# RIPARIAN LINKS AND NITROGEN SINKS HW RIPARIAN CONNECTIVITY AND INVASIVE SPECIES EFFECT NITROGEN CYCLING IN URBAN RIPARIAN ZONES

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Abstract. Riparian ecosystems are important nutrient sinks that are useful in preventing excessive nitrogen loading into aquatic ecosystems. The primary mechanism of N removal in riparian ecosystems is denitrification, an anaerobic microbial process that converts inorganic nitrate into N gas. The process is carried out primarily by heterotrophic bacteria, and is optimized in the oxygen poor and carbon rich environments that are characteristic of wetland and riparian ecosystems. Unfortunately, increasing urbanization has lead to a suite of degrading effects in riparian ecosystems including hydraulic disconnection, erosion, drier soils and increased vulnerability to exotic plant invasions. Two prominent invasive plants found in urban riparian zones are Aliliaria petiolata and Microstigium vimineum, herbaceous C3 and C4 plants, respectively, that are known to alter soil chemistry and composition through various secondary effects. The objective of this study was to determine how hydrologic disconnection and the presence of A. petiolata and M. vimineum affect the ability of urban riparian ecosystems to function as N sinks. Soil samples collected from urban riparian zones in Baltimore were analyzed for denitrification potential, potential net N mineralization and nitrification, soil moisture, and ion concentrations. Denitrification was found to be significantly higher in the hydraulically connected DR2 site compared to the more disconnected HHB1 site and the strongly eroded and incised DR5 site. Differences in denitrification potential were strongly correlated to differences in soil nitrate pools across sites. Differences in soil nitrate pools were not the result of differences in internal N-cycling across sites, and were likely the result of varying exposure to nitrate rich stream water. Thus, it appears that more connected riparian sites have a greater potential to function as N-sinks. Among plant species, denitrification potential was found to be significantly lower in soils beneath M. vimineum than in soils beneath A. petiolata and S. foetidus. The lower denitrification potential in M. viminum colonized soils may be the result of *M. vimineum* altering N cycling process within the soil through the release of allelopathic chemicals or high levels of labile carbon. However, it is also possible that M. vimineum simply colonizes soils which have inherently lower nitrogen cycling and denitrification potential.

#### INTRODUCTION

Riparian zones provide a wide range of ecosystem services that play an important role in maintaining aquatic ecosystem health and function (Jones, 2010). By forming terrestrial-aquatic junctions, riparian zones have the potential to function as "buffer zones" that reduce loading of pollutants into aquatic systems (Vidon, 2010).

Due to changes in land use, nitrogen (N) loading into aquatic systems has been increasing drastically over recent decades (Galloway et. al, 2008; Vitousek *et. al*, 1997) and poses a threat to both human and environmental health (Townsend et. al, 2003, Davidson et al. 2012). Nitrate, (NO<sub>3</sub><sup>-</sup>), the most common and mobile form of reactive N, is a prominent water pollutant that is poisonous to humans in high concentrations (Ward *et. al*, 2005). Excessive N loading into coastal waters also results in eutrophication-increased algae growth which results in anoxic zones and the death of aquatic organisms (Turner and Rabalais, 1994; Ryther and Dunstan, 1971). The ability of riparian zones to function as N sinks is hence important to conserving the health of aquatic ecosystems.

The primary mechanism for N removal in riparian ecosystems is denitrification; an anaerobic microbial process that converts  $NO_3^-$  and nitrite  $(NO_2^-)$  to N gas (Seitzinger *et. al*, 2006). As the most mobile form of N,  $NO_3^-$  is the primary concern to drinking water and eutrophication (Nolan and Stoner 2000). The process of denitrification is facilitated primarily by facultative anaerobes - organisms that, in the absence of oxygen, can use N oxides as electron acceptors during respiration (Groffman *et. al*, 1999). The most abundant denitrifying bacteria are heterotrophic, meaning they use organic carbon as a source of energy (Knowles, 1982). Denitrification is hence controlled by N, oxygen and carbon concentrations in the soil, and is optimized in wet, high organic C, anaerobic conditions (Decamps *et. al*, 2009).

