

Original Research Paper

## Turbidity more than herbivory, negatively correlated with *Podostemum ceratophyllum* Michx. stem length and biomass in slow water habitats in three Appalachian Plateau rivers

Ashley Vankirk<sup>1</sup>, & James Wood <sup>1\*</sup>

<sup>1</sup>Department of Biology, West Liberty University, West Liberty, WV, USA;

### Article history

Received: 12 November 2024

Revised: 6 January 2025

Accepted: 8 January 2025

\*Corresponding Author: James Wood, West Liberty University, Department of Biology, West Liberty, WV, USA; Email: James.Wood@WestLiberty.edu

**Abstract:** The effects of environmental stressors (water chemistry, habitat characteristics, herbivory, etc.) on macrophytes are understudied and fewer studies have focused on how stressors impact macrophytes in swiftwater habitats. *Podostemum ceratophyllum*, a swiftwater macrophyte native to Eastern North America, has been identified as a foundation species of Appalachian rivers because of the plant's positive influence on macroinvertebrates, fish, and ecosystem processes. We initially sought to explore the effects of herbivory on the plant in three river systems on the Appalachian Plateau, however, we found little evidence of herbivory limiting *P. ceratophyllum* stem length and biomass. Instead, we observed that when *P. ceratophyllum* was exposed to low-flow conditions for approximately two weeks, stem length significantly decreased, and reduced stem length and plant biomass were correlated with turbidity in these rivers. Continued research exploring how environmental stressors influence *P. ceratophyllum* growth and survival could improve river management and support the conservation and restoration of *P. ceratophyllum* to eastern North American rivers.

**Keywords:** *Podostemum*; turbidity; water velocity; macrophyte; aquatic plant

## Introduction

Macrophytes (aquatic plants) in lotic habitats were thought to be primarily controlled by the physio-chemical conditions (e.g., water velocity, benthic substrate characteristics, light availability, pH, ionic concentration, etc.) but herbivory on macrophytes is increasingly recognized to be an important factor that influences plant biomass and community structure (Lodge 1991, Miller and Provenza 2006). Physical habitat, specifically water velocity, has previously been shown to negatively influence grazing on lotic macrophytes. For example, in a lowland stream in England, Wood et al. (2018) found that high-velocity conditions increase macrophyte biomass by limiting grazing access by mute swans (*Cygnus olor*). Similarly, in two Piedmont streams in Georgia, USA, Wood et al. (2019) found that reduced water velocity facilitated increased herbivory on the swift water macrophyte *Podostemum ceratophyllum* Michx.

(Podostemaceae), more commonly known as hornleaf riverweed or threadfoot. Water velocity also influences lotic macrophyte habitat through the deposition or removal of fine sediments and organic materials on plant tissue (roots, stems and leaves) (Chambers et al. 1991, Fritz et al. 2004), and as predicted by the Hjulström curve, even modest reductions in water velocity can facilitate rapid deposition of sediments (Hjulström 1935). Thus, water velocity can influence macrophyte distribution along the river continuum (slow water pools vs high-velocity cascades) through multiple interactions between macrophyte and physical habitats. However, for many macrophytes, specific habitat requirements, responses to environmental stressors, and the influence of herbivory are not well understood. Addressing knowledge gaps in macrophyte ecology can lead to the development of species-specific responses to environmental stressors (effects of extreme flooding, drought, sediment

loading, etc.) and management actions (establishing minimum environmental flows, effects of pulse flows, etc.).

We sought to explore the effect of herbivory on the macrophyte *Podostemum ceratophyllum* in short-term low water velocity conditions. The native range of *Podostemum ceratophyllum* extends across streams and rivers in Eastern North America's montane and Piedmont regions with disjunction populations in the Dominican Republic and Honduras (Philbrick and Crow 1983). *Podostemum ceratophyllum* grows attached to stable, benthic substrates in high water velocity habitats (Philbrick and Crow 1983, Wood and Freeman 2017) including bedrock, cobbles, gravels as well as on mussel shells, tires, metal, and wood (Per. Obs Wood). *Podostemum ceratophyllum*, henceforth referred to as *Podostemum* is the only member of the family Podostemaceae found in North America, and was identified as a foundation species in eastern rivers by Wood and Freeman (2017) due to the plant's considerable potentially high biomass (Hill and Webster 1985) and positive influence on benthic biota, including fish (Argentina et al. 2010b, Baynes et al. 2022) and invertebrates (Grubaugh and Wallace 1995, Hutchens et al. 2004), and its contribution to food webs (Nelson and Scott 1962, Rodgers et al. 1983, Parker et al. 2007). *Podostemum* is generally thought to be restricted to high-velocity habitats, and both Parker et al. (2007) and Wood et al. (2019) have observed that high water velocity conditions limit grazing on the plant. However, the water velocity-herbivory relationship has only been documented in the southeastern portion of the plant's range (Fig 1; Parker et al. 2007, Wood et al. 2019).

To better understand how herbivory and water velocity influence *Podostemum*, we conducted an herbivory exclusion study in three rivers on the Appalachian Plateau of West Virginia and Pennsylvania to assess how low water velocity habitats affect *Podostemum*. We hypothesize that when *Podostemum* experiences low water velocity conditions, herbivory will increase and decrease plant stem length and biomass. Specifically, we asked the following questions: (1) In the Appalachian plateau region, do low water velocity conditions facilitate herbivory on *Podostemum ceratophyllum*, reducing stem length and biomass on *Podostemum*? (2) In the absence of macro-consumers, do low-velocity conditions alone reduce *Podostemum* stem length and overall biomass? (3)

and post hoc we asked, how might turbidity impact *Podostemum* in low water velocity habitats?

