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Assessing the feasibility of freshwater mussel restoration in urban streams.

John-Reid Ryan

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By John-Reid Ryan, B.S. Environmental Studies Virginia Commonwealth University
Center for Environmental Studies, Virginia Commonwealth University,
ryanjr2@mymyail.vcu.edu

Thesis Advisor: Paul Bukaveckas Ph.D. Professor, VCU Center for Environmental Studies

Virginia Commonwealth University
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Abstract

The main objective of this study was to determine whether introduced freshwater mussels (Alewife floater, Utterbackiana implicata) can survive and grow in urban streams in the James River watershed. A secondary objective was to assess differences in U. implicata survival and growth of in the context of differing water quality and food resource conditions among three urban sites and three rural sites. Results from this study show large differences in growth and survivorship of mussels across sites. Higher survivorship was observed among mussels stocked into rural streams (35% and 44%) in comparison to urban streams (3%, 6% and 14%). High mortality in urban streams was largely due to washout and burial of mussels. These findings suggest that the "flashy" hydrology typically associated with urban streams is a significant impediment to successful introduction at these sites. High growth rates were observed in one of the rural streams (Herring Creek: 57 mg/d), whereas growth rates were less than 15 mg/d at all other sites. Food resource metrics showed statistically significant differences among sites with higher values of TSS, particle density, organic matter content and chlorophyll-a content at rural sites relative to urban sites. These findings suggest that rural sites had more favorable food resources than rural streams, though we did not find that food metrics were a significant predictor of variation in growth rates among sites. We did not find that water quality metrics (temperature, dissolved oxygen) were a significant predictor of variation in mussel growth rates. Overall, these findings suggest that hydrologic conditions in urban streams pose a significant challenge to the successful reintroduction on native mussels.

Introduction

North America has the largest freshwater mussel diversity in the world (Williams et al., 1993). In addition to being a diverse and unique fauna, mussels provide important ecosystem services by removing particulate matter from the water column (Vaughn, 2018). Nutrients contained in particulate matter may be stored in mussel shells and tissues, or excreted as 'bio deposits' (Vaughn, 2018). The transfer of nutrients from suspended particulate matter to benthic deposits makes mussels an important link within nitrogen and phosphorous cycles as it increases the likelihood that nutrients will be sequestered (through in-stream burial) or lost via denitrification (Hoellein et al., 2017). Prior work on urban rivers showed that freshwater mussels stimulate microbial activity and denitrification through waste production. Hoellein et al. (2017) showed that nitrogen uptake and denitrification rates in sediment alone were around 2%, while rates in sediments with mussels were 8-12%. Ecosystem services provided by freshwater mussels may be beneficial to mitigating anthropogenic pollution of Chesapeake Bay. Human activities such as agriculture and urban development increase sediment and nutrient transport via tributary streams transporting sediment to the Bay (Eshleman & Sabo, 2016). These non-point sources, coupled with point source inputs such as wastewater treatment plants have degraded water quality within the Bay and its tributaries (Eshleman & Sabo, 2016). Reducing sediment and nutrient loads to the Bay to improve water clarity will depend on implementation of best management practices in upland areas and tributary streams (McConnell, 2017).

Mussel populations have declined in many watersheds throughout the United States over the past 50 years (Williams et al., 1993). The Nature Conservancy estimates that 55% of mussel species in North America have progressed to extinction or imperiled status (Williams et al., 1993). Declines in mussel populations are partly due to habitat degradation associated with land use change (urbanization and agriculture). Urban streams are often devoid of mussel populations, and in unrestored urban streams their biodiversity can be 47% less than reference streams (Smucker & Detenbeck, 2014). Recent advances in the ability to propagate mussels provides an opportunity to restore populations, however little is known regarding in-stream conditions that would influence the success of restoration (e.g. food and water quality conditions). Generally, mussels require well-oxygenated flowing water with suitable substrate conditions (a mixture of sand, gravel and silt; NRCS, 2007). Food quantity and quality is likely to be dependent on the abundance of suspended particulate matter and its composition, including contributions from suspended algae (Jeager and Cherry, 1994). The presence of impoundments, such as storm water retention ponds, may increase food quantity and quality as they trap inorganic particulate matter, and may promote growth of phytoplankton (Winter and Duthie 1998). Further research is needed to document the success of mussel introduction efforts in diverse stream habitat conditions to better inform management efforts.

Conditions found in urban streams may present special challenges to mussel restoration (Walsh et al., 2016). These include “flashy” streamflow conditions due to rapid runoff from impervious surfaces (Nedeau et al.,2003; Walsh et al., 2016). High discharge events alter stream morphology due to increased bank erosion which causes unstable sediments and burial of mussels (Walsh et al., 2016). Urban streams are also subject to scouring events where the stream bottom is removed (Walsh et al., 2016). This removes substrates including leaf litter and organic deposits that make up mussel habitat (Walsh et al., 2016). The absence of mussels in urban streams may also be from years of poor water quality conditions (low dissolved oxygen, toxic pollutants) that were prevalent prior to passage of the Clean Water Act.

The Chesapeake Bay has suffered from eutrophication and sediment pollution for a number of decades. In order to properly manage this issue, there is a need for restoration practices that reduce sediment and nutrient loads. This is accomplished by implementing best management practices (BMPs). Improvements in stream condition have been brought about by a variety of management practices that seek to reduce urban runoff (e.g., via storm water retention), improve water quality (e.g., by preventing CSO events), and, in some cases by undertaking stream restoration projects, which reshape the stream channel to reduce erosion and withstand “flashy” hydrology (NRCS, 2007). Typically, stream restoration projects focus on the geomorphology of the channel and do not consider the potential for biological restoration as a means to improve ecosystem services (NRCS, 2007). In contrast, biological restoration is used within the Bay itself to achieve water quality targets. For example, the Chesapeake Oyster BMP (Cornwell et al., 2016) was established on the basis that oysters filter particulate matter from the water column and increase nitrogen removal through enhanced denitrification (Cornwell et al., 2016). Research from the Partnership for Delaware Estuary (PDE) has shown that the clearance and filtration rates of freshwater mussels rival that of oysters. Kreeger et al. (2017) reported that mass-specific clearance rates for freshwater mussels ranged from 0.5 to 3.4 $l\ h^{-1}\ g^{-1}$, while Eastern Oysters had a clearance rate of 6 to 6.4 $l\ h^{-1}\ g^{-1}$. The same study suggested that mussel beds have a higher clearance rate than oyster beds, due to higher population density. The critical ecosystem services that oysters contribute to the environment are potentially the same ecosystem services that freshwater mussels could provide in upstream areas. These findings suggest that stocking freshwater mussels in tributary streams may be a useful means for reducing nutrient and sediment inputs to the Bay.