Riparian zones are considered to be potential "hot spots" for denitrification in the landscape because they are often wet and anaerobic, with high levels of soil carbon and they intercept  $NO_3^-$  moving from upland land uses (Groffman *et. al*, 1998; McClain *et. al*, 2003). Realization of this potential is largely dependent upon hydrologic connectivity of surface and subsurface water flow to and from aquatic ecosystems (Racchetti *et. al.* 2010). In order for denitrification to occur,  $NO_3^-$  from runoff and other sources must interact with the anaerobic, high carbon soils characteristic of riparian ecosystems.

Unfortunately, urbanization has resulted in riparian zone degradation and disconnection through various effects collectively referred to as the Urban Stream Syndrome (Walsh *et. al*, 2005). Effects include increased incision and erosion due to runoff from impervious soils and lowered water tables due to decreases in infiltration (Paul, 2001). Riparian zone disconnection has several effects on riparian ecosystems that ultimately results in lower potentials for denitrification. Due to incision, flowpaths from uplands towards the stream are deepened and therefore don't interact with the wet, mucky, anaerobic soil that has the highest denitrification potential (Groffman *et. al*, 2002). Over time, these wet, mucky soils become drier and as a result lose organic carbon, are more aerobic and as a result their denitrification potential decreases (Groffman *et. al*, 2003). Additionally, because of changes in soil composition resulting from decreases in riparian connectivity, riparian biodiversity and plant community structures change to more closely resemble upland flora and fauna (Groffman *et. al*, 2003, Bain *et. a.*, 2012).

Degradation associated with urbanization has also made urban riparian zones more susceptible to plant invasions (Ehrenfeld 2003). Two common plant invaders of riparian zones found throughout the entire northeastern megalopolis are the C3 herbaceous plant *Alliaria petiolata* and the C4 grass *Microstigium vimineum*. Studies have determined that *A. petiolata* can alter nutrient cycling processes in soils by influencing leaf litter decomposition rates (Vikki *et. al*, 2008), disrupting mycorrhizal symbioses with plants (Roberts and Anderson, 2001), and by inhibiting the growth of native plant seedlings (Stinson *et. al*, 2006), likely preventing native plants from influencing soil chemistry. *M. vimineum* has been shown to reduce inorganic N levels presence within wetland soils compared to native species possibly by enhanced denitrification rates (DeMeester and Richter, 2010). No research has determined how invasive plants such as *A. petiolata* and *M. vimineum* may influence denitrification rates in riparian zones.

Previous studies have demonstrated the ability of invasive plants to influence nutrient composition, oxygen levels and organic carbon availability within soils. Angeloni *et. al* (2006) found higher concentrations of soluble ammonium,  $NO_3^-$  and phosphate in wetlands invaded by the invasive *Typha glauca*, suggesting that the invasion reduced the ability of wetlands to function as a nutrient sink. Additionally, denitrifers were found to be less abundant in the invaded wetlands (Angeloni *et. al*, 2006), which could result in reduced denitrification rates.

In order to better understand the effects of urbanization on riparian zones' ability to function as N sinks; I examined denitrification potentials of riparian zones as a function of varying levels of hydraulic connectivity and in the presence of *A. petiolata and M. vimineum*. I hypothesized that as riparian connectivity decreases, denitrification rates will also decrease. Along this gradient, I also hoped to find a way to quantify how connected riparian areas are to adjacent streams by comparing chloride

concentrations in the streams and soils. Additionally, because invasive plants are likely more prominent in disturbed areas (Lake and Leishmen, 2004) I investigated the effects of two prominent riparian plant invaders, *A. petiolata and M. vimineum*, on N cycling within riparian soils. Because both invasive plant species have been observed to either disrupt microbial processes or alter nutrient cycling processes in the soil, I hypothesized that denitrification potentials in soils beneath *A. petiolata* and *M. vimineum* would be lower than in soils taken from beneath native riparian plant species.