## Materials and Methods

### Study Location

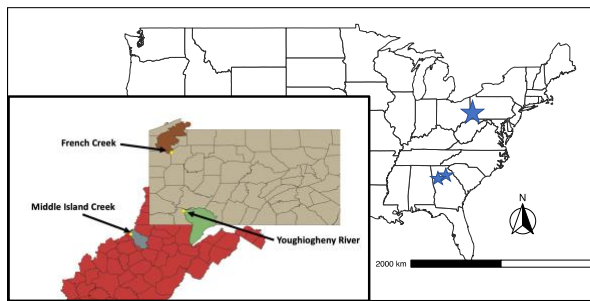
Studies were conducted in one river in West Virginia and two rivers in Pennsylvania where *Podostemum* grows with relative abundance: Middle Island Creek, WV, French Creek, PA, and Youghiogheny River, PA (Fig. 1). All sites were in mid-order rivers with an open canopy and fast-flowing shoals, consisting mainly of cobbles and gravel, intermixed with bedrock and sand. The study was conducted from 22 June 2022 through 06 July 2022 at French Creek, a 6th order stream and tributary to the Allegheny River in northwestern Pennsylvania; from 29 June 2022 through 13 July 2022 in Middle Island Creek, a 5th order stream and a tributary to the Ohio River in the upper Ohio River Valley; and from 3 August 2022 through 17 August 2022 in the Youghiogheny River, henceforth the Yough, a 6th order river and tributary to the Monongahela River in southwestern Pennsylvania. Sites were chosen based on herbarium records from the Carnegie Museum of Natural History and from previous studies of *Podostemum* in the region (Canfield 2019).

### Effects of low water velocity

To assess whether low water velocity conditions facilitate herbivory on *Podostemum*, we conducted a consumer exclusion experiment. At each of the three study sites we identified approximately 50 m reaches of the river colonized by *Podostemum* where water velocity was relatively fast ( $> 0.5\text{m/s}$ ) and plants were attached to bedrock and cobble substrates. We then collected ten cobble substrates, approximately 15 x 15 x 15 cm in size, that were densely colonized with *Podostemum*, and stems were at least 3 cm long and relatively uniform. Substrates were kept in shallow water while processing to prevent desiccation and damage to the plants. Each of the collected substrates was randomly assigned into paired sets of a consumer exclusion "Caged" treatment and a consumer accessible "Uncaged" treatment. The cage used in the caged treatment was a cylindrical wire mesh minnow trap, where openings were approximately 1cm x 1 cm, and the larger openings at each end were zipped tied closed to

prevent macroconsumers (i.e., consumers larger than the macroinvertebrates) from entering the enclosure.

Pairs of substrates were uniquely identified with colored zip ties, creating five pairs (Cage & Uncaged) of substrates. Then, ten stems on each substrate were randomly measured to estimate the average stem length of that substrate. The paired substrates were then placed into low-velocity habitats (< 0.5 m/s) in the stream, either upstream or downstream from where they were collected. Rocks in each pair were placed in close proximity to each other (< 0.5 meters apart) and each pair was placed at least 1 m apart away from any other substrate pair. Substrates were allowed to incubate *in situ* for approximately two weeks. After two weeks, the substrates were collected, and 10 stems from each substrate were randomly measured to assess changes in stem length during the low-flow velocity treatment.



**Figure 1.** The background map of the USA shows the location of this study (large star) and the location of the other herbivory studies (Parker et al. 2007 and Wood et al. 2019) (small stars). The inset map shows the watersheds focused on in this study, Middle Island Creek (WV), French Creek (PA), and the Youghiogheny River (PA).

At each location where a pair of substrates were deployed, water velocity and depth were recorded using the Hach EM950 Flow Meter and wading rod, and the water chemistry parameters, dissolved oxygen (DO, mg/L), specific conductance (SPC,  $\mu\text{s}/\text{cm}$ ), chloride (Cl, mg/L), water temperature ( $^{\circ}\text{C}$ ), turbidity (NTU), and pH were collected for each site with a calibrated YSI Quatro Handheld meter or Hach 2100QTurbidimeter (Table 1; Table S2).

To estimate the water velocity *Podostemum* experienced inside the caged treatment, we measured water velocity inside the cage by placing the velocity meter inside the cage after the substrate was

removed. Water velocity measurements inside the cages were taken at the approximate location of the caged treatment at the Yough and French Creek sites. However, the water velocity in the cage was not collected at Middle Island Creek; therefore, we estimated water velocity in the cage using a flow reduction factor of the cage based on measurements at French Creek and the Yough (Table S1). The flow reduction factor was estimated with the equation, water velocity in cage / water velocity outside cage = reduction factor, and we estimated that cages reduced water velocity by 56%.

After stem length was measured on each substrate post-treatment, *Podostemum* biomass was collected from 35.2  $\text{cm}^2$  area with a custom biomass sampler. All plant material in the sampling area was scraped from the substrate and placed in a labeled plastic bag. Labeled bags were placed on ice and transported to the laboratory. In the laboratory, the samples were washed over a 1,000 micron-sieve to remove detritus, macroinvertebrates, and sediments. Plant material was then dried at  $60^{\circ}\text{C}$  for 48 hrs, weighed, ashed in a muffle furnace at  $500^{\circ}\text{C}$  for 4 hrs, and weighed again to calculate the ash-free dry mass (AFDM) for each sample ( $\text{AFDM} = \text{Dry Wt} - \text{Ashed Wt}$ ). Biomass was assessed as  $\text{g AFDM m}^{-2}$ .

Statistical analysis was performed using the R studio software package v4.1.3 and figures were produced using the R package ggplot (R Core Team 2022). At each site we assessed pre- and post-stem length in both treatments, changes in stem length before and after cage treatments, and post-treatment differences in biomass (AFDM) using paired t-tests. Because we only had site-level turbidity data, we used linear mixed effect models to account for pseudo-replication within each site with the “lme4” package in R with a random effect of “Site”. We modeled change in stem length and biomass as predicted by turbidity and exposed water velocity individually and with an additive model. To improve the model fit, we modeled the natural log of turbidity in the biomass model. Additionally, we assessed how the change in stem length and biomass were related to turbidity and water velocity in a series of linear regression models with single and additive fixed effects. Because turbidity was a site-level predictor, we were not able to model the effect of turbidity within the sites and thus, at the site level, we assessed the effects of water velocity on change in stem length and biomass.

**Table 1.** Summary of water chemistry parameters for all sites . See supplemental Table S1 for water chemistry parameters for each sample event.

