa were identified in the lab. All individuals were identified to class, and insects were identified to order. Analysis of variance (ANOVA) was used to test whether habitat types supported different taxonomic richness and total abundances of invertebrates. **Crayfish.** To examine whether crayfish abundance and body size varied with water quality, timed crayfish surveys were conducted. Crayfish

were surveyed for repeated 45 min intervals at three sites (Sites 1, 2 & 4; Appendix B) in 4.5 m long sections of stream of similar width (3 m) and depth. All crayfish encountered in these timed samples were collected, measured, and then released at the end of the sampling period. Concurrently, the following aspects of water quality were measured: pH, dissolved oxygen, nitrate (in collaboration with Animal Physiology students, below), and turbidity (Test Assured Secchi disk, Inlet Innovations). Correlations between measures of water quality and crayfish size and abundance were investigated with regression and ANOVA in Excel.

**Animal Physiology.** Students from the animal physiology course focused on assessing water quality and fish health at different sites along the stream (Appendix B). Dissolved oxygen readings were measured in the field at sites 2 and 4 using a dissolved oxygen pen (Sper Scientific). Water samples were also collected from each of the four sites and brought to the lab for immediate assessment of pH (Vernier), nitrate, and nitrite (NutraFin) levels. Fish were collected using dip nets or small seines and immediately placed into an anesthetic bath of tricaine methanesulfonate (MS-222; Western Chemical) at a concentration of 50 mg/L. Fish were photographed with a scale to determine species identity, in an effort to assess general biodiversity at each location. Fish were then either released, used for physiological testing in the field, or transported to the lab for further experimentation on organismal health. The McDaniel College IACUC reviewed all methods for animal care and use. The blacknose dace, *Rhinichthys atratulus*, was the most abundant species and was thus the choice for physiological pilot studies.

Assessment of primary, secondary, and tertiary stress responses occurred both in the lab and in the field. Briefly, to assess tertiary swimming responses in dace, high-speed video was collected from individuals from different sites using a small aquarium filled with stream water with a 1 cm grid in view. Videos were taken using an iPhone 6 at a rate 240 frames per second and were imported into free NIH ImageJ software (Schneider et al. 2012) to quantify movements. To analyze biochemical indicators (glucose, Accu-Chek, Roche, and cortisol, Cayman ELISA kit, #500360) of fish stress in blood plasma, fish were humanely sacrificed in the field using an overdose of buffered MS-222. The caudal fin was severed and blood from the caudal vein was collected using heparinized capillary tubes. For swimming endurance tests, dace were transported to the lab in an anesthetic bath and then placed into a 10-gallon aquarium with water collected from the stream. Fish were acclimated to room temperature by floating ice packs to ensure no more than a 2°C change per day (per Nelson et al. 2008). Individual fish were placed in a flume (flow rate 0.4 L/min) constructed by students per Bolduc et al. 2002. The fish were determined to be at maximum exhaustion when they avoided the flow and stopped swimming. To answer whether water chemistry and physiological indicators differed among locations, ANOVA was used in JMP software (version 13, SAS) and a Tukey-Kramer HSD post hoc test was used to differentiate how the sites differed. In the cases where only two sites were being compared, a two-tailed t-test was run in Microsoft Excel.

## RESULTS

**Conservation Biology.** The total number of potential mammal species in the Piedmont region of Maryland whose range and habitat preferences overlap with the site is 36 species. Of the 36 species that could be present at the site, nine species were captured on the cameras (Table 1; please see Appendix C for all Tables and Figures). All nine species captured, with the exception of the coyote, are species that are generalists and tolerant of human influence. By order of most common, the following mammals were present at the site: whitetail deer, red fox, raccoon, rabbit, squirrel, opossum, groundhog, coyote, and skunk (scientific names provided in Table 1). The habitat use of wildlife at Haines Branch varied depending on the species (Table 2; Fig. 1). Forest areas had the highest capture frequency for foxes, raccoons, rabbits, opossums, skunks, squirrels and groundhogs. Wetlands had the highest capture frequency for deer and coyotes. The grasslands did not have the greatest capture frequency for any species; however, they did have the highest diversity of mammalian species. Generally, the locations

where each species was found aligned with the Mid-Atlantic field guide (Alden 1999) and their known habitat, with the exception of coyotes that were not spotted in the forests and opossums that were not spotted in the wetlands. However, these exceptions may be due to the low number of image captures of coyotes and opossums ( $n = 3$  and  $n = 11$ , respectively). Across all species, capture frequency of mammals departed significantly from expectations based on the percent cover of habitats ( $\chi^2 = 517.42$ ,  $df = 3$ ,  $P < 0.0001$ ). Specifically, mammals were captured more frequently than expected in wetlands (265 observed, 131 expected) and forests (218 observed, 98 expected), slightly less frequently than expected in grasslands (170 observed, 196 expected), and far less frequently than expected in the agricultural area (0 observed, 229 expected; similar results were obtained when expected frequencies were calculated based on the number of camera traps per habitat). In terms of fox activity, individuals were most active during the time code '7', or 6PM - 9PM. High levels of activity were also observed between the hours of 9PM - 12AM and 6AM - 9AM. Foxes were the least active during the hours of 3PM - 6PM (Fig. 2).

**Ecology. Tree Health.** Although *J. virginiana* and *Q. alba* differed in size (e.g., *Q. alba* individuals were significantly taller,  $F = 16.85$ ;  $P = 0.0002$ ), no aspect of tree size varied significantly with distance to a water source (Fig. 3;  $P > 0.05$  for all regressions). Similarly, although the number of galls on individuals of *J. virginiana* varied considerably, this variation was not related to distance to a water source (Fig. 4;  $P > 0.05$ ). **Terrestrial Invertebrates.** In total, 301 individuals of 8 invertebrate taxa were captured in the pitfall trap study. The abundance of taxa differed markedly; springtails (Order Collembola) were by far the most abundant taxon identified in this study, followed by spiders and flies (Fig. 5A). The abundance of each taxon, however, was unrelated to habitat type, with the exception of mosquitoes (Culicidae:  $F = 7.7$ ,  $P = 0.017$ ; all others:  $P > 0.05$ ). The taxonomic richness of invertebrates also did not differ among habitat categories (Fig. 5B). **Crayfish.** Crayfish body length did not differ significantly among sampling sites ( $F = 0.268$ ,  $P = 0.766$ ; Fig. 6A). Crayfish abundance, however, differed substantially among sites; the most crayfish were collected from the farthest upstream site (site 4), and only one crayfish was collected mid-reach across the combined 90 minutes of sampling (site 2; Fig. 6B). Of the water quality variables, turbidity varied substantially among sites and was related to crayfish abundance, with more crayfish collected at sites with clearer water (deeper Secchi Disk depth; Fig. 6C). This relationship, though visually striking, was not statistically significant ( $R^2 = 0.946$ ,  $n = 3$ ,  $P = 0.148$ ), likely due to a small sample size.

**Animal Physiology. Water quality.** Water quality measurements differed among sites. pH differed significantly among the four sites ( $F = 58.52$ ,  $P < 0.0009$ ) with the most alkaline regions located upstream (Fig. 7A). Both nitrate and nitrite levels differed significantly across sites ( $F = 28.3$ ,  $P < 0.0001$  and  $F = 5.67$ ,  $P < 0.0077$ , respectively): Site 3 had significantly higher nitrate levels than all other sites (Fig. 7B). Sites 1 and 3 had the highest nitrite levels, though only site 1 had significantly higher nitrite than the rest of the sites (Fig. 7C). The average concentration of dissolved oxygen at site 4 (17.35 mg/mL) was significantly higher than that at site 2 (12.125 mg/mL;  $P = 0.0020$ ). **Biodiversity.** The blacknose dace *Rhinichthys atratulus* was by far the most abundant species in the stream, however we found several other species, particularly at site 2, which was the most species-rich site. The next most common fish species in this study was the tessellated darter, *Etheostoma olmstedii*. Several longnose dace, *Rhinichthys cataractae*, and white suckers, *Catostomus commersoni*, were also caught and released. Single individuals of two other species were caught and released at site 2: a juvenile bullhead catfish, *Ameiurus nebulosus*, and a minnow, likely a *Notropis* species (species-level identification would have required sacrificing the animal). **Physiology.** Pilot experiments evaluating fish health focused primarily on optimizing protocols and validating physiological measurements. None of the physiological variables differed statistically among sites. However, several preliminary results are worth reporting, as we plan to build on these in subsequent years. Blood collection proved difficult from such small fish, and glucose and cortisol levels were not significantly different among dace from different sites. A major result from the swimming performance (flume) pilot project was that blacknose dace can successfully survive the potential stressors of transport to the lab, minor experimental handling, and transport back to the field site. The average time to exhaustion for fish from site 2 was slightly higher than the average for site 4



(8.84 minutes, and 7.95 minutes, respectively), but there was no significant difference between sites. To quantify reaction time and escape response (C-start) variables, high-speed videos from the field were digitized to obtain kinematic variables but there were no differences among fish from different sites.