### METHODS

### Sites

Four riparian sites were chosen for sampling in Baltimore, MD USA based on visual evidence of their level of connectivity or disconnection. Of the four sites, one was clearly disconnected, one clearly connected, and two were of intermediate connectivity. Sites were located in the Horsehead Branch (HHB) and Dead Run (DR) watersheds that are subwatersheds of the Gwynns Falls, the main study watershed for the Baltimore urban long-term ecological research (LTER) project, the Baltimore Ecosystem Study (BES). Analysis of high resolution topographic maps and field reconnaissance was used to locate the gradient of sites. Onsite measurements of stream width (bank to bank) and depth (at deepest point) were made using surveying equipment.

In addition to riparian sites, floodplain wetlands at one of the HHB sites were sampled for native vs. invasive plant influenced soils; native skunk cabbage, *Symplocarpus foetidus*, vs. A. Petiolata and M. vimineum. Field reconnaissance was used to locate these sites. To control for differences in soil moisture within the site, each M. vimineum and A. petiolata patch was paired with a neighboring S. foetidus patch for comparison.

#### Site Sampling

For the connectivity comparisons, 1 meter soil cores were taken 5 meters from either side of the stream and at each of the four sampling locations (eight cores total). For the plant comparisons, soil samples (0 - 10 cm) were taken from beneath each plant species using a hand trowel. All samples were labeled and stored at 4 °C until transported from Baltimore to Millbrook.

A water sample was also collected from each stream at each of the four different sites for analysis of nitrate, chloride and sulfide concentrations. These values were compared with levels of these ions in soils to develop an index of connectivity, i.e. sites where soil and stream concentrations are similar were considered to be "connected" (Sudduth *et. al*, 2013).

Soil cores were divided into four depths (0-10cm, 10-30cm, 30-70cm, and 70-100cm). Each section was homogenized to remove roots and rocks and was then weighed and stored in plastic bags to yield a total of 32 riparian soil samples. Wetland samples taken from beneath the native and invasive plant species were also homogenized and stored in plastic bags for a total of 8 wetland samples.

### Soil Moisture and Organic Carbon

Subsamples of each of the 40 total soil samples were weighed into crucibles of known mass and placed in a drying oven for 48 hours at  $60^{\circ}$  C. After this drying period, the soil and crucibles were weighed, and the initial and final masses were compared to determine water loss. The samples were then placed back into the oven overnight at 100 ° C to remove any additional water and the dry soil masses were weighed (McInnes *et. al*, 1994). After the second drying period and mass measurements, the samples were left in the drying oven over night at 500 ° C to determine organic matter by loss upon ignition.

#### Soil Extractions

Inorganic N (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>) was extracted from soils with 2M KCL and analyzed colorometrically with a flow injection analyzer. Chloride, NO<sub>3</sub><sup>-</sup> and sulfate were extracted with water and analyzed by ion chromatography (Tabatabai and Dick, 1983). Stream water samples were also analyzed for inorganic N, chloride, nitrate and sulfate by ion chromatography.

### Denitrification Potential

Denitrification Potential was determined using the short-term denitrification enzyme anaerobic slurry assay (DEA) developed by Smith and Tiedje (1997) as described by Groffman et. (1999). Samples were amended with  $NO_3^-$  (100mg N/kg), dextrose (40 mg/kg), and chloramphenicol (10 mg/kg) and incubated under shaken, anaerobic conditions for 90 minutes. Gas samples were taken at 30 and 90 min, stored in evacuated glass tubes, and analyzed for N<sub>2</sub>O by electron capture gas chromatography.