Site	Dissolved Oxygen (mg/L)	Specific Conductivity ( $\mu\text{s}/\text{cm}$ )	Chloride (mg/L)	Water Temperature ( $^{\circ}\text{C}$ )	Turbidity (NTU)	pH
French Creek	9.9	267.9	17.0	21.7	14.5	8.0
Middle Island Creek	5.4	227.0	13.3	27.1	43.8	7.6
Youghiogheny River	9.7	232.0	13.2	21.5	4.7	7.3

## Results

Water chemistry between the three sites was similar, although mean DO was lower and mean turbidity and temperature were higher in Middle Island Creek. Specific conductance and turbidity were lowest in the Yough River (Table 1; Table S2). Exposed low-water velocity treatments across all sites ranged from 0.004 – 0.28 m/s ( $\mu = 0.08$ ) and the depth ranged from 24 cm – 58 cm ( $\mu = 40.2$ ).

At Middle Island Creek, the uncaged pretreatment stem length averaged 8.0 cm (SE = 1.3), while uncaged post-treatment averaged 3.5 cm (SE = 1.2). Caged pretreatment stem length averaged 5.8 cm (SE = 0.5) and caged post-treatment averaged 2.1 cm (SE = 0.4). *Podostemum* stem length significantly decreased in both caged and uncaged treatments (uncaged:  $t(8) = 2.54$ ,  $p = 0.03$ ; caged:  $t(8) = 5.95$ ,  $p < 0.001$ ) during the study (Fig. 2a – b; Fig. 3a – b; Fig. 4a). Change in stem length at Middle Island Creek (caged  $\mu = -3.8$  cm, SE = 0.4; uncaged  $\mu = -4.5$ , SE = 1.6) was not significantly different between the treatments ( $t(4) = 0.48$ ,  $p = 0.66$ ; Fig. 4b), but post-treatment biomass (caged  $\mu = 43.2$  g AFDM  $\text{m}^{-2}$ , SE = 13.7; uncaged  $\mu = 117.6$ , SE = 27.8) was significantly higher in the uncaged treatment ( $t(4) = -3.03$ ,  $p = 0.04$ ; Fig. 4c).

At French Creek, the uncaged pretreatment stem length averaged 6.4 cm (SE = 0.4), while uncaged post-treatment averaged 4.4 cm (SE = 0.62). Caged pretreatment stem length averaged 5.9 cm (SE = 0.4), and caged post-treatment averaged 3.7 cm (SE = 0.7) and stem length significantly decreased in both caged ( $t(8) = 2.9$ ,  $p = 0.02$ ) and uncaged treatments ( $t(8) = 2.74$ ,  $p < 0.03$ ) during the study (Fig 2c – d; Fig. 3c – d; Fig. 4d). Change in stem length was not significantly different between caged ( $\mu = -2.3$ , SE = 0.5) and uncaged ( $\mu = -2.0$ , SE = 0.7) treatments ( $t(4) = -0.44$ ,  $p = 0.68$ ; Fig. 4e) and *Podostemum*

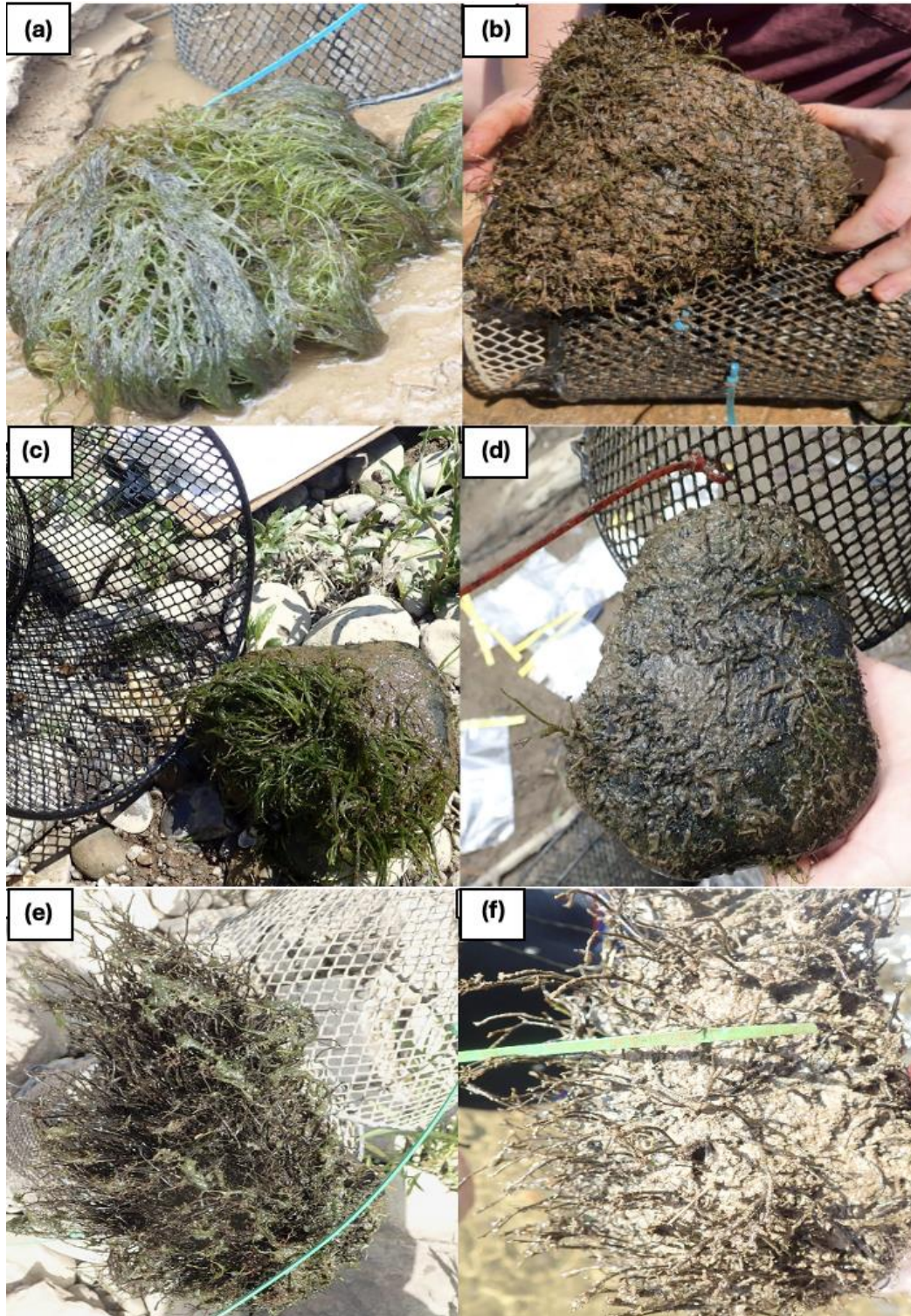
biomass was not significantly different between the caged ( $\mu = 64.8$  g AFDM  $\text{m}^{-2}$ , SE = 23.9) and uncaged ( $\mu = 93.2$ , SE = 18.5) treatments ( $t(4) = -0.84$ ,  $p = 0.45$ ; Fig. 4f).