## DISCUSSION

The purpose of this study was to develop and pilot a portable framework to engage professors and their college students in evaluating the effectiveness of the Company's restoration activities. As intended, our results demonstrate that this framework can be successfully implemented in college classes encompassing a variety of scientific disciplines. Importantly, our results also build upon and expand the scientific insights and management recommendations that can be obtained from the Company's existing "Indicators for the Representation of Successful Reconstruction Measures" (Rademacher et al. 2010). Below, we review the major scientific findings of our research, with particular emphasis on specific management recommendations for the Haines Branch restoration site. We describe the utility of this framework for assessing the restoration of ecological function and the promotion of biodiversity, and we conclude by highlighting additional benefits of the framework for the Company moving forward.

**Conservation Biology.** The use of terrestrial animals as indicators of ecosystem health and restoration effectiveness is well established. Our finding that only 9 of 36 mammal species that should be present were present is certainly concerning. Some of the mammals may in fact be present but were not detected by the cameras due to their small size and life close to the ground. Additionally, colder seasonal effects could have skewed the sample with some species that may have been hibernating or simply less active. However, our findings still show that the Haines Branch restoration site is not functionally intact relative to undisturbed sites (Paradiso 1969). This finding draws the conclusion that to improve the integrity and function of the ecosystem through wildlife presence, additional restoration activities are needed. One management recommendation to improve the habitat available for mammals is to increase the amount of forest cover on the site, as increased woody biomass is known to support mammal species diversity (Ferrer-Castán, et al. 2016) and only ~15% of the site is currently forested. Another management recommendation to improve mammal species diversity is to increase the amount of edge habitats on the property as edges are known to support a wider variety of species, especially small mammals, which were underrepresented in the results (Menzel et al. 2009). Future wildlife surveys on the site may be more robust if they exclude rare species because these species are not good indicators of species richness in the eastern US (Meyers and Joly 2001).

Our study comparing mammal presence by habitat type offers several insights to inform management of the Haines Branch site. The first is the relatively low use of the grassland habitats of the site compared to the forest area, which contrasts with where red foxes are typically more prevalent. This finding may indicate the grassland habitats should be enhanced, including removal of existing invasive species and increased edge habitat through planting of bushes near forest edges to increase the abundance of small mammals that foxes prefer to eat (Alden 2014). The other major finding of this analysis was the complete absence of mammals in the agricultural area. This finding may indicate that the abandoned fields are poor habitats for most species and could be improved through ecological restoration. Such actions could include removing invasive grass species now flourishing on the site and planting the agricultural areas under forest or grass cover. Thus, improving the habitat quality of the agricultural areas is a top priority for enhancing wildlife abundance and diversity on the site. The study of red fox activity patterns shows there is a large population of red foxes using the site, which indicates a similarly large population of prey species, such as rabbits and mice. Likewise, the high activity levels of foxes during the time periods we would expect the highest level of activity under natural conditions (i.e., dawn and dusk) indicate that the site is under relatively low levels of human pressure. Collectively, these findings show that the site has a functional predator-prey dynamic for foxes and thus a relatively high

level of ecosystem function, but that the site could be further improved for red foxes and their prey by increasing the amount of edge habitat and forest cover across the site.

**Ecology.** Through the complementary approaches of investigating tree health, terrestrial invertebrate abundance and richness, and crayfish size and abundance, we found mixed support for our initial hypotheses that variation in within-site conditions would influence the health, abundance, and richness of taxa at Haines Branch. When successfully established, trees can sequester carbon, intercept nutrient runoff, mitigate air pollution, and provide habitat, protection, and resources for diverse consumers (MEA 2005); thus, the survival and growth of trees is integral to the ecological function of restoration sites. We had hypothesized that tree health would depend on distance from the stream or wetlands, using distance as a proxy for water availability. Our study of 52 individuals of *J. virginiana* and *Q. alba* demonstrated that tree size and the presence and abundance of fungal galls did not differ with distance to the stream. We did, however, note high variability in tree size and gall abundance, and visits to Haines Branch after the conclusion of the study revealed localized oak mortality. Although both of these tree species are widely used in ecological restoration due to their broad abiotic tolerances (Eggemyer et al. 2009), their health and growth can be negatively affected by water stress and other abiotic conditions (McKinley 2005, LeBlanc and Terrell 2009). We are therefore developing follow-up studies on tree health, to be conducted this fall. These studies will focus on the following priorities: (1) assessing survival, size, and incremental growth of trees; (2) directly measuring soil quality; (3) determining the relationship between the health of *J. virginiana* and the abundance of fungal galls; and (4) assessing the ecosystem function provided by surviving trees. Should these follow-up surveys discover widespread losses of *Q. alba*, we would strongly suggest replanting with tree species more tolerant of local conditions and transplant stress.

Our study of invertebrates revealed little difference in the biodiversity of terrestrial invertebrates across habitats and interesting correlations between aquatic invertebrate abundance and water quality. We found few differences in the abundance and taxonomic richness of invertebrates in our pitfall trap samples when comparing grassland, wetland, and habitats adjacent to the stream. Clearly, invertebrates have colonized the restored vegetation at Haines Branch, but the cold weather during our study period may have limited our ability to differentiate the capacity of these habitats to support different levels of biodiversity. We had originally set out to compare the activity density of carabid beetles with the pitfall traps. Carabids (family Carabidae) are used worldwide as a standard indicator taxon, as species in this beetle family vary in environmental sensitivity and habitat requirements (Rainio and Niemelä 2003). However, with such cold temperatures, most large invertebrates such as carabids are unlikely to be active. Repeating the study during the growing season and identifying captured individuals to a lower taxonomic level should afford the opportunity to (1) compare the species richness and abundances of invertebrates across habitats and (2) determine whether rare, wetland specialist species have colonized and established populations in and around the restored wetlands at the site. Whereas the terrestrial invertebrate activity was likely limited by the cold, crayfish were abundant in the stream, and sampling revealed correlations between crayfish abundance and abiotic conditions, especially water clarity. Crayfish are considered indicator species that are moderately tolerant of environmental degradation (McDonald et al. 1991), so our initial results suggest adequate water quality at Haines Branch for at least some aquatic species. As our results are correlative and restricted to one group of species, future studies at this site are being planned, with an emphasis on (1) sampling aquatic species of varying environmental sensitivities and (2) investigating the causal mechanisms leading to the correlation of crayfish abundance with water clarity.

**Animal Physiology.** Fishes rely on matter exchange (e.g., gases, ions) with the surrounding water for all of their basic physiological functions. No fish were found at site 3, a location with high pH, nitrates, and nitrites (Fig. 7A-C). The pH level of water has been shown to impact ion balance in the cells

of fish. Both highly acidic and alkaline water have been linked to high potassium ion concentrations in blood (Garcia et al., 2014) as well as sodium and chloride imbalance (Cone, 1988; Val et al., 1998; McGeer and Eddy, 1998); all of these ions are necessary for successful firing of neurons. Increased nitrate and nitrite levels can build up in fish tissues, affect fish health, and damage reproductive modes of freshwater fishes (Eddy and Williams, 1987). Environmental stress can also affect whole-animal status, impacting swimming performance (e.g., speed, endurance; Nelson et al. 2008) and reflexes (Davis, 2010) or escape responses such as C-starts, all of which have strong ecological relevance (Plaut, 2001). While measurements of fish health did not vary significantly across stream locations, the dace from site 2 had slightly higher times to exhaustion; combined with the fact that site 2 also had the highest fish diversity, this preliminarily suggests that anastomosed channel sections are effective for creating pockets of fish biodiversity and healthier conditions for fish in restored freshwater streams.