#### Potential net N mineralization

Soils were incubated in 946 mL "mason" jars with lids fitted with septa to allow for gas sampling for a 10 day period to provide an estimate of potential net N mineralization and nitrification and microbial respiration. Inorganic N was extracted and analyzed as described above. Potential net N mineralization was calculated from the accumulation of inorganic N ( $NH_4^+$  plus  $NO_3^-$ ) over the course of the incubation and nitrification was calculated from the accumulation of  $NO_3^-$  alone (Robertson *et. al*, 1999). Gas samples were taken after the incubation period and analyzed for carbon dioxide ( $CO_2$ ) by gas chromatography. Microbial respiration was calculated from the accumulation of  $CO_2$  over the course of the incubation.

#### Statistical Analysis

Differences in DEA, potential net N mineralization and nitrification, soil respiration, ion concentrations, soil moisture and soil organic matter content among riparian sites were assessed using two-way analysis of variance with site and depth as main effects, with interactions. Differences among sites with different vegetation were assessed with one-way analysis of variance. Duncan's multiple comparisons test was used to assess specific differences between sites and depths. Relationships between variables were assessed with correlation analysis.

#### RESULTS

#### Connectivity

We hypothesized that ratios of soil to stream chloride concentrations and stream width to depth ratios would increase with increasing connectivity between streams and riparian zones. However, stream to soil chloride ratios showed no relationship to obvious visual riparian connectivity (Table 1). The most heavily degraded and clearly disconnected site, DR5, had the greatest soil: stream chloride ratio  $(1.43 \pm 0.01 \text{ mean} \pm \text{SD}, \text{N=8})$  whereas DR2, a site with little incision and obvious connectivity to the adjacent stream had the lowest soil: stream chloride ratio  $(0.41 \pm 0.07)$ . Soil to stream NO<sub>3</sub><sup>-</sup> and sulfate ratios also showed no relationship to riparian site connectivity, nor did comparisons of soil: stream chloride ratios to stream width/depth ratios (Figure 1).

Denitrification potential and related variables in riparian sites

Denitrification potential (Figure 2A) was significantly higher in surface (0 - 10 cm) soils of the DR2 riparian site than in soils from both the HHB1 and DR5 ( $\alpha < 0.1$ , N=2) sites. Microbial respiration was significantly higher in DR2 and DR5 soils than in HHB soils (Figure 1B,  $\alpha < 0.1$ , N=2). All other variables associated with N cycling and availability showed no significant differences among sites. Soil organic matter and soil moisture showed no significant differences among sites (Figures 2C, 2D respectively). Soil moisture and soil organic matter were higher in the HHB sites than the DR sites (Figures 2D and 2C, respectively). Potential net nitrification (Figure 2E), soil NH<sub>4</sub><sup>+</sup> pools (Figure 2G), and potential net mineralization (Figure 2F) also showed no significant differences among sites. Potential net nitrification was highest in HHB1 (Figure 2E) and potential net mineralization (Figure 2F) was higher in the HHB sites than in the DR sites. Total inorganic N was higher in HHB1 and DR2 than in HHB2 and DR5, but not significantly. Soil NO<sub>3</sub><sup>-</sup> (Figure 2H) was higher in DR2 than all other sites (not significant), showing very similar patterns to denitrification potential.

All variables related to N cycling and availability decreased with increasing soil depth. Comparing soil depths for all sites collectively, denitrification potential was significantly higher in the top 0-10 cm of soil than all other depths (Figure 3A,  $\alpha < 0.1$ , N=8). Soil moisture and soil NO<sub>3</sub><sup>-</sup> pools were also significantly higher in the 0-10 cm soil depth than all other depths (Figures 3B and 3C, respectively,  $\alpha < 0.1$ , N=8). Soil NH<sub>4</sub><sup>+</sup> pools (Figure 3D), potential net mineralization (Figure 3E) and nitrification (Figure 3F), and total soil inorganic N pools (Figure 3G) were significantly higher in the 0-10 cm depth than in the 30-70 cm and 70-100cm depths ( $\alpha < 0.1$ , N=8). Soil organic matter (Figure 3H) was significantly higher in the 0-10 cm depth than all other depths ( $\alpha < 0.1$ , N=8).