Mussel restoration efforts have occurred throughout the Atlantic slope, though these typically focus on species that are a priority for conservation, and in high quality habitats. In addition to state and federal facilities, some environmental groups have also begun mussel restoration activities to augment the population of Alewife Floater, Utterbackia implicata, mussels (Delaware Estuary, 2016) . The National Strategy for Conserving Freshwater Mussels enumerates several goals for propagating mussels and understanding what factors degrade the population (Haag & Williams, 2014). However, the strategy provides little guidance as to where the restorations should occur (Haag & Williams, 2014). Many stocking programs focus only on augmenting population, and only in areas where other members of the same species can be found. As a result, propagation programs leave out streams where mussels are or were historically absent, and which may benefit from mussel restoration to improve local and downstream water quality. Data are needed to assess the viability of stocking mussels into impaired streams such as those found in urban environments and to better understand the factors which affect their performance (e.g., survivorship, growth rate).

Objectives and Hypothesis

My thesis project focused on the question: Can freshwater mussels be restored in urban streams? To address this question, *A.implicata* were stocked in three urban streams in the metro area of Richmond, Virginia. As a control for comparison, *A. implicata* were stocked in two rural streams and a hatchery pond. All of the streams used in this study are tributaries of the

James River. One of the rural streams (Herring Creek) is also the source of water for the fish hatchery pond. Data were collected to assess mussel survivorship and growth (mass and length) at 1-2 month intervals. The primary objective was to determine whether freshwater mussels can survive and grow in urban streams. A secondary objective was to assess inter-stream differences in survivorship and growth in relation to in-stream habitat conditions such as water quality, food quantity, and food quality. Water quality metrics of interest included water temperature, pH and dissolved oxygen. In urban streams, low pH and oxygen conditions may arise from chronic or episodic inputs of wastewater (e.g., CSO events), which may have detrimental effects on mussel growth and survivorship. Temperature would generally be expected to have a positive effect on growth rates, though high temperature conditions (e.g., in the absence of a riparian canopy) may be detrimental. Metrics of food quantity such as Total Suspended Solids (TSS) or particle density, may be positively related to growth and survivorship, though in urban streams, bed and bank erosion may suspended materials of low food quality (e.g., sand, silt and clay). Therefore, measures of food quality (organic matter and chlorophyll-a content) were also used to assess their relationship to growth rates.

Methods

Juvenile *A. implicata* mussels (mean length 42mm STD dependent on site) were placed in cages installed in each of three urban streams and two non-urban streams. Alewife Floater mussels were chosen due to their availability from Harrison Lake National Fish Hatchery (Charles City, VA), because they are native to the region and have been used in prior restoration projects (Kreeger et al., 2017). The mussels were individually tagged and monitored on a monthly basis for survivorship and growth. Water quality, substrate characteristics and food quality were documented for each stream to interpret differences in survivorship and growth. The same data were collected from control sites: a rearing pond at the Harrison Lake Fish Hatchery, and two non-urban streams (Kimages Creek, Herring Creek). The Hatchery pond was chosen as a control site as it has been used to successfully rear mussels for local stocking efforts. Herring Creek and Kimages Creek are located nearby and were chosen based on their rural location and access. Kimages Creek is located at the VCU Rice Rivers Center and has a long-term record of bi-monthly water quality monitoring.

The Richmond urban streams selected for this study were part of a network of 7 sites currently monitored by VCU. Bi-monthly data are collected to measure discharge and water quality (temperature, pH, conductivity, turbidity, dissolved oxygen and TSS). Individual streams were chosen based on their accessibility and prior data characterizing hydrology, geomorphology and water quality. Habitat conditions (hydrology) and food availability (quantity and quality of particulate matter) were expected to differ among the study sites. Broad Rock Creek, Reedy Creek and Gillies Creek drain predominantly urban areas with a high proportion of impervious surfaces and low proportion of forested areas in their watershed (Table 1). At Broad Rock, the presence of an upstream impoundment was expected to provide more stable flow conditions, and potentially improve food quality by trapping sediment and allowing for phytoplankton production that could have possibly imitated impoundments at Herring Creek. It was expected from land use data that rural streams would harbor improved mussel survival and growth rates, but the rise in impervious surfaces at Kimages creek could show variation in mussel survivorship and growth between rural sites.

In-Stream Deployment of Mussels

Juvenile A. implicata mussels were stocked at all sites in April-May 2019 with stocking rates ranging from 106 to 129 per site. Supplemental stockings (20-60 mussels) were carried out at some sites following loss of individuals due to washout. A total of 880 mussels were used in this study. Six enclosures (Figure 1) containing ~19 individuals per enclosure were placed at each site. Stream enclosures were constructed from plastic crates (approx. 25 x 25 x 25 cm) with plastic wire mesh (1 cm) added along the sides (2 layers), bottom (3 layers) and top (1 layer). Two metal poles with flat tops commonly used to secure mulch barriers were placed at the front two corners to secure the enclosure to the stream bottom. Mussel enclosures at the fish hatchery site consisted of floating baskets (Patterson et al. 2018). Three enclosures containing 40 mussels per basket were placed into a pond at the Harrison Lake National Fish Hatchery. All mussels were given a unique ID tag (number 1 to 1,300) on their shell below the umbo using a Zing laser engraver.

Data Collection

Survivorship and growth were monitored at 1-2 month intervals from the time of stocking (April-May 2019) to the conclusion of the experiment (December 2019). Survivorship was determined from the number of alive mussels in all enclosures at a site for a given census date. Separate tallies were recorded for dead mussels (recovery of empty shells) vs. lost mussels (arising from loss of cages or missing mussels within recovered cages). Due to loss of enclosures, and in some cases, the low number of surviving individuals per enclosure, we did not derive separate estimates of survival for each enclosure. Length (mm) and weight (wet mass; g) were measured for all individuals. Growth rates (mm/d and mg/d) were derived for each monitoring interval based on repeat measurements of tagged individuals. In the later stages of the experiment, some tags became unreadable. We recorded length and weight for these unknowns, and derived population-based estimates of growth rate based on the change in mean length and mass during the monitoring interval. Water quality data (temperature, pH, conductivity, turbidity and dissolved oxygen) were measured in conjunction with monitoring of mussels using a YSI Pro DSS sonde. Water samples were collected to assess food quantity and quality based on total suspended solids (TSS), organic matter (OM) content, and chlorophyll-a (CHLa) content. Sample analysis followed protocols developed for the VCU Environmental Analysis Lab, a state-accredited water quality testing facility. Samples for TSS and CHLa were filtered through a GF/A glass fiber filters (0.5-µm nominal pore size). Filters for CHLa analyses were extracted for 18 h in buffered acetone and analyzed on a Turner Design TD-700 Fluorometer. Filters for TSS were dried at 60^{o} C for 48 h and analyzed using a Perkin-Elmer CHN Analyzer to determine the organic matter content, expressed as particulate organic C (POC). Particle size and density were measured between 2.16 and 60 µm using a Coulter Counter Multisizer4e (Beckman Coulter, Pasadena, California). Samples were preserved with Lugol's iodine solution and refrigerated. Samples were diluted with electrolyte solution starting with 5ml of sample to 5ml of electrolyte and repeated three times for a range of concentrations (2x, 4x, 8x, and 16x). The Coulter Counter measures all particles within the specified size range inclusive of cells (bacteria, phytoplankton) and non-living particulates (e.g., silt, clay, etc.) with results reported as number of particles per unit volume.