Armed with the results of water quality, fish biodiversity, and physiological pilot projects, our work will continue in a longitudinal study of organismal health at the restored stream. The physiological pilot projects produced optimal designs for experiments, and the Spring 2019 Animal Physiology class will build on this year's best design to improve the functionality and reliability of the protocols. Our preliminary work also generated empirical ranges of water quality that will serve as the treatment parameters in next year's controlled physiological experiments in the lab. The goal of these structured trials will be to define causal, mechanistic relationships between water quality metrics and fish health. In this way, we will ultimately identify which water chemistry indicators are most related to fish health.

**The benefits of this framework for the Company and its community partners are many.**

Many local colleges and universities lack access to field sites, limiting the potential for their students to conduct field research applicable to real-world scenarios. Through the implementation of our framework to evaluate ecosystem function at the Company's restoration sites, professors and students gain increased opportunities, both to engage in scientific research and to form a valuable partnership with an international company committed to sound environmental stewardship. The Company benefits from applying our framework at its restoration sites around the world because of the added value of research results pertaining to (1) the effectiveness of its restoration efforts for the promotion of biodiversity and (2) the generation of science-based restoration and management recommendations. Additionally, because the framework implies additional relationship-building between the Company and local partners, the Company will benefit from increased exposure to local students, community members, and the media. These new connections have the potential to generate positive press for the Company in local media markets and expand the access of the Company to talent pools of students who are aware of the Company and its corporate social responsibility efforts.

**The benefits of this framework to science are many, both globally and locally.** Globally, this framework builds on the recent growth of engaging local citizens in citizen science (Bonney et al. 2014). This framework expands that effort to include local citizens (students) in scientific cooperation with local corporations. To the best of our knowledge, our framework is the first to explicitly generate citizen-corporate relationships centered around the collection of scientific data. Another global scientific benefit of this framework is its potential to scale the number of scientific studies investigating the effectiveness of restoration efforts, leading to long-term improvements to the practice of restoration ecology. Locally, the scientific benefits of our pilot study and the application of our framework to assess restoration effectiveness include increasing awareness among local decision-makers and company managers about the presence and abundance of local wildlife, as well as the condition of environmental variables of interest, such as water quality, stream structure, and soil composition. For example, we found no significant difference in fish health among study sites despite the fact that there were significant differences in water chemistry among sites. Similarly, we found that a diversity of invertebrates established in the terrestrial habitats and that crayfish abundance is likely correlated with water quality.

We also found that tree health varied, but not with distance to the stream and wetlands. Together, these findings show that a variety of indicator species are present on the site and they indicate favorable restoration outcomes to date. Lastly, our pilot research generated a set of management interventions that could be used to improve the ecological condition of the site. For example, our wildlife pilot study found that a diversity of wildlife are using the restoration site, but many species that should be present were absent or at low abundance and that by increasing the amount of forest cover and edge habitat substantial increases in wildlife abundance and diversity are likely. Lastly, one way our framework could be easily expanded to increase its scientific benefits and community impacts is to incorporate BioBlitz's, which are community-based activities that engage citizens and scientists in a joint effort to inventory the presence of local wildlife and plant species. When conducted over time, BioBlitz's can be used to monitor the health of local species populations and detect important changes.

## **FINAL CONCLUSIONS**

In closing, this research project expands on the Company's "Indicators for the Representation of Successful Reconstruction Measures" through the development and testing of a framework to assess restoration effectiveness in partnership with educational institutions. Our research team developed and piloted this framework and showed how it can be used to engage college students and their professors in evaluating the ecological function and effectiveness of the Company's restoration and reconstruction activities. Our research results demonstrate that the restoration project at Haines Branch is effective to date and generated a variety of management recommendations to improve the quality of the site for local wildlife and ecosystem services. As shown, our framework can be applied by the Company at its restoration, reconstruction, and conservation sites around the world with resulting benefits for community-corporate relations, science, and the promotion of biodiversity.

## **ACKNOWLEDGEMENTS**

This project would not have been possible without the support of top managers at Lehigh Hanson's Union Bridge Quarry in Union Bridge, Maryland. We are especially grateful to Environmental Engineer Kurt Deery and Plant Manager Kent Martin for their encouragement and support of this project. We would also like to thank the administration of McDaniel College for their support of this project, particularly, Provost Julia Jasken, Josh Ambrose, and Erin Giles. Dr. Randy Morrison provided advice on local stream fauna.

**Project Tags (select all appropriate):**

This will be use to classify your project in the project archive (that is also available online)

**Project focus:**

- ☒ Beyond quarry borders
- ☒ Biodiversity management
- ☐ Cooperation programmes
- ☒ Connecting with local communities
- ☒ Education and Raising awareness
- ☐ Invasive species
- ☒ Landscape management
- ☐ Pollination
- ☒ Rehabilitation & habitat research
- ☒ Scientific research
- ☐ Soil management
- ☒ Species research
- ☒ Student class project
- ☐ Urban ecology
- ☒ Water management

**Flora:**

- ☒ Trees & shrubs
- ☐ Ferns
- ☒ Flowering plants
- ☐ Fungi
- ☐ Mosses and liverworts

**Fauna:**

- ☐ Amphibians
- ☐ Birds
- ☐ Insects
- ☒ Fish
- ☒ Mammals
- ☐ Reptiles
- ☒ Other invertebrates
- ☐ Other insects
- ☐ Other species

**Habitat:**

- ☐ Artificial / cultivated land
- ☐ Cave
- ☐ Coastal
- ☒ Grassland
- ☐ Human settlement
- ☐ Open areas of rocky grounds
- ☐ Recreational areas
- ☐ Sandy and rocky habitat
- ☐ Screes
- ☐ Shrub & groves
- ☐ Soil
- ☐ Wander biotopes
- ☒ Water bodies (flowing, standing)
- ☒ Wetland
- ☒ Woodland