At the Yough River sites, the uncaged pretreatment stem length averaged 6.3 cm (SE = 0.7) and the post-treatment averaged 5.6 cm (SE = 0.4), while caged pretreatment stem length averaged 5.6 cm (SE = 0.6) and the post-treatment averaged 4.8 cm (SE = 0.4). In both the caged and uncaged treatment, no significant change in stem length was observed between pre- and post-experiment (caged  $t(8) = 1.04$ ,  $p = 0.32$ , uncaged ( $t(8) = 0.84$ ,  $p = 0.42$ ) during the study (Fig. 2e – f; Fig. 3e – f; Fig. 4g). Change in stem length was not significantly different between the caged and uncaged treatments ( $t(4) = -0.15$ ,  $p = 0.88$ ; Fig. 4h). Even though mean *Podostemum* biomass was considerably higher in the uncaged treatments ( $\mu = 589.4$  g AFDM  $\text{m}^{-2}$ , SE = 65.2) compared to the caged treatments ( $\mu = 327.8$  g AFDM  $\text{m}^{-2}$ , SE = 97.1), biomass was not significantly different between treatments overall ( $t(4) = -1.88$ ,  $p = 0.13$ ; Fig. 4i).

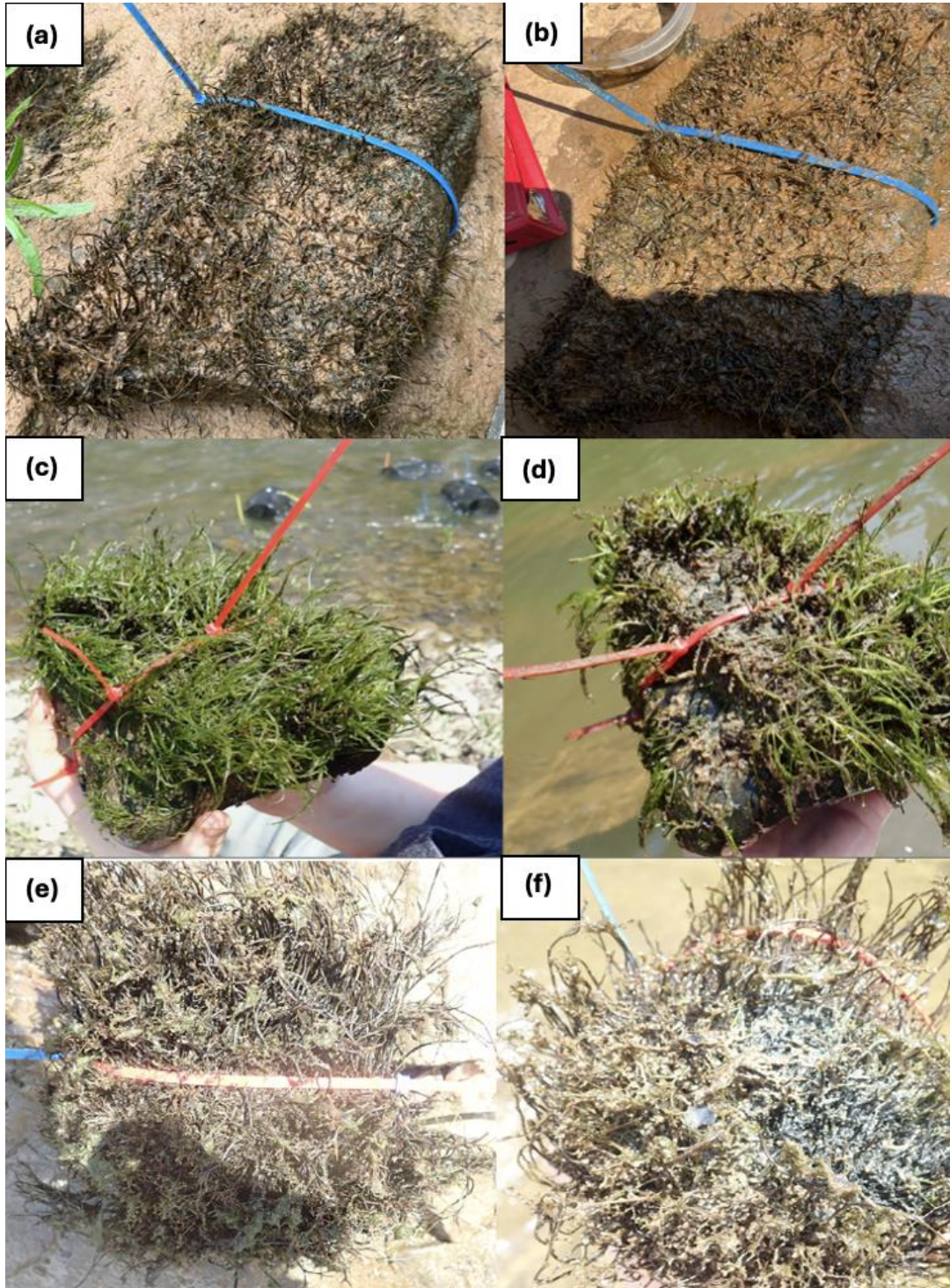
We did not find a significant effect of water velocity on change in stem length in the low-flow habitats (Fig. 5a) but the range of water velocity was small. However, our linear mixed-effects models suggest that increased turbidity increased the degree to which *Podostemum* stems were reduced in length when modeled across all study sites ( $\text{mR}^2 = 0.43$ ,  $\text{cR}^2 = 0.43$ ,  $p < 0.001$ ; Fig. 5b), and we found that increasing stream water turbidity was correlated with lower biomass ( $\text{mR}^2 = 0.31$ ,  $\text{cR}^2 = 0.31$ ,  $p < 0.001$ ; Table 2; Fig. 5c) but our low sample size of stream turbidity measurement suggests caution when interpreting our results. The greatest reduction in stem length occurred at Middle Island Creek where