Statistical Analysis

Water quality data and metrics of food quantity and quality were analyzed using a one-way ANOVA to determine whether differences among sites were statistically significant. Statistical analysis was limited to the 4 sites for which bi-monthly data were available; Reedy Creek, Gillies Creek, Broad Rock Creek and Kimages Creek.

Results

Survivorship of mussels varied among the 6 sites (Figure 2). Highest survivorship was observed at the fish hatchery pond where 88% of the stocked mussels survived through the end of the experiment (May-November). Only a small number (N=3) were lost due to mortality (presence of dead mussels), while others were unaccounted for and designated as lost. Survivorship at the rural sites (Kimages and Herring Creeks) was 35% and 44%, respectively. At these sites, relatively few mussels were lost due to mortality (Kimages = 21 individuals; Herring = 13 individuals), but a larger number of individuals (40-60 mussels per site) were lost due to washout of cages following a July storm event. Lowest survivorship was observed among the three urban sites. At Reedy Creek, only 4 mussels survived through mid-August (3% survivorship), while at Broad Rock and Gillies Creek, the number of surviving mussels was 12 and 20 individuals (6% and 14% survivorship, respectively). At the urban sites, the majority of the decline (84%) was due to loss of cages either through washout or burial, though higher rates of mortality were also observed (Reedy = 42 individuals, Gillies = 28 individuals).

Length-based growth rates varied seasonally and among sites (Table 2; Figure 3). Highest individual-based growth rates (0.23 ± 0.06 mm/d) were measured in Herring Creek during July to September. Average growth rates at this site were 0.093 mm/d across all census periods. By comparison, growth rates at the hatchery pond and Kimages Creek were 0.047 mm/d and 0.044 mm/d, respectively. The high growth rates at Herring Creek correspond to an increase in length of 31% (from 45.4 ± 0.3 mm to 59.5 ± 0.8 mm) over the 5-month period (May-October). Mussels stocked at the hatchery pond and Kimages Creek increased in length by 10% and 9%, (respectively) over the same period. The low number of surviving individuals limited the number of dates for which growth rates could be calculated at the urban sites. Average growth rates were lower among the urban streams in comparison to the non-urban sites: 0.025 mm/d (Gillies), 0.021 mm/d (Broad Rock) and 0.015 mm/d (Reedy).

Highest mass-based growth rates were observed in Herring Creek where mean growth rates were 57 mg/d and peak values were 94 ± 9 mg/d from July to September (Table 3; Figure 3). High mean growth rates were also observed during September-October at this site (48 ± 14 mg/d) and at the hatchery pond (40 ± 2 mg/d) during May-July. Among other sites, average growth rates ranged from 1 mg/d at Broad Rock to 15 mg/d at the hatchery pond and Reedy Creek. Negative growth rates were recorded in some census periods.

Growth rates estimated using the larger dataset of all measured individuals (including those that could not be identified by tag number) generally showed good agreement with those based on repeated measurements of tagged individuals (Figure 4). Mass-based growth rates, whether derived from repeat measurements of individuals or population mean values showed a high degree of correspondence ($R^2 = 0.96$, $p < 0.001$). Length-based estimates derived by the two methods showed a weaker agreement ($R^2 = 0.52$, $p < 0.001$). Increases in length showed a strong correspondence to increases in mass for both the individual-based ($R^2 = 0.65$, $p<0.001$) and population-based assessments ($R^2 = 0.72$, $p<0.001$). Slopes derived from the two datasets were not statistically different (0.0017 ± 0.0003 and 0.0016 ± 0.0002 mm/mg, respectively).

Water quality conditions varied seasonally and among sites (Table 4; Figure 5). Statistical analysis of these data focused on the 4 sites for which bi-weekly data were available (N = 17 measurements during May-December). Water temperatures were similar among the 3 urban and one non-urban (Kimages) streams (mean range = 16.4 to 18.0 C; $p = 0.8$). Peak water temperatures during the period of study were ~22 C at these sites. Higher water temperatures were observed in the hatchery pond, which exceeded 25 C during June-September (peak = 32 C). Specific conductance was significantly different among the 4 sites ($p < 0.001$) with lowest values at Kimages Creek (mean = 104 ± 11 uS/cm) and higher values among urban streams (mean range = 144 to 197 uS/cm). Similar conductivity values were observed in Herring Creek (mean = 150 ± 41 uS/cm), whereas the hatchery pond exhibited low values relative to the streams (mean = 38 ± 5 uS/cm). Dissolved oxygen concentrations differed significantly among sites ($p < 0.001$) with lowest values observed in Kimages Creek (mean = 6.7 ± 0.3 mg/L; saturation = 70 ± 3%). Dissolved oxygen values were higher among the three urban streams (range of means = 9.0 to 9.6 mg/L; saturation = 91-100%). Low oxygen conditions were also observed in Herring Creek (mean = 5.8 ± 1.4 mg/L; saturation = 59 ± 12%) and on one occasion in the hatchery pond (June = 2.2 mg/L; 27% saturation). Highest discharge was observed in Herring Creek (mean = 233 ± 114 L/s). Among the urban sites, Gillies Creek exhibited the highest average discharge (mean = 74 ± 4 L/s) and highest peak values >100 L/s in June and October). Average discharge was similar among Broad Rock, Reedy and Kimages Creeks (range of means = 14 to 39 L/s).

Food quantity and quality varied between sites (Figure 6). TSS concentrations (mg/L) were significantly different among the 4 sites for which bi-monthly data were available ($p < 0.001$). Highest concentrations were measured in Kimages Creek (mean = 16.2 ± 5.2 mg/L). TSS concentrations were lower among the three urban streams (range of means = 1.7 to 2.2 mg/L). Relatively few measurements of TSS were obtained from the hatchery pond and Herring Creek (N = 3 and 4, respectively), but these showed somewhat elevated levels relative to the urban streams (mean = 8.7 and 3.8 mg/L, respectively).