**Stakeholders:**

- ☐ Authorities
- ☐ Local community
- ☐ NGOs
- ☐ Schools
- ☒ Universities

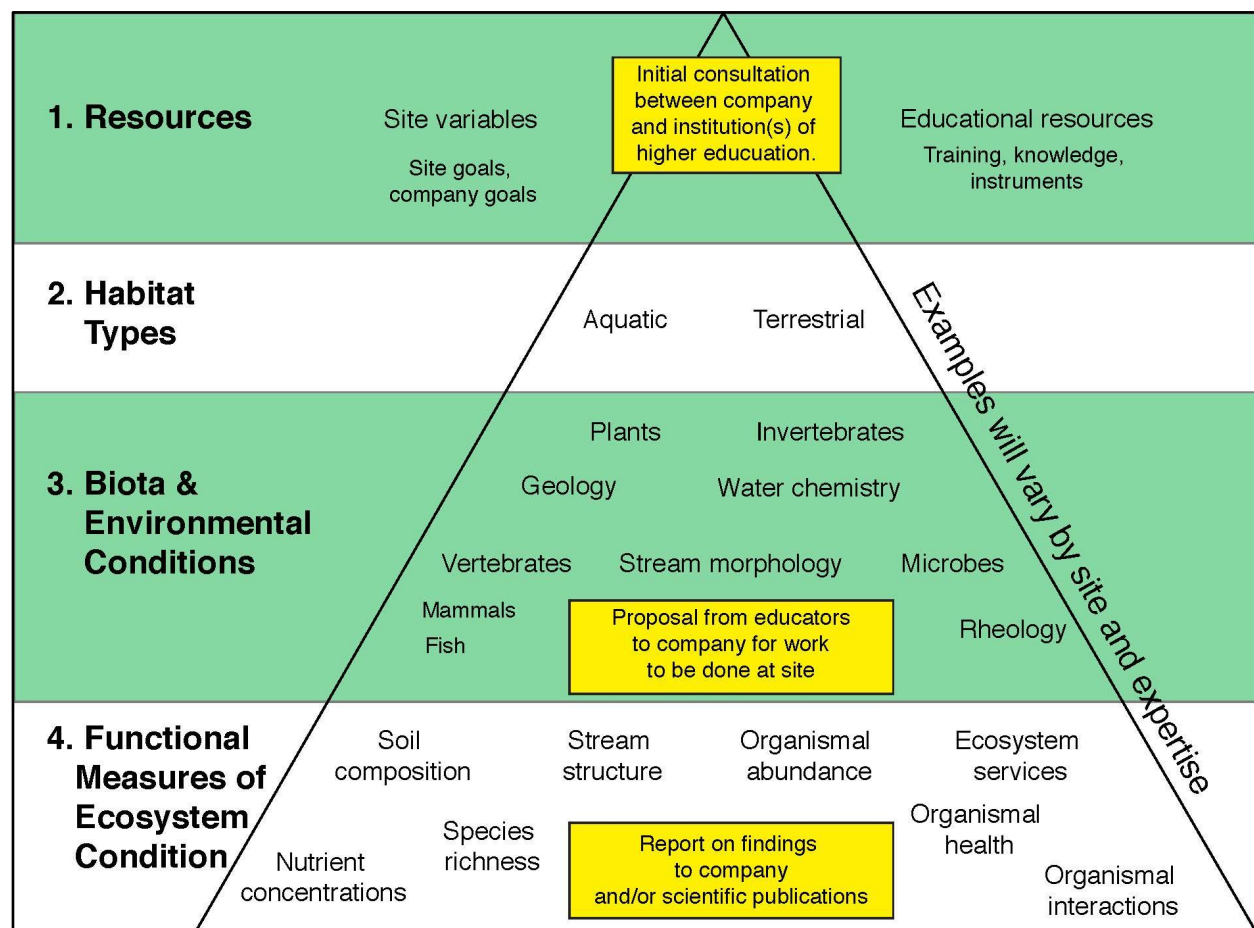
## APPENDIX A. EDUCATIONAL PARTNERSHIP FRAMEWORK

In order to accomplish our scientific objectives, we developed a framework to infuse authentic research projects into undergraduate classrooms. In this way, the number of scientists – and replicates – is increased, strengthening the power of the data and the impact of the collaboration.

In the above report we present the results from a pilot of this scholarly educational framework using three college courses: Conservation Biology, Ecology, and Animal Physiology. Students from each course assessed different aspects of ecosystem health to provide a robust analysis of restoration success. We propose that this framework can be incorporated into partnerships around the world and can vary based on expertise of local scientists. Consequently, a system of sharing protocols and data could contribute to increasing the effectiveness of restoration projects around the world.

### Collaborative Workflow - Decision Tree

Each partnership will vary based on resources, habitat types, etc. Representatives from the Company and the educational institutions will collaborate throughout the process (yellow boxes).



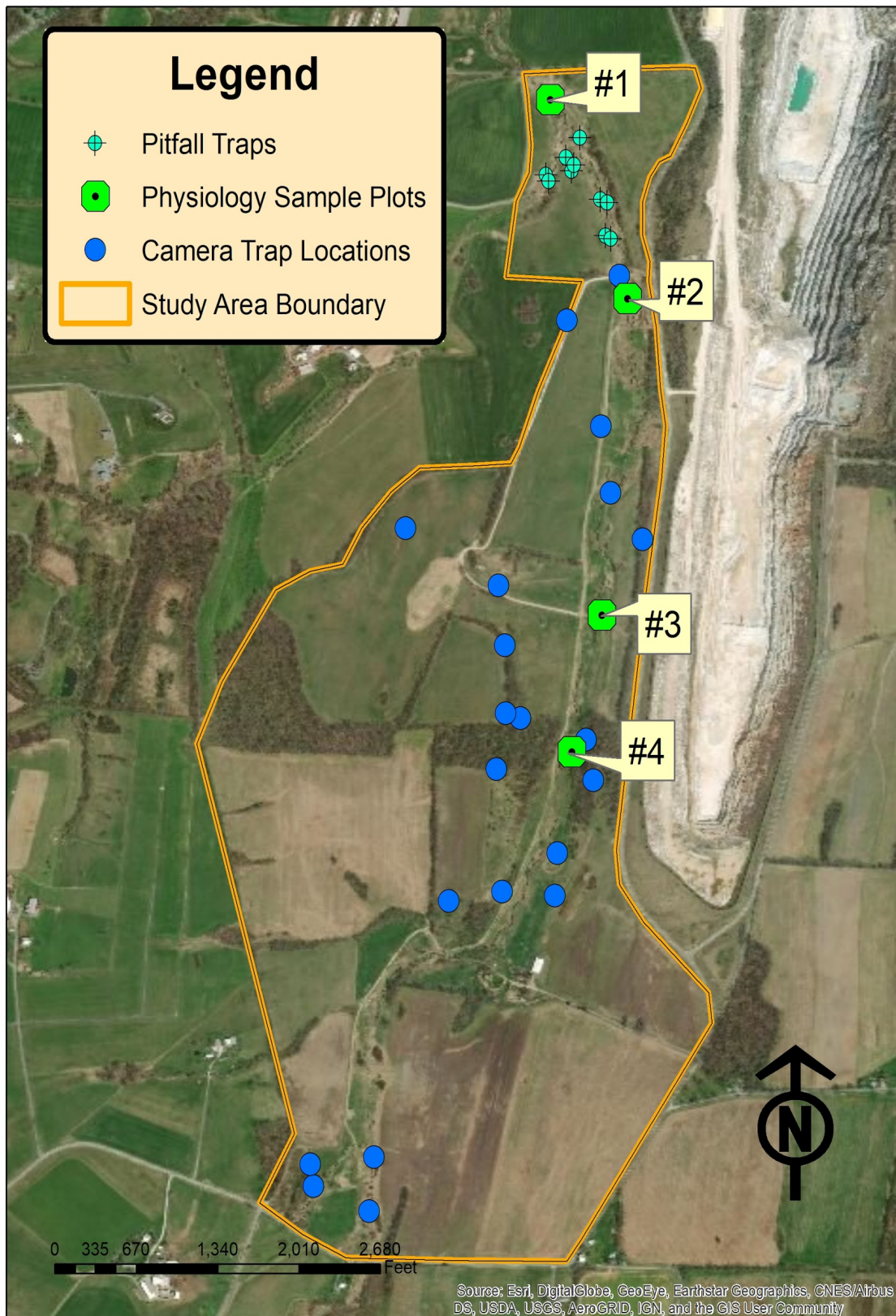
## APPENDIX B. SITE DESCRIPTIONS AND STUDY AREA MAP

**Aquatic Sites.** Because each project assessed an aspect of water quality and animal health, microhabitats were chosen along the stream based roughly on differences in the riparian zone. Site 1 is the northernmost sampling site, borders a dairy farm, and is most downstream of the sites sampled. Site 2 is closest to a road and the limestone quarry and includes a small network of anastomosed channels and confluences. Site 3 is a single thread channel, also in close proximity to the road and the quarry, and is bordered by grasslands. Site 4 is most upstream, and is bordered by small areas of forest.

**Terrestrial Sites.** The pitfall traps are replicated in three different habitat types (i.e., wetlands, grasslands, and forests). The wildlife camera traps were distributed across the landscape by habitat type and specifically by locations where mammals were likely to occur, including game trails, watering holes, and ridgelines. Trees were sampled throughout the field site and are not shown in the figure below.



# Study Area Map of Research Plots, Camera Trap Locations, and Pitfall Traps at Haines Branch





## APPENDIX C. TABLES AND FIGURES

**Table 1. List of Wildlife Species Captured on Camera Traps.** (A) Relative to Species Expected Based on Historical Presence in Mid-Atlantic Piedmont Ecoregion (B).