**Table 2.** Summary of model parameters estimates predicting change in stem length (CSL) and ash free dry mass (AFDM) during the experiment. Fixed effects were model as additive effects. LMER = Linear Mixed Effect model, LM = linear model,  $mR^2$  = model fit of fixed effects only,  $cR^2$  = model fit of fixed and random effects.

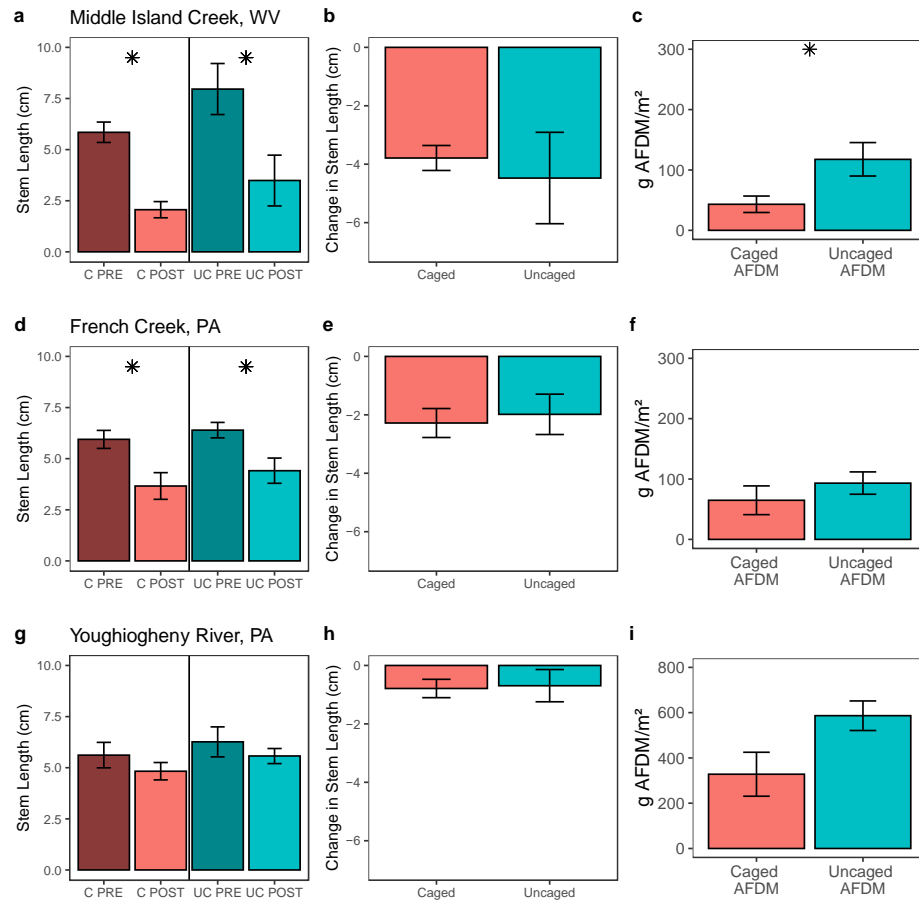
Model type	Response variable	Treatment	Turbidity	Exposed Water Velocity	Intercept	$R^2$
All Sites						
LMER	CSL	all	<b>-0.090 (p &lt; 0.001)</b>	-5.605 (p = 0.23)	-0.007 (p = 0.99)	<b><math>mR^2 = 0.43</math> <math>cR^2 = 0.43</math></b>
LMER	CSL	all		-4.44 (p = 0.42)	-1.975 (p = 0.19)	$mR^2 = 0.02$ $cR^2 = 0.52$
LMER	CSL	all	<b>-0.083 (p = 0.001)</b>		-0.606 (p = 0.23)	<b><math>mR^2 = 0.41</math> <math>cR^2 = 0.41</math></b>
LMER	AFDM	all	<b>(ln) -6.841 (p = 0.58)</b>	474.59 (p = 0.30)	310.41 (p = 0.41)	$mR^2 = 0.17$ $cR^2 = 0.82$
LMER	AFDM	all	<b>(ln) -7.434 (p = 0.001)</b>		361.101(p < 0.001)	<b><math>mR^2 = 0.31</math></b> <b><math>cR^2 = 0.31</math></b>
LMER	AFDM	all		488.86 (p = 0.28)	<b>166.06 (p = 0.34)</b>	<b><math>mR^2 = 0.02</math></b> <b><math>cR^2 = 0.74</math></b>
LM	CSL	all	<b>-0.090 (p &lt; 0.001)</b>	-5.605 (p = 0.23)	-0.007	<b>0.45</b>
	CSL	caged	<b>-0.071 (p &lt; 0.001)</b>		-0.799 (p = 0.07)	<b>0.63</b>
LM	CSL	caged	<b>-0.088 (p &lt; 0.001)</b>	<b>-21.819 (p &lt; 0.01)</b>	0.596 (p = 0.28)	<b>0.80</b>
LM	CSL	uncaged	<b>-0.094 (p = 0.02)</b>		-0.410 (p = 0.66)	<b>0.36</b>
LM	CSL	uncaged	<b>-0.104 (p = 0.02)</b>	-5.534 p = 0.49	0.416 (p = 0.79)	<b>0.39</b>
LM	AFDM	all	<b>-168.43 (p &lt; 0.001)</b>		<b>653.37 (p &lt; 0.001)</b>	<b>0.50</b>
LM	AFDM	caged	<b>-127.14 (p &lt; 0.01)</b>		<b>483.35 (p = 0.001)</b>	<b>0.45</b>
LM	AFDM	uncaged	<b>-209.71 (p &lt; 0.001)</b>		<b>823.39 (p &lt; 0.001)</b>	<b>0.63</b>



**Figure 2.** A representative sample of the caged treatments pre- (a, c, e) and post- (b, d, f) caged treatments for Middle Island Creek, WV (a-b), French Creek, PA (c-d), and Youghiogheny River, PA (e-f).