Variation in CHLa concentrations among sites generally tracked differences in TSS. Highest concentrations were observed at the hatchery pond (mean = 5.65 µg/L ± 1.16). Average concentrations at Herring and Kimages Creek were 2.68 ± 0.63 and 2.46 µg/L ± 0.58, respectively). Urban sites had lower CHLa concentrations relative to rural sites with mean values ranging from 0.76 ± 0.12 µg/L (Broad Rock) to 1.91 ± 0.29 µg/L (Reedy). Differences in CHLa among sites were statistically significant ($p < 0.001$). POC concentrations generally followed TSS and CHLa showing lower concentrations at the urban sites. Kimages Creek and the hatchery pond exhibited the highest POC concentrations (means = 1.16 ± 0.22 and 1.08 ± 0.11 mg/L, respectively), followed by Herring Creek (0.66 ± 0.08 mg/L). Urban streams had two-fold lower POC concentrations compared to the rural sites with mean values ranging from 0.22 ± .03 (Gillies) to 0.22 ± 0.04 mg/L (Broad Rock) to 0.31 ± 0.03 mg/L (Reedy). Differences in turbidity among sites mirrored variations in TSS, CHLa, and POC. Kimages Creek and the hatchery pond had the highest average concentrations of all sites (14.1 ± 3.8 and 10.2 ± 1.4 NTU, respectively). There was no consistent seasonal pattern in CHLa, TSS, Turbidity, or POC concentrations among the sites for which bi-monthly data were available.

Particle density and particle size varied between sites. Higher particle densities were observed among the rural sites relative to the urban sites. The hatchery pond exhibited the highest particle density (mean = 158,000 ± 40,000 #/ml), though this estimate was based on only 4 measurements. Among the 4 sites for which bi-monthly data were collected, Kimages Creek exhibited the highest particle density (mean = 91,400 ± 3,000 #/ml). Average particle densities in Herring Creek were 53,1000 ± 4,000 #/ml. Urban sites had less than half the particle density of rural sites (Gillies = 31,700 ± 3000, Broad Rock = 29,400 ± 7600, Reedy 34,900 ± 7500 #/ml). Differences among the 4 sites where bi-monthly data were available were marginally significant ($p = 0.052$). Median particle size was highest at Broad Rock (mean = 3.27 µm) but was otherwise similar among sites (range = 2.89 to 2.98 µm; $p = 0.006$).

Seasonal and inter-stream variation in mussel growth rates was analyzed in relation to water quality and food resource metrics (Figure 7). Water quality metrics generally did not show a statistically significant relationship with mussel growth rates, with the exception of dissolved oxygen. Growth rates were generally similar among sites where oxygen saturation was greater than 60%, but two of the sites where high growth rates were measured (Herring Creek and hatchery pond) exhibited low dissolved oxygen. This resulted in a weak ($R^2 = 0.25$ but statistically significant ($p = 0.029$) negative relationship between growth and dissolved oxygen. Food metrics did not show statistically significant relationships with mussel growth rates with the exception of a marginally significant ($p = 0.07$) positive relationship with CHLa ($R^2 = 0.23$).

Discussion

Differences in survivorship among urban and rural sites reflected expectations based on the urban stream syndrome. Specifically, issues of “flashy” hydrology and sedimentation were evident at urban sites (Walsh et al., 2016). Multiple cage losses occurred at each of the urban sites (e.g., 3 at Gillies, 4 at Broad Rock and 6 at Reedy Creeks). Most of these cage losses occurred from late May through June. Though we lack site-specific discharge data, it is likely that high runoff from impervious surfaces resulted in large increases in stream discharge following rainfall events, which resulted in enclosures being swept away. One of the study sites (Reedy Creek) has a 3-year record of water level data (2015-2018), which was used in combination with twice-monthly measurements of discharge to develop a stage-discharge relationship (P. Bukaveckas, unpubl. data). At baseflow, water level was 10-20 cm and discharge less than 10 L/s. During events, water level exceeded 2 m and discharge exceeded 3000 L/s. The large range of variation in depth and discharge highlight the difficulties in establishing resident populations of mussels. Sediment deposition from bank erosion was also evident at each of these sites, resulting in silt and sand loads in cages that may have suffocated mussels. Prior studies in urban systems using the Asian Clam (*Corbicula*) also reported high rates of (Nobles & Zhang, 2015), whereas enclosures in non-urban systems reported higher rates of survivorship (e.g., 60-90%; Haag et al., 2019), which were comparable to those observed at our rural sites. Mortality effects measured in this study reflect in part the use of enclosures, which are needed to recover individuals for measurements of growth and survivorship. Loss of cages is ascribed to mortality, though it is possible that mussels stocked directly into streams may be better able to avoid washout, or to find suitable habitat in downstream areas. Some loss of mussels may have also occurred due to escape from cages, as indicated by the absence of mussels and lack of dead shells, but this was minor component of the overall mortality in comparison to washout losses.

Alewife Floater mussels were selected for this study in part due to their expected high growth rates. Observed growth rates based on length ranged from 0.04 to 0.23 mm/d, exceeding those reported for other mussels (e.g., *Lampsilis cardium = 0*.001 - 0.020 mm/d; Ohlman & Pegg, 2020). Highest growth rates were observed at Herring Creek and the hatchery pond. For comparison Alewife floater mussels have a fast growth rate in hatchery systems capable of 0.219 mm/d (Kreeger et al., 2018). It is possible that growth rate could have been limited by a concentrated population size of 40 per floating basket as opposed to 20 per cage. It is possible that the concentration of 40 per basket was at a threshold where mussels were competing with each other for feeding and physically limited due to lack of space within the basket. If this were true, it might would explain why the growth rate dropped from 0.10 to 0.05 mm/d. Growth rates in the urban streams and Kimages Creek were generally below Herring Creek and the hatchery pond, with only a few peaks reaching equal growth. Inter-site differences in mass-based growth rates generally followed survival rates, showing that urban streams had lower growth rates and higher mortality compared to rural streams. While

Kimages Creek does not have all the attributes of the urban sites such as flashy hydrology or a watershed with high amounts of impervious surfaces, the growth rate and survivorship of mussels is more comparable to urban sites than rural sites.