*Table 1A. Wildlife Species Captured.*

Order	Common Name	Scientific Name
Artiodactyla	White tailed deer	<i>Odocoileus virginianus</i>
Carnivora	Coyote	<i>Canis latrans</i>
Carnivora	Raccoon	<i>Procyon lotor</i>
Carnivora	Red fox	<i>Vulpes vulpes</i>
Carnivora	Striped skunk	<i>Mephitis mephitis</i>
Lagomorpha	Eastern cottontail	<i>Sylvilagus floridanus</i>
Marsupialia	Virginia opossum	<i>Didelphis virginiana</i>
Rodentia	Eastern gray squirrel	<i>Sciurus carolinensis</i>
Rodentia	Groundhog	<i>Marmota monax</i>
Non-Native Wildlife	Domestic cat	<i>Felis silvestris catus</i>
Non-Native Wildlife	Domestic dog	<i>Canis lupus familiaris</i>
Non-Native Wildlife	Human	<i>Homo sapiens</i>

*Table 1B. Wildlife Species Historically Present.*

Order	Common Name	Scientific Name
Artiodactyla	White tailed deer	<i>Odocoileus virginianus</i>
Carnivora	Coyote	<i>Canis latrans</i>
Carnivora	Raccoon	<i>Procyon lotor</i>
Carnivora	Red fox	<i>Vulpes vulpes</i>
Carnivora	Bobcat	<i>Lynx rufus</i>
Carnivora	Gray fox	<i>Urocyon cinereoargenteus</i>
Carnivora	Long-tailed weasel	<i>Mustela frenata</i>
Carnivora	Mink	<i>Neovison vison</i>
Carnivora	North American river otter	<i>Lontra canadensis</i>
Carnivora	Striped skunk	<i>Mephitis mephitis</i>
Chiroptera	Big brown bat	<i>Eptesicus fuscus</i>
Chiroptera	Eastern red bat	<i>Lasiurus borealis</i>
Chiroptera	Keen's myotis	<i>Myotis keenii</i>
Chiroptera	Little brown bat	<i>Myotis lucifugus</i>
Insectivora	American pygmy shrew	<i>Sorex hoyi</i>
Insectivora	Eastern mole	<i>Scalopus aquaticus</i>

Insectivora	Masked shrew	<i>Sorex cinereus</i>
Insectivora	North American least shrew	<i>Cryptotis parva</i>
Insectivora	Northern short-tailed shrew	<i>Blarina brevicauda</i>
Insectivora	Star-nosed mole	<i>Condylura cristata</i>
Lagomorpha	Eastern cottontail	<i>Sylvilagus floridanus</i>
Marsupialia	Virginia opossum	<i>Didelphis virginiana</i>
Rodentia	American red squirrel	<i>Tamiasciurus hudsonicus</i>
Rodentia	Eastern chipmunk	<i>Tamias striatus</i>
Rodentia	Fox squirrel	<i>Sciurus niger</i>
Rodentia	House mouse	<i>Mus musculus</i>
Rodentia	Meadow jumping mouse	<i>Zapus hudsonius</i>
Rodentia	Meadow vole	<i>Microtus pennsylvanicus</i>
Rodentia	Muskrat	<i>Ondatra zibethicus</i>
Rodentia	North American beaver	<i>Castor canadensis</i>
Rodentia	North American deer mouse	<i>Peromyscus maniculatus</i>
Rodentia	Norway rat	<i>Rattus norvegicus</i>
Rodentia	White-footed mouse	<i>Peromyscus leucopus</i>

Rodentia	Woodland vole	<i>Microtus pinetorum</i>
Rodentia	Eastern gray squirrel	<i>Sciurus carolinensis</i>
Rodentia	Groundhog	<i>Marmota monax</i>
Non-Native Wildlife	Domestic cat	<i>Felis silvestris catus</i>
Non-Native Wildlife	Domestic dog	<i>Canis lupus familiaris</i>
Non-Native Wildlife	Human	<i>Homo sapiens</i>

**Table 2. The Habitat Use of Wildlife at Haines Branch by Habitat Type.** Table 2a. Total number of mammal species captured on the camera traps at Haines Branch by habitat type.

<b>Species</b> <b>Habitat Type</b>	White Tailed Deer	Red Fox	Raccoon	Rabbit	Skunk	Squirrel	Coyote	Opossum	Groundhog
Grassland	577	127	43	23	1	8	2	2	2
Forest	286	90	69	66	1	27	0	9	4
Wetland	431	69	49	8	0	0	1	0	0
Former Agriculture	4	0	0	0	0	0	0	0	0

Table 2b. Capture frequency of mammals captured on the camera traps at Haines Branch by habitat type.

<b>Species ----- Habitat Type</b>	White Tailed Deer	Red Fox	Raccoon	Rabbit	Skunk	Squirrel	Coyote	Opossum	Groundhog
Grassland	124.9	27.5	9.3	5	0.2	1.7	0.4	0.4	0.7
Forest	113.4	35.8	27.4	26.2	0.4	10.7	0	3.6	1.2
Wetland	205.2	32.8	23.3	3.8	0	0	0.5	0	0
Former Agriculture	0	0	0	0	0	0	0	0	0