**Figure 3.** A representative sample of the uncaged treatments pre- (a, c, e,) and post- (b, d, f,) for Middle Island Creek, WV (a-b), French Creek, PA (c-d), and Youghiogheny River, PA (e-f).



**Figure 4.** Mean and standard error (SE) of pre and post-experiment stem length in both uncaged and caged treatments for Middle Island Creek (a), French Creek (d) and Youghiogheny River (e). Mean and SE of the change in stem length and between caged and uncaged treatment in Middle Island Creek (b), French Creek (e), and the Youghiogheny River (h), and mean Ash Free Dry Mass (AFDM) and SE for the caged and uncaged treatments for Middle Island Creek (c), French Creek (f), and Youghiogheny River (i). The asterisk (\*) indicates significance ( $p < 0.05$ ) groups. Red colors represent the caged treatment and blue colors represent the uncaged treatments.

turbidity was highest, followed by French Creek, and the smallest change was at our lowest turbidity site in the Yough. Modeling of change in stem length with a simple linear regression yielded similar results (Table 2; Table S3). Only at the Middle Island Creek site, in the cage treatment, did we find an effect of water velocity on change in stem length, which was negative (Table 2). We did not find any significant effects of water velocity on biomass.

## Discussion

Our results differed from the findings of Wood et al. (2019) as we did not find compelling evidence that herbivory was limiting *Podostemum* stem length and biomass in low-water velocity conditions at our study sites, even though we did observe modest

herbivory at Middle Island Creek (noted on one uncaged substrate with a ~10cm reduction in stem length). Conversely, our data suggests that turbidity negatively affect *Podostemum* in low water velocity habitats ( $< 0.5\text{m S}^{-1}$ ) by reducing stem length and biomass through plant dieback. We observed a considerable buildup of fine sediments on the roots and haptera (holdfast structures) of *Podostemum* over the course of the study. The accrual of fine sediments may rapidly create anoxic conditions in the root zone, limit sunlight to the photosynthetically active roots, and stress the plants. However the direct effects of sediment accrual on *Podostemum* growth remain largely unexplored.

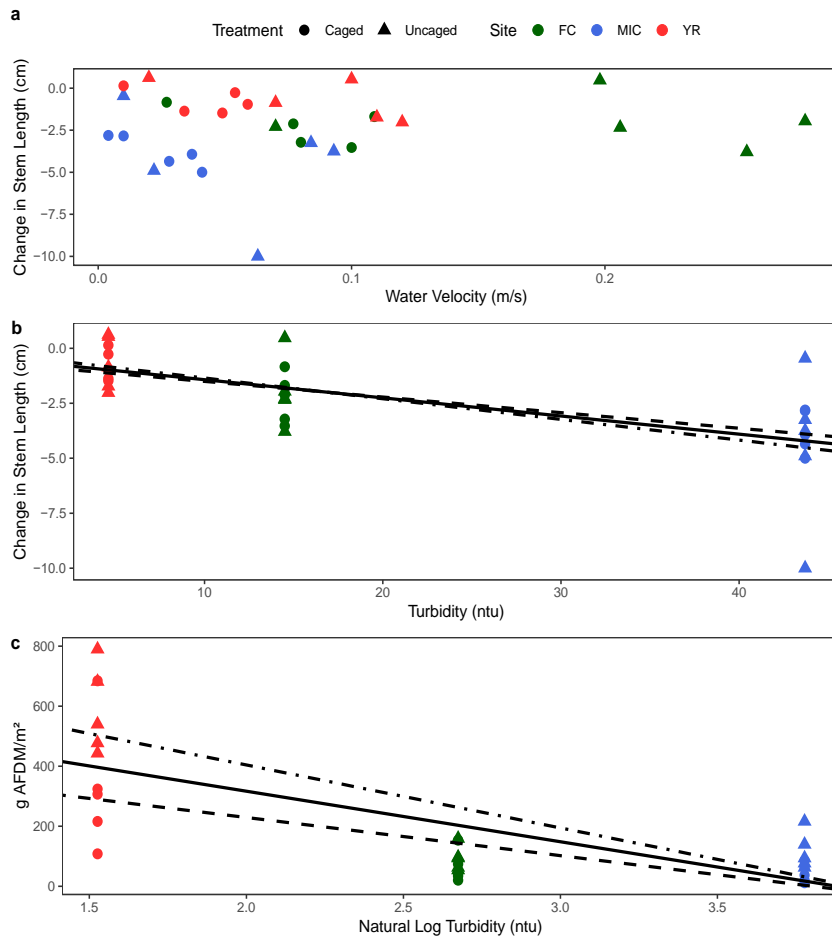
Our study adds to the literature that has observed negative effects of turbidity on *Podostemum*. For example, Connelly et al. (1999) reported



*Podostemum* populations had declined in the Roanoke River, VA, likely due to an increase in sedimentation. Additionally, Canfield and Wood (2022) working in West Virginia rivers where *Podostemum* had historically been observed, noted an absence of *Podostemum* at sites where considerable deposition of fine sediments had occurred. Similarly, Grubaugh and Wallace (1995) concluded that a decline in agriculture and sedimentation over time had likely increased *Podostemum* biomass in the Middle Oconee River. Our results suggest that sedimentation related to turbidity in low-flow habitats may be of considerable concern for conservation and restoration efforts focused on *Podostemum*. Ultimately resulting in the loss of plant biomass and a decline in population health and size. Sediment accumulation and turbidity can negatively affect the growth of submerged macrophytes by reducing light availability (Barko and Smart 1986, Köhler et al. 2010) and *Podostemum* is known to prefer high-light habitats (Everitt and Burkholder 1991, Argentina et al. 2010a). Additionally, the accumulation of fine sediments on the stems and leaves could stress the plants by reducing light availability. Benthic sediments can

also create anoxic environments in the root zone, which could limit root respiration (Sand-Jensen et al. 1982, Caraco et al. 2006). Our observations were that *Podostemum* stems became brittle, and roots were easily detached from their substrate after only two weeks of incubation in low-water velocity habitats. We suspect that turbidity and the resulting sedimentation in low-velocity habitats negatively affected *Podostemum* in our study by limiting oxygen availability to the roots and possibly negatively affecting the biofilm community associated on the haptera (Jäger-Zürn and Grubert 2000, Vankirk 2023). Nonetheless, our biomass estimates in the Yough were similar to 3 times those reported by Hill and Webster (1985) in the New River, but in our more turbid rivers, our biomass estimates were generally lower. Future studies that focus on quantifying sedimentation and root zone activity (i.e., oxygen demand) could provide additional insight into the causal relationship between turbidity, sedimentation, and *Podostemum* growth and senescence.