Trends in food quality and quantity indicate that differences between sites may partly account for differences in survival and growth. The response of freshwater mussels to increased algal or suspended matter flux with water velocity is complex and varies by species due to morphology (Mistry & Ackerman, 2015.; Tuttle-raycraft & Ackerman, 2019). High concentrations of suspended particulate matter are indicative of potentially greater food resources, though much of this particulate matter may be of low quality (e.g., silt and clay). Studies have shown that the clearance rate (number of particles filtered) of mussels decreases with high levels of TSS (Tuttle-raycraft & Ackerman, 2019). It is therefore important to also consider metrics of food quality such as organic matter content and chlorophyll a content. Rural sites exhibited higher TSS, POC, chlorophyll-a and particle density relative to urban sites. More favorable food resource conditions may account for higher growth rates observed at some of the non-urban sites (Herring Creek, Hatchery pond) though we did not find significant relationships for these, or water quality parameters, in predicting variation in growth rates.

Conclusion

Prior studies have documented the effects of urbanization on stream hydrology and geomorphology. Typically, these are associated with high runoff from impervious surfaces during storm events, which lead to bed and bank erosion. These conditions are often associated with impairment of stream function (e.g., reduced sediment and nutrient retention; low biodiversity). Results from this study further highlight the challenges to restoring urban streams as exemplified by the high rate of washout and burial of introduced mussels. Restoration efforts within the catchment are needed to reduce urban runoff, which may then allow for successful re-introduction of mussels and associated improvements in stream ecosystem services. Data from this study show that while introduction of mussels to urban streams had limited success, growth and survivorship in nearby rural streams was indicative of greater potential for success. The stocking of freshwater mussels into these, and potentially, restored urban streams, may be a useful approach to mitigating nutrient and sediment transport to Chesapeake Bay. Further studies are needed to better understand water quality and food conditions that are conducive to successful establishment of mussels, and how stream restoration efforts may be geared to accommodating mussel introduction.

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Table 1. Location, watershed area, substrate composition, and surrounding land use from NOAA land use database and graduate student thesis (Lucas, 2019) (NOAA, 2010) of five streams selected for mussel introduction.

Site	Watershed Area (sq km)	Substrate	Impervious	Forest	Lat/Long
Broad Rock	7	Cobble	28%	16%	37.487301,-77.441020
Gillies	36.8	Cobble-Clay-Sand	28%	16%	37.527887,-77.393051
Reedy	9.8	Cobble-Silt	33%	35%	37.514287,-77.473190
Kimages	12.7	Clay-Silt	8%	50%	37.338097,-77.205687
Herring	78	Pebble-Sand	0%	63%	37.334301,-77.185604

Table 2. Growth rates of mussels (length in mm/d) stocked into urban and non-urban streams. Individual growth rates are based on repeat measurements of the same individual; population growth rates are derived from the average length of all individuals. Stocking dates for each site are shown in the first column and census dates in the second column.

		Individual GR (mm/d)			Population GR (mm/d)	
Site	Date	Mean	SE	N	Mean	N
Hatchery	7/15/2019	0.072	0.003	116	0.071	119
5/15/2019	8/27/2019	0.022	0.006	99	0.007	112
	10/2/2019	0.040	0.027	92	-0.002	104
	11/2/2019	0.052	0.027	86	0.015	100
Kimages	7/24/2019	0.031	0.003	45	0.044	65
5/18/2019	9/1/2019	0.071	0.043	39	0.014	45
	9/30/2019	0.006	0.006	31	0.007	47
	10/29/2019	0.096	0.041	23	-0.005	40
	11/21/2019	0.014	0.040	17	-0.006	33
Herring	7/14/2019	0.052	0.006	17	0.052	37
5/17/2019	9/7/2019	0.225	0.061	14	0.137	52
	10/22/2019	0.082	0.015	18	0.075	62
Gillies	8/3/2019	0.013	NA	1	0.02	29
5/27/2019	9/15/2019	0.005	0.001	21	0.009	23
	11/2/2019	0.057	0.051	17	-0.011	21
Broad Rock	7/1/2019	0.028	0.023	4	-0.031	4
5/20/2019	8/15/2019	0.034	0.029	13	0.034	13
	9/17/2019	0.001	0.004	11	0.001	11
Reedy						
5/22/2019	7/9/2019	0.015	0.027	4	0.027	10
7/7/2019	8/12/2019	0.064	0.050	2	0.076	4

Table 3. Growth rates of mussels (mass in mg/d) stocked into urban and non-urban streams. Individual growth rates are based on repeat measurements of the same individual; population growth rates are derived from the average length of all individuals. Stocking dates for each site are shown in the first column.

		Individual GR (mg/d)			Population GR (mg/d)	
Site	Date	Mean	SE	N	Mean	N
Hatchery	7/15/2019	40.1	1.6	116	39.3	119
5/15/2019	8/27/2019	10.5	1.4	99	4.3	112
	10/2/2019	1.2	3.3	92	-1.1	104
	11/2/2019	5.7	2.6	86	1.3	100
Kimages	7/24/2019	10.5	1.6	45	16.0	65
5/18/2019	9/1/2019	13.8	1.9	39	14.2	45
	9/30/2019	6.6	4.9	31	3.2	47
	10/29/2019	-1.1	6.2	23	-10.7	40
	11/21/2019	-2.5	4.3	17	-5.5	33
Herring	7/14/2019	29.0	4.9	17	31.5	37
5/17/2019	9/7/2019	94.2	9.1	14	77.0	52
	10/22/2019	48.0	13.9	18	39.6	62
Gillies	8/3/2019	9.5	NA	1	9.4	29
5/27/2019	9/15/2019	10.6	1.9	21	8.4	23
	11/2/2019	-3.7	1.3	17	-8.3	21
Broad Rock	7/1/2019	7.5	2.3	2	0.0	8
5/20/2019	8/15/2019	-4.6	5.3	13	-4.6	13
	9/17/2019	0.9	1.4	11	0.9	11
Reedy						
5/22/2019	7/9/2019	11.3	8.4	4	10.2	10
7/7/2019	8/12/2019	15.8	1.3	2	0.0	4

Table 4. Mean values of water quality variables recorded at each site during April to December 2019.

Site	Temp C	pH	Conductivity µS/cm	DO %	DO mg/L	Discharge m^3/s
Herring	18.7	6.9	150.3	59.4	5.8	0.233
Pond	23.2	7.3	38.4	81.4	7.3	N/A
Kimages	16.4	7.1	103.7	69.5	6.7	0.021
Broad Rock	17.9	7.5	144.0	99.8	9.6	0.039
Reedy	17.2	7.4	196.8	90.9	9	0.014
Gillies	17.8	7.2	157.6	94.1	9.1	0.072

Figure 1. Design of in stream mussel enclosures

Figure 2. Stocking number and fate of Alewife Floater mussels in three urban streams (Reedy, Broad Rock, and Gillies), 2 non-urban streams (Herring, Kimages) and a fish hatchery rearing pond. The bars represent the number of mussels, blue representing stocked mussels, orange representing number of mussels alive, grey representing number of mussels lost, and yellow representing number of mussels' dead.