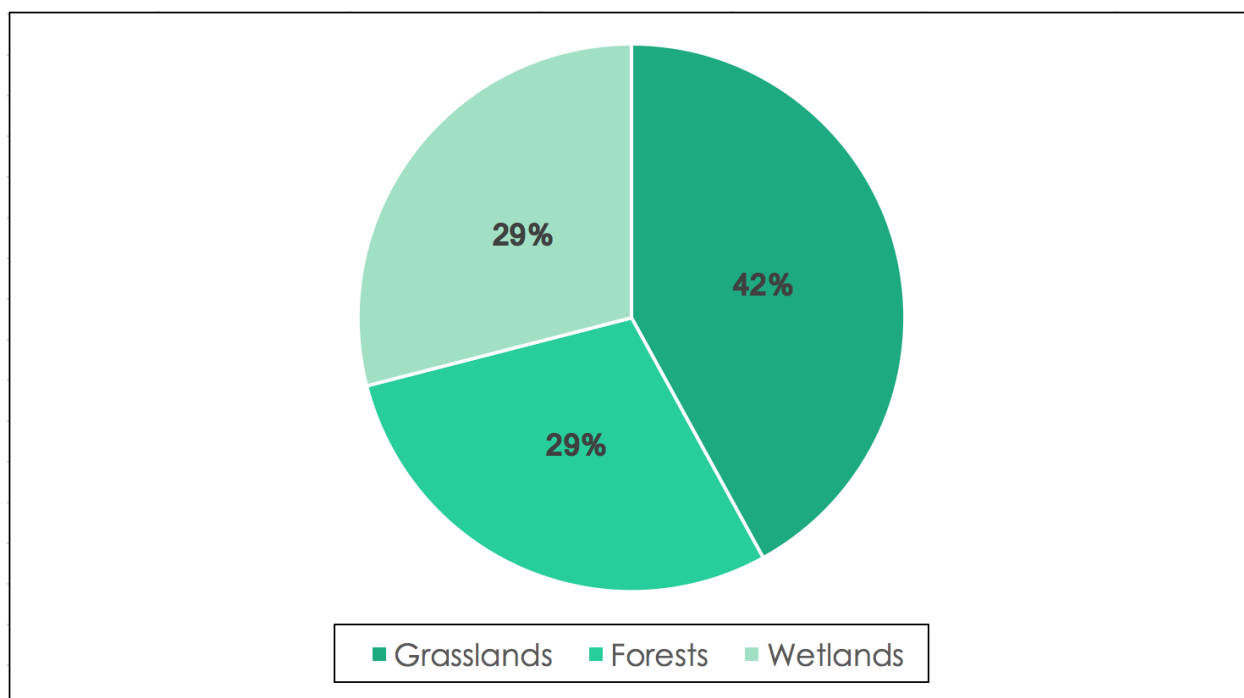
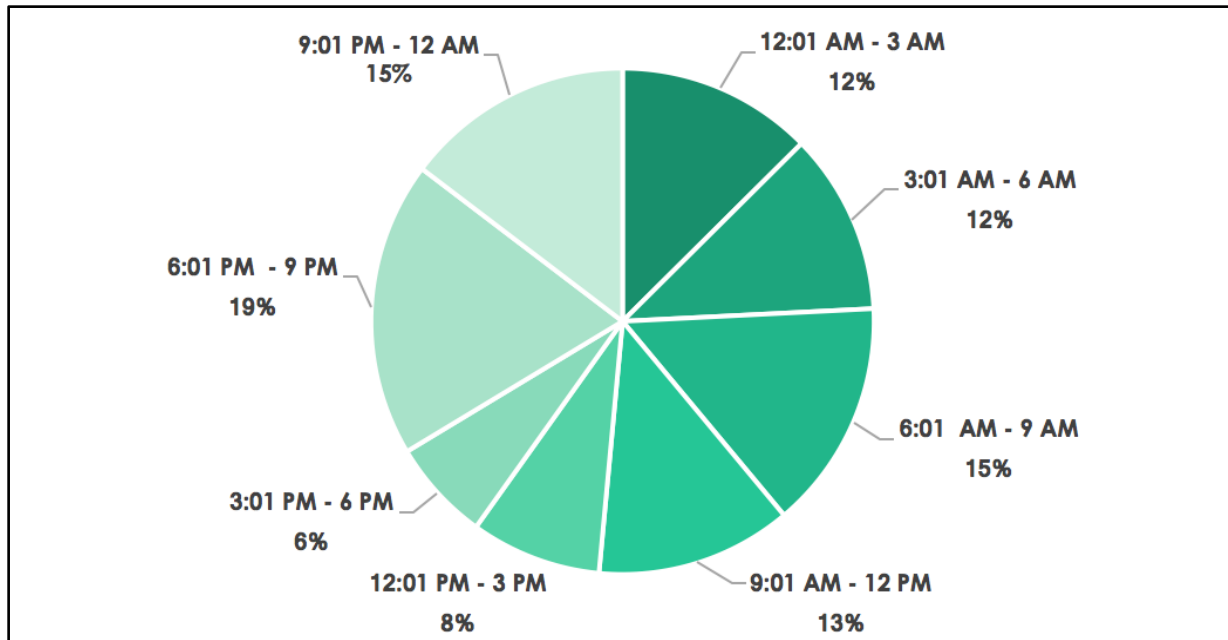
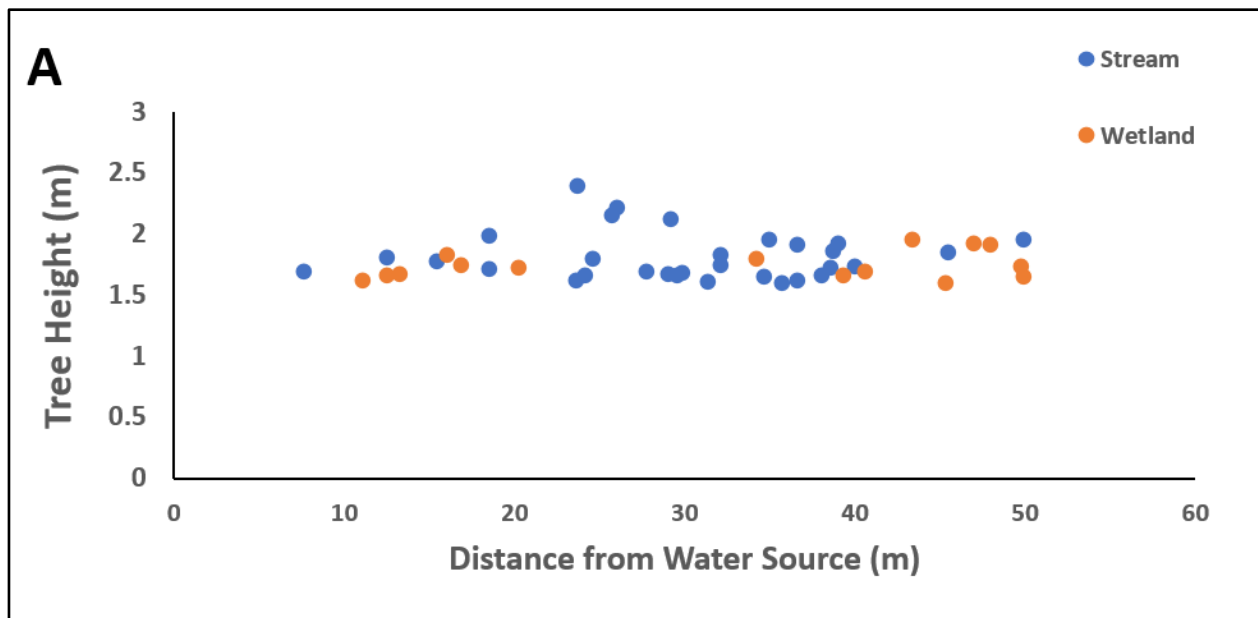


Figure 1. Percent total of mammal species captured in each habitat type at Haines Branch.



**Figure 2. Activity Patterns of Foxes at Haines Branch.** *The figure shows the recorded activity patterns of red foxes in three hour time blocks as a percentage of all red foxes captured.*



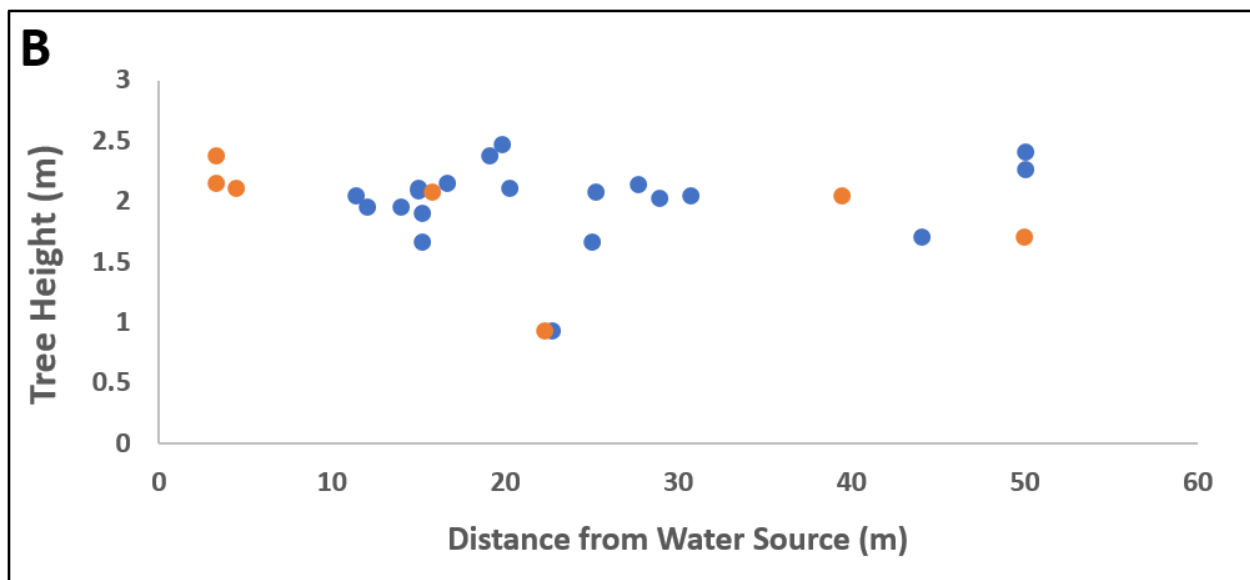


Figure 3. The Relationship between Tree Height and Distance to the Nearest Body of Water for (A) *Juniperus virginiana* and (B) *Quercus alba*.