Significant correlations were found between DEA and respiration (r = 0.61, p=.0003), NO<sub>3</sub><sup>-</sup> (Figure 4A, r=0.83, p<.0001), total inorganic N (r=0.79, p<.001), soil organic matter (Figure 3B, r=0.55, p=0.0018) and soil moisture (Figure 4c, r=0.75, p<0.001).

#### Denitrification potential and related variables in floodplain wetlands with native versus exotic vegetation

Denitrification potential (DEA Figure 5A) was significantly lower in soils colonized by the invasive exotic plant *M. vimineum* than in soils beneath the invasive exotic *A. petiolata* and the native *S. foetidus* ( $\alpha < 0.1$ , N=2 for *M. vimineum* and *A. petiolata*, N=4 for *S. foetidus*). In general, variables related to N cycling and availability were lower under *M. vimineum*. Potential net N mineralization and nitrification rates (Figures 5B and 5C) were lower (statistically significant only for mineralization) in soils beneath *M. vimineum* than in soils beneath *A. petiolata* and *S. foetidus* ( $\alpha < 0.1$ , N=8 for each test). Soil NO<sub>3</sub><sup>-</sup> pools (Figure 5D) were lowest under *M. vimineum* (although not significantly) and soil NH<sub>4</sub><sup>+</sup> pools (Figure 5E) were significantly higher under *M. vimineum*.

*A. petiolata* showed significantly higher respiration than did *S. foetida* (Figure 6F). *M. vimineum* had the lowest soil moisture and organic matter content but these differences were not statistically significant (Figures 6G and 6H).

Among the plant comparison samples, significant correlations were found between DEA and soil  $NO_3^-$  (Figure 5A, r=0.91, p=0.0019), soil organic matter ( r=0.69, p=0.0602) and soil moisture (r=0.86, p=0.0082).

#### DISCUSSION

### Denitrification potential and related variables in riparian sites

Our results suggest that denitrification potential in riparian areas may be controlled by hydrologic connectivity between streams and riparian soils. The most obviously connected site, DR2 had significantly higher denitrification potential than the eroded, incised, and highly disconnected DR5 site as well as the less obviously degraded HHB sites.

The observed differences in denitrification potential could be the result of differences in soil moisture, soil organic matter, or soil  $NO_3^-$  pools across sites (Groffman *et. al*, 2002). Soil moisture affects the amount of available oxygen within sites; drier soils are more aerobic and hence less suitable for denitrification to occur (Seitzinger *et al*, 2006). Similarly, because carbon is a source of energy for denitrifying bacteria, greater amounts of soil organic matter can support a larger community of denitrifying bacteria, allowing for greater denitrification potential. We did not observe any differences across sites for either soil moisture or soil organic matter, therefore it does not appear that these factors caused the higher denitrification at the connected DR2 site relative to the other sites. Consistent with previous studies (Groffman and Crawford, 2003; Bettez and Groffman, 2012), we observed strong correlations between denitrification potential and both soil moisture and soil organic carbon matter. While these factors clearly influence variation denitrification potential among the samples we took, they did not vary systematically with site and therefore do not account for the significant differences that we observed between DR2 and the other sites.

Results suggest that differences in soil  $NO_3^-$  caused the site differences observed in this study. Soil  $NO_3^-$  pools were highest in DR2, the site with the highest denitrification potential. Moreover, a strong correlation was found between denitrification potential and  $NO_3^-$  supporting the idea that the observed differences in denitrification potential across sites were likely due to differences in soil  $NO_3^-$ .