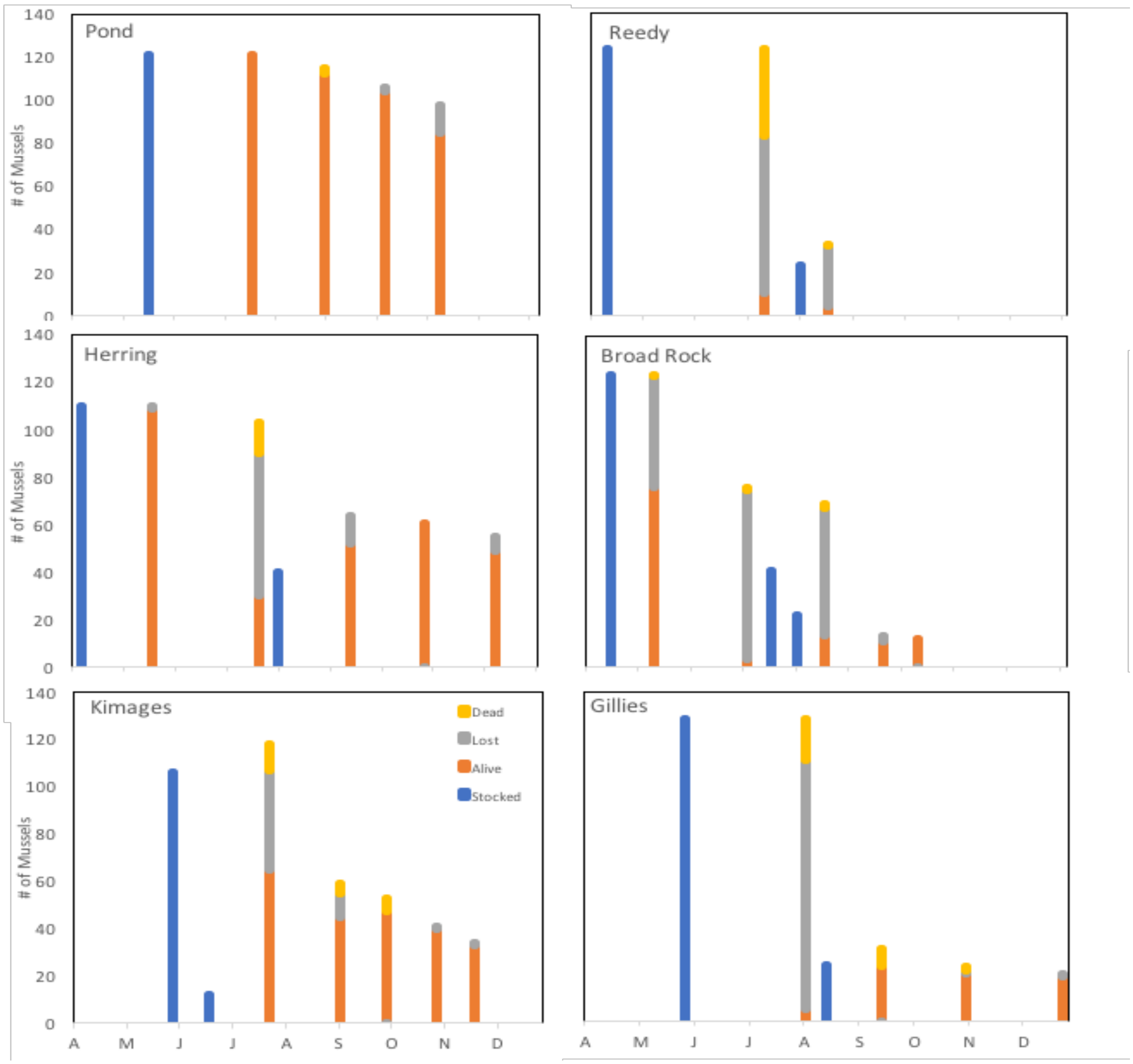

Figure 3. Length and mass based growth rates (with standard error) of Alewife Floater mussels in urban and rural streams as well as the hatchery pond. Data shown are average values based on repeat measurements of individually tagged mussels.