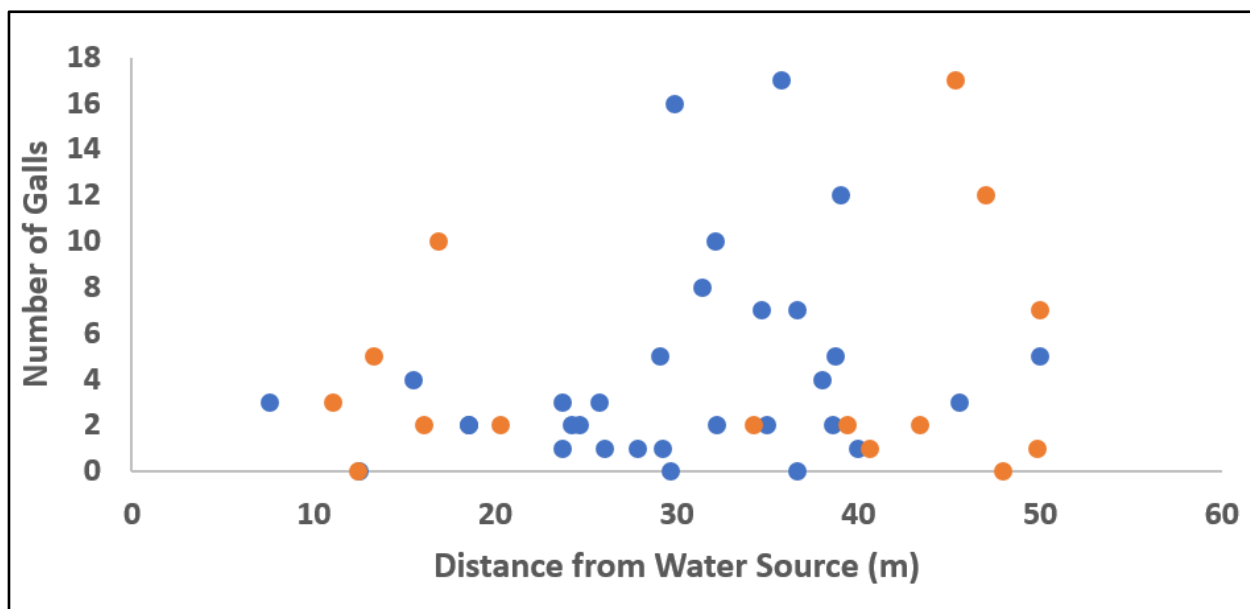
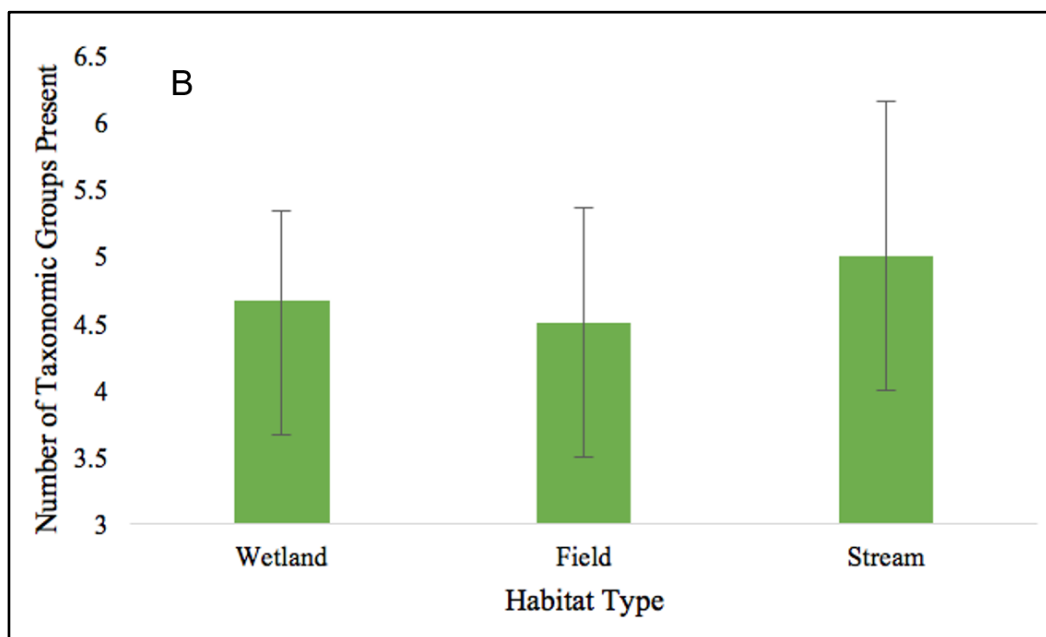
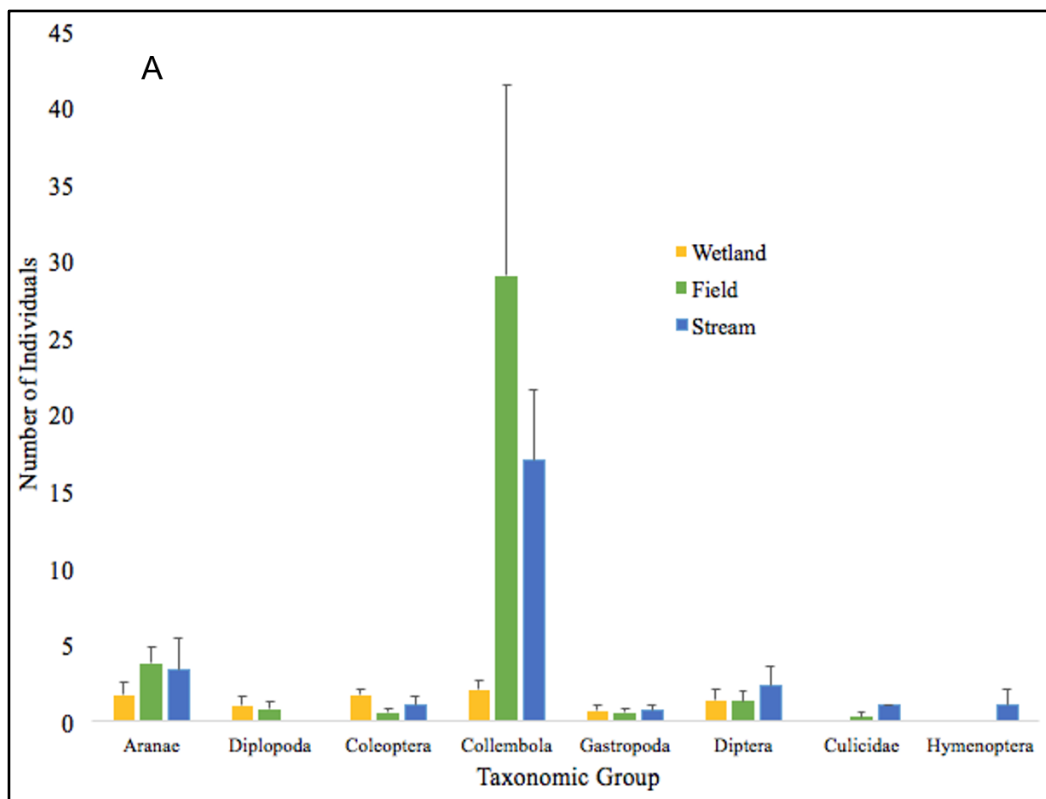
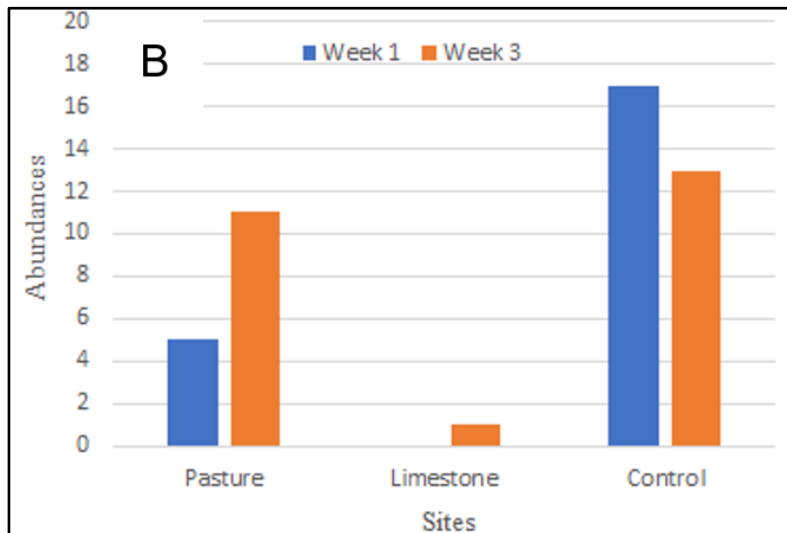
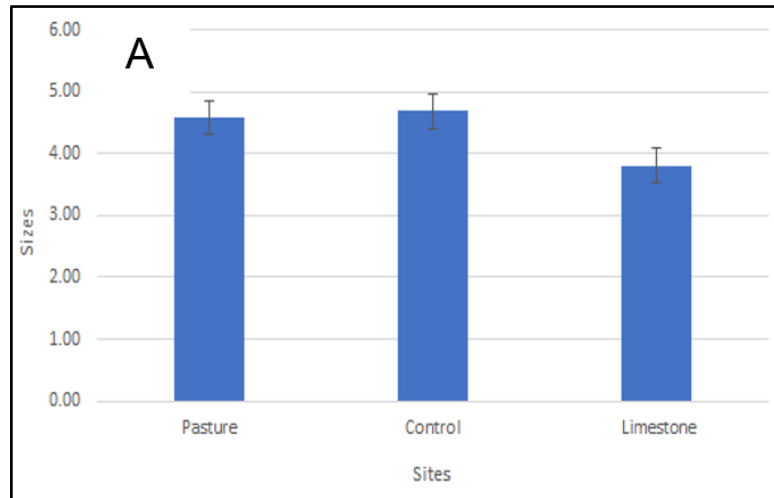