The reason our results differed from the findings of Wood et al. (2019), may be due to regional



**Figure 5.** Water velocity was not significantly correlated with change in stem length (a), but stream water turbidity was correlated ( $p < 0.05$ ) with change in stem length across all sites and in individual treatments (b), and turbidity was negatively correlated with biomass across all sites and in individual treatments ( $p < 0.05$ ) (c). Solid line represents the trend line for both cage and uncaged treatment, dashed line represents caged sites, and dot-dash line represents the trend line for the uncaged sites. See Table 2 for complete model results.

differences in fauna, specifically turtle diversity, which may be partly responsible for the modest indications of herbivory we observed in our study. One of the most likely consumers suggested in Wood et al. (2019)'s study was the turtle *Pseudemys concinna* (river cooter), which is abundant in the Piedmonts of Georgia. *Pseudemys concinna* are known to be primarily herbivorous as adults. Aresco and Dobie (2000), found abundant *Podostemum* in the stomachs of *P. concinna* in an Alabama study. Additionally, Wood et al. (2019) and Parker et al. (2007), indicated crayfish and Canada geese grazed on the plant. We hypothesized that herbivory would also occur at our sites, however this hypothesis was not generally supported. The native range of *Pseudemys concinna* (river cooter turtle) extends to the eastern and southwestern portion of West Virginia and there are no documented populations in Pennsylvania (Seidel and Dreslik 1996). Crayfish, Canada geese and other turtle species (e.g.,

*Chrysemys picta*, painted turtles) are possible consumers in our study area with wide geographic distribution but they were not observed during our study. Communications with a member of the local community indicated that turtles were occasionally hunted from the bridge immediately downstream of our study site on Middle Island Creek, likely decreasing the local abundance of turtles. Additional conversations with property owners near the study site on Middle Island Creek indicated that white-tailed deer (*Odocoileus virginianus*) have been observed to occasionally consume *Podostemum* from the stream, but this was not observed in our study.

While our study was originally focused on documenting herbivory, our findings suggest that in our study sites, *Podostemum* is more negatively affected by stream water turbidity in low water velocity habitats than herbivory and that the negative impacts of sedimentation can occur quickly (~ two weeks). Unfortunately, our study design prevented a

more nuanced and more statistically rigorous assessment of the effects and interactions between turbidity and water velocity on *Podostemum*, and we cannot rule out another factor that could also influence our results. For example, Middle Island Creek had a lower DO and higher water temperature than the other streams, which may stress the plant. Reduced stem length could also be due to handling effects, although efforts were made to keep the plants submerged and prevent damage to the plant during the transplant and we do not think the reductions in stem length observed were due to handling and transport. Additionally, variation in flow may have differentially affected the streams, however, the duration of the study was short (two weeks) and stream flow variation was minimal at all sites during the study, thus we do not think changes in flow (discharge or velocity) were major factor affected this study. While we found only minimal evidence of herbivory in the Mid-Atlantic Appalachian plateau region, additional herbivory studies in other regions of Appalachia would help elucidate the effects of aquatic and terrestrial herbivory on this widespread Appalachian species.

Overall, this study provided new insight into the effects of low water velocity conditions and sedimentation on *Podostemum* stem length and biomass. We found that when *Podostemum* was exposed to low water velocity conditions, stem length and biomass decreased, and the loss of stem length was correlated with higher stream water turbidity. Our results suggest that high turbidity in low water velocity habitats can lead to rapid degradation of the plant. Because *Podostemum* is a foundation species of Appalachian rivers and appears to be a general bioindicator of ecosystem health (Meijer 1976), its loss could alter food webs, reduce macroinvertebrate biomass and habitat for fishes, decrease stream bed stability, and reduce nutrient uptake in eastern rivers (Wood and Freeman 2017, Wood et al. 2024). Continued research exploring how environmental stressors (e.g., sedimentation, low water velocity conditions, extreme flood events, changes in water chemistry) can influence *Podostemum* growth and physiology could help conserve and restore the plant to eastern North American Rivers.

### Acknowledgments

We would like to thank the WVNASA and

WLU Department of Biology for their support of this project as well as Emily Huff, Mycah Richie-Yoho and Montana Fonner for their assistance with field work.