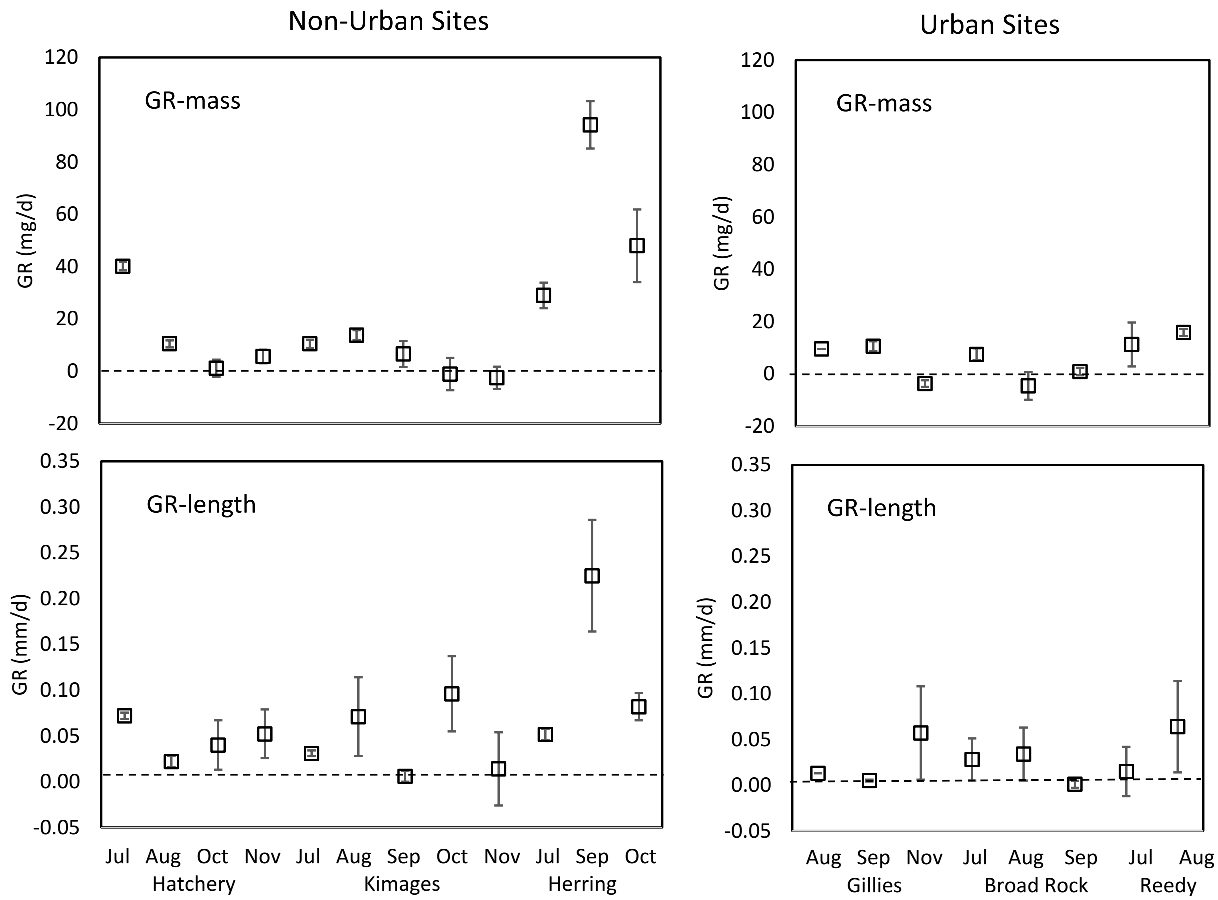

Figure 4. Comparisons of individual and population based growth rates as mass and length (upper panels). Comparisons of mass and length based growth rates derived from repeated measurements of individuals and population means (lower panels). All regressions $p < 0.001$.

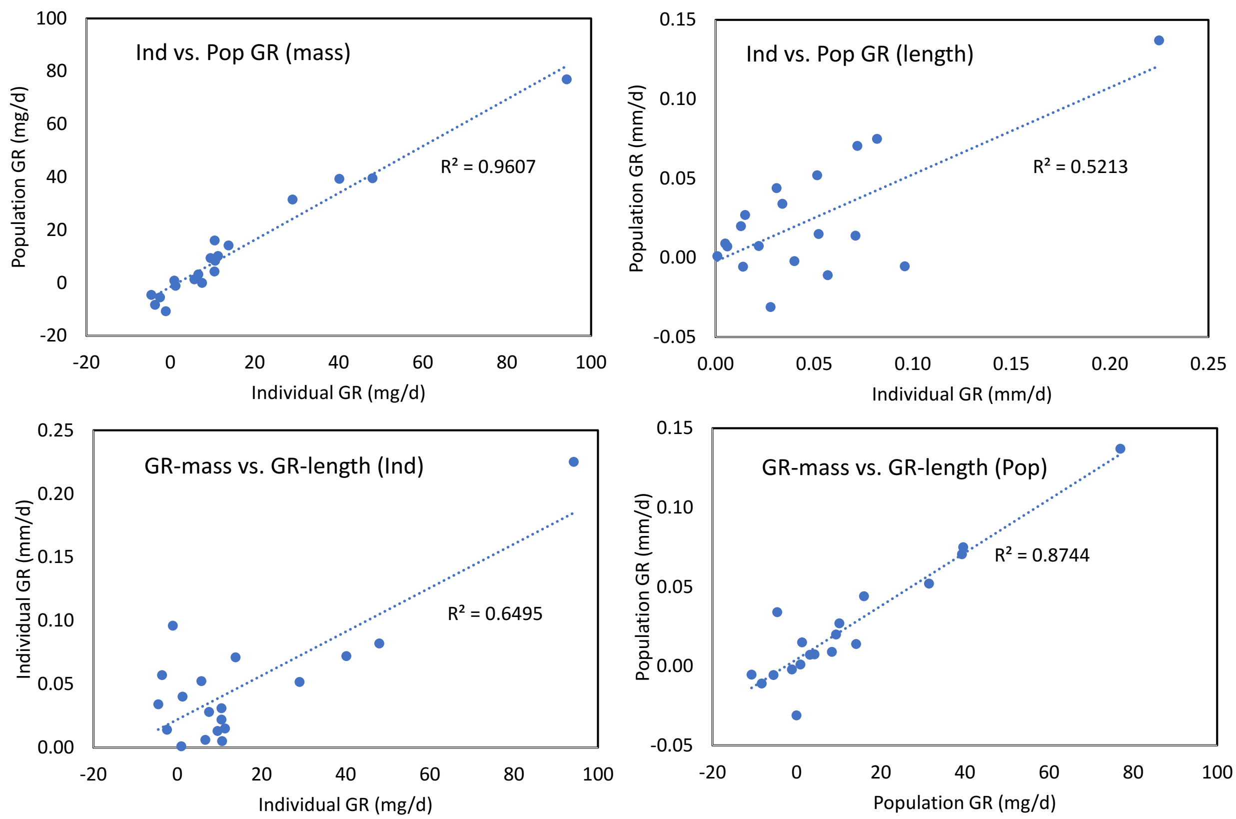

Figure 5. Water quality conditions at study sites during the period when mussels were deployed from June-December 2019.