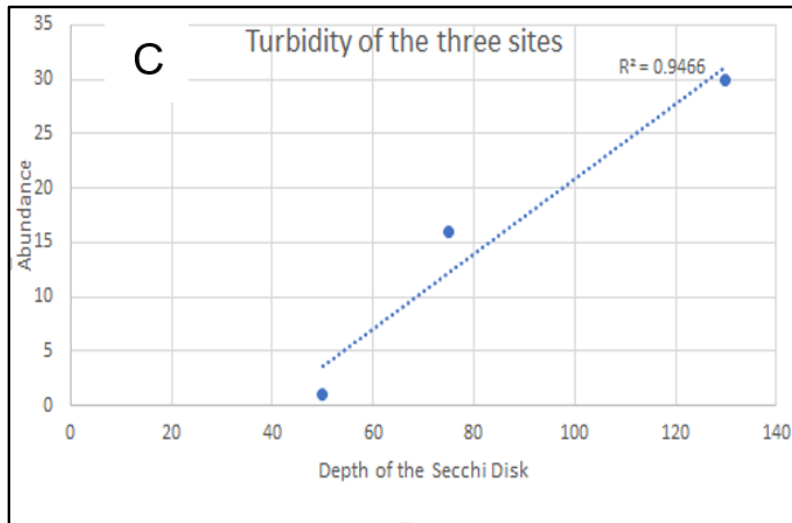
Figure 4. Number of Galls on Individuals of *J. virginiana* with Respect to Distance to the Nearest Body of Water.



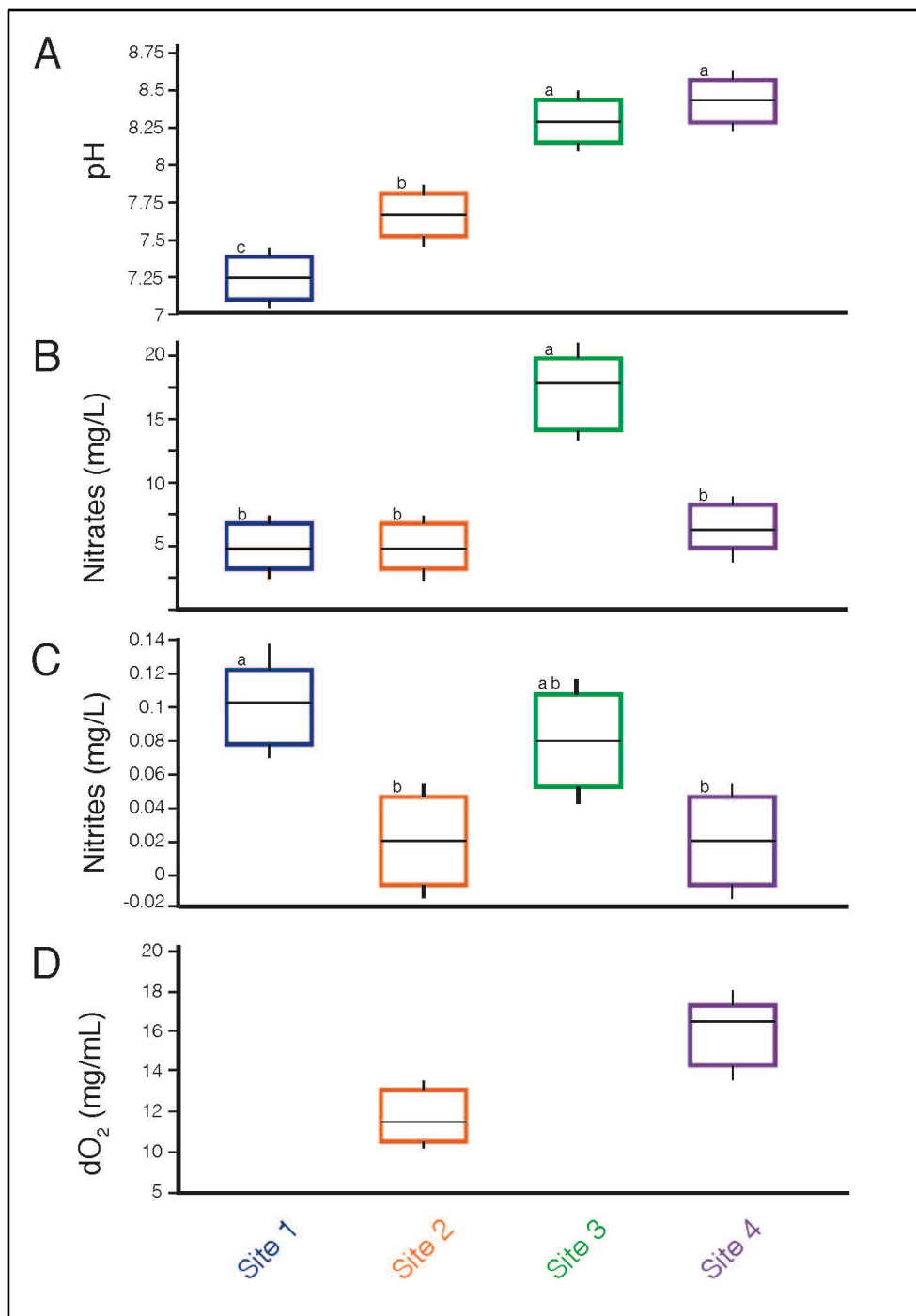


**Figure 5. The Abundance (A) and Taxonomic Richness (B) of Invertebrates Captured in Pitfall Traps 4-11.** Plotted are means ( $\pm 1$  SE). Samples were collected in wetlands (adjacent to submerged regions of seasonal wetlands), fields (grasslands), and stream (adjacent to stream channel); pitfalls in forest sites were lost to trampling. Taxonomic richness was assessed as the total number of groups (classes and orders) depicted in part A for each pitfall trap.





**Figure 6. Crayfish Size (A), Abundance (B), and Relationship of Total Crayfish Abundance with Turbidity (C) at Three Sites:** Pasture (corresponding to Site 1 in Appendix B), Limestone (Site 2), and Control (Site 4). Crayfish were collected for 45 minutes during each of two intervals. Size is measured in cm. Turbidity of the water is the inverse of the depth of the Secchi Disk; as the disk is lowered, the ability to resolve contrasting colors on the disk at greater depth is facilitated by clear (non-turbid) water.



**Figure 7: Water Chemistry Differs Significantly by Location Along the Stream.** (A): Water pH was highest upstream (Site 4; adjacent to forest) and was lowest downstream (Site 1; adjacent to dairy farm) (B): Nitrate levels were significantly higher at site 3. (C): Nitrite levels were highest at site 1, but site 3 was not significantly different than site 1. (D): Dissolved oxygen was significantly higher at site 4 than at site 2. Boxes with different letters signify statistically significant differences among sites.

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