### Literature Cited

- Aresco, M. J., and J. L. Dobie. 2000. Variation in shell arching and sexual size dimorphism of river cooters, *Pseudemys concinna*, from two river systems in Alabama. *Journal of Herpetology* **34**:313-317.
- Argentina, J. E., M. C. Freeman, and B. J. Freeman. 2010a. Predictors of occurrence of the aquatic Macrophyte *Podostemum ceratophyllum* in a Southern Appalachian River. *Southeastern Naturalist* **9**:465-476.
- Argentina, J. E., M. C. Freeman, and B. J. Freeman. 2010b. The response of stream fish to local and reach-scale variation in the occurrence of a benthic aquatic macrophyte. *Freshwater Biology* **55**:643-653.
- Barko, J. W., and R. M. Smart. 1986. Sediment-related mechanisms of growth limitation in submersed macrophytes. *Ecology* **67**:1328-1340.
- Baynes, A. Y., M. C. Freeman, S. K. McKay, and S. J. Wenger. 2022. Habitat associations of riverine fishes among rocky shoals. *Ecology of Freshwater Fish*:336-347.
- Canfield, S. A. 2019. Investigating the Dynamics of a Lotic Macrophyte Community in West Virginia with an Emphasis on the Distribution and Niche Requirements of *Podostemum ceratophyllum*.
- Canfield, S. A., and J. L. Wood. 2022. New Populations of the Rheophytic Macrophyte *Podostemum ceratophyllum* Michx. (Hornleaf Riverweed) in West Virginia. *Castanea* **86**:214-224, 211.
- Caraco, N., J. Cole, S. Findlay, and C. Wigand. 2006. Vascular plants as engineers of oxygen in aquatic systems. *BioScience* **56**:219-225.
- Chambers, P. A., E. E. Prepas, H. R. Hamilton, and M. L. Bothwell. 1991. Current velocity and its effect on aquatic macrophytes in flowing waters. *Ecological Applications* **1**:249-257.
- Connelly, W. J., D. J. Orth, and R. K. Smith. 1999. Habitat of the riverweed darter, *Etheostoma podostemone* Jordan, and the decline of riverweed, *Podostemum ceratophyllum*, in the tributaries of the Roanoke River, Virginia. *Journal of Freshwater Ecology* **14**:93-102.
- Everitt, D., and J. Burkholder. 1991. Seasonal dynamics of macrophyte communities from a stream

- flowing over granite flatrock in North Carolina, USA. *Hydrobiologia* **222**:159-172.
- Fritz, K. M., M. M. Gangloff, and J. W. Feminella. 2004. Habitat modification by the stream macrophyte *Justicia americana* and its effects on biota. *Oecologia* **140**:388-397.
- Grubaugh, J. W., and J. B. Wallace. 1995. Functional structure and production of the benthic community in a piedmont river: 1956-1957 and 1991-1992. *Limnology and Oceanography* **40**:490-501.
- Hill, B. H., & Webster, J. R. (1984). Productivity of *Podostemum ceratophyllum* in the New River, Virginia. *American journal of botany*, **71**(1), 130-136.
- Hjulstrom, F. (1935). Studies of the morphological activity of rivers as illustrated by the River Fyris, Bulletin. Geological Institute Upsalsa, **25**, 221-527
- Hutchens, J. J., J. B. Wallace, and E. D. Romaniszyn. 2004. Role of *Podostemum ceratophyllum* Michx. in structuring benthic macroinvertebrate assemblages in a southern Appalachian river. *Journal of the North American Benthological Society* **23**:713-727.
- Jäger-Zürn, I., and M. Grubert. 2000. Podostemaceae depend on sticky biofilms with respect to attachment to rocks in waterfalls. *International Journal of Plant Sciences* **161**:599-607.
- Köhler, J., J. Hachoł, and S. Hilt. 2010. Regulation of submersed macrophyte biomass in a temperate lowland river: Interactions between shading by bank vegetation, epiphyton and water turbidity. *Aquatic Botany* **92**:129-136.
- Lodge, D. M. 1991. Herbivory on freshwater macrophytes. *Aquatic Botany* **41**:195-224.
- Meijer, W. A Note on *Podostemum ceratophyllum* Michx., as an indicator of clean streams in and around the Appalachian Mountains. *Castanea*, **41**(4), 319-324.
- Miller, S. A., and F. D. Provenza. 2006. Mechanisms of resistance of freshwater macrophytes to herbivory by invasive juvenile common carp. *Freshwater Biology* **52**:39-49.
- Nelson, D. J., and D. C. Scott. 1962. Role of Detritus in the Productivity of a Rock-Outcrop Community in a Piedmont Stream. *Limnology and Oceanography* **7**:396-413.
- Parker, J. D., D. E. Burkepile, D. O. Collins, J. Kubanek, and M. E. Hay. 2007. Stream mosses as chemically-defended refugia for freshwater macroinvertebrates. *Oikos* **116**:302-312.
- Philbrick, C. T., and G. E. Crow. 1983. Distribution of *Podostemum ceratophyllum* Michx. (Podostemaceae). *Rhodora* **85**:325-341.
- R Core Team. 2022. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL <https://www.R-project.org/>. R Foundation for Statistical Computing, Vienna, Austria.
- Rodgers, J. H., M. E. McKeivitt, D. O. Hammerlund, K. L. Dickson, and Cairns J, Jr. 1983. Primary production and decomposition of submergent and emergent aquatic plants in two Appalachian rivers. Pages 283-301 in T. D. Fontaine Iii and S. M. Bartell, editors. Ann Arbor Science Publishers, Ann Arbor, MI.
- Sand-Jensen, K., C. Prahl, and H. Stokholm. 1982. Oxygen release from roots of submerged aquatic macrophytes. *Oikos*:349-354.
- Seidel, M. E., and M. J. Dreslik. 1996. *Pseudemys concinna*.
- Vankirk, A. N. 2023. Understanding How Water Velocity Affects Herbivory Pressure on *Podostemum ceratophyllum* Michx. and the Identification of Cyanobacterial Biofilms Associated with Attachment. West Liberty University.
- Wood, J., and M. Freeman. 2017. Ecology of the macrophyte *Podostemum ceratophyllum* Michx. (Hornleaf riverweed), a widespread foundation species of eastern North American rivers. *Aquatic Botany* **139**:65-74.
- Wood, J. L., L. H. Dietterich, D. R. Leasure, S. C. Jantzi, T. R. Maddox, S. J. Wenger, J. W. Skaggs, A. D. Rosemond, and M. C. Freeman. 2024. Elemental composition and potential toxicity of the riverine macrophyte *Podostemum ceratophyllum* Michx. reflects land use in eastern North America. *Science of The Total Environment* **954**:176118.
- Wood, J. L., J. W. Skaggs, C. Conn, and M. C. Freeman. 2019. Water velocity regulates macro-consumer herbivory on the benthic macrophyte *Podostemum ceratophyllum* Michx. *Freshwater Biology* **64**:2037-2045.
- Wood, K. A., R. A. Stillman, R. T. Clarke, F. Daunt, and M. T. O'Hare. 2018. Water velocity limits the temporal extent of herbivore effects on aquatic plants in a lowland river. *Hydrobiologia* **812**:45-55.