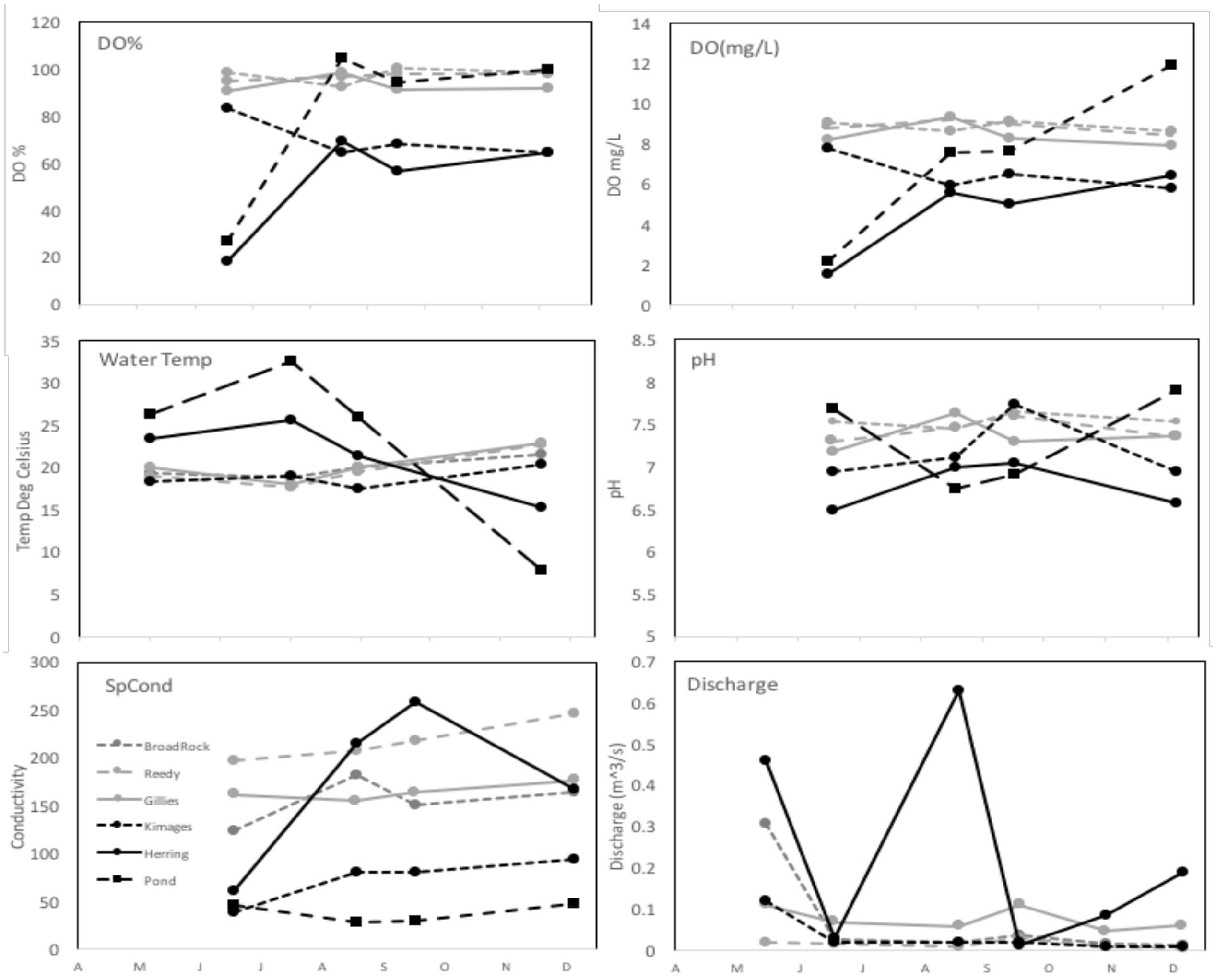

Figure 6. Food quantity and quality metrics for urban streams (Gil = Gillies, BR = Broad Rock, Ree = Reedy) and 3 non-urban sites (Kim = Kimages, Her = Herring, Pon = hatchery pond). Data shown are medians (dark black line), 25% and 75% quartiles (boxes), 95% confidence intervals (bars) and outliers (circles).

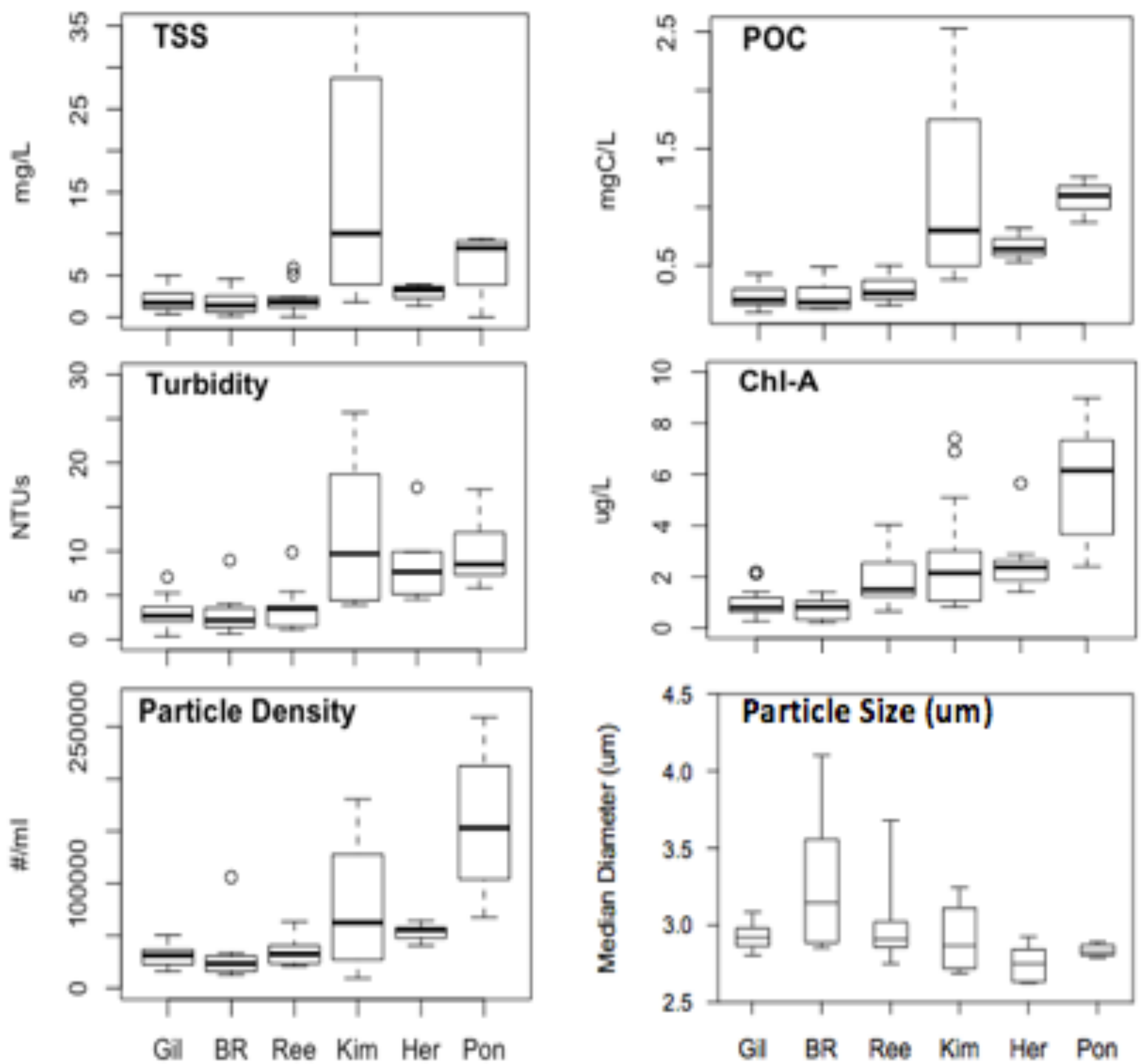

Figure 7. Water quality and food metrics as predictors of mussel mass growth rates. Regression lines denote statistically significant relationships.