Differences in soil  $NO_3^-$  pools across sites could be the result of differences in internal N cycling and  $NO_3^-$  production across the four sites. However, there was no significant variation in either potential net N mineralization or potential net N nitrification across sites (Figure1F and 1E). Both potential net nitrification and mineralization were very weakly correlated to denitrification potential. These results indicate that the differences in soil  $NO_3^-$  pools across sites were not the result of differences in internal N cycling processes and suggest that the sites must have been affected (or not) by an external source of  $NO_3^-$ 

The most likely external source of  $NO_3^-$  for these riparian sites was from adjoining streams. Other studies have suggested that waterways contaminated by agricultural and urban runoff may act as sources of  $NO_3^-$ , accounting for differences in soil  $NO_3^-$  pools across sites (Bettez and Groffman, 2012). High  $NO_3^$ concentrations were found within the streams of all four riparian sites (Table 1). HHB1 had the highest  $NO_3^-$  concentrations, meaning that if all sites were equally exposed to their adjoining streams, HHB1 would have the highest soil  $NO_3^-$  pools, which is not the case. Rather, the amount of  $NO_3^-$  that riparian soils receive from the streams depends on how connected the stream is to these riparian soils. More connected riparian zones would interact more with their adjoining stream and accumulate higher concentrations of  $NO_3^-$  within their soils. This reasoning is consistent with our results where DR2, the most obviously connected site, had the highest soil  $NO_3^-$  pools and denitrification potential. Conversely, DR5 had much lower nitrate pools and denitrification potential than DR2, due to the site being greatly incised and disconnected from its adjacent stream.

Denitrification potential and  $NO_3^-$  pools were also found to drastically decrease with increasing depth. Such trends have been observed in other studies (Groffman *et. al*, 2002; Gift *et. al*, 2010) that suggest that denitrification potential is highest in the uppermost organic layer of the soil. If differences in denitrification potential are in fact being controlled by stream  $NO_3^-$ , this means that even slight incision and disconnection of riparian soils from stream water would result in a substantial decrease in denitrification potential. This is because even slight incision would prevent the upper most layers of the soil from receiving the  $NO_3^-$  from the streams that is driving denitrification within these sites. These results suggest that the potential for riparian zones to function as N sinks depends greatly on the level of connection between riparian soils and stream water. To maximize the ability of riparian zones to function as denitrification hotspots that successfully remove N pollutants from the environment, riparian restoration projects should focus on maximizing riparian connectivity to streams, specifically by finding ways to prevent erosion and incision within riparian zones.

### Denitrification potential and related variables in floodplain wetlands with native versus exotic vegetation

Denitrification potential was significantly lower under *M. vimineum* than under both *A. petiolata* and *S. fetidus*. This appeared to be driven by differences in N cycling beneath each plant species. Soils beneath *M. vimineum* had lower potential net N mineralization and nitrification rates, accounting for lower soil  $NO_3^-$  pools beneath *M. vimineum*. Soil  $NO_3^-$  pools were also strongly correlated to denitrification potential. Soils beneath *M. vimineum* also had the lowest soil moisture, but these differences were not significant. Soil moisture was strongly correlated to denitrification potential.

One possibility that explains the lower denitrification potential in soils beneath *M. vimineum* compared to *A. petiolata* and *S. foetidus* is simply that *M. vimineum* more aggressively colonizes drier, N poor sites. Drier soils are more aerobic resulting in lower rates of potential denitrification. Low soil moisture can also reduce rates of  $NO_3^-$  production and in turn reduce denitrification potential. However, an alternative explanation is that *M. vimineum* is consuming more water and N that the other species, causes soils to be drier and have low levels of available N and denitrification.

The low N cycling in soils beneath *M. vimineum* could also be the result of *M. vimineum* releasing allelopathic chemicals that inhibit nitrification. Many studies have found evidence that allelopathic plants, many of which are invasive, can prevent denitrification by inhibiting nitrifying bacteria through allelopathy (Lodhi and Killingbeck 1980, Thibault *et. al*, 1982). Allelopathic chemicals emitted by *M. vimineum* have not yet been identified, but research has shown *M. vimineum* to inhibit germination of plant seedlings and alter soil microbial communities, suggesting that this species does release these compounds (Pisula and Meiners 2010, (Kourtev *et. al*, 2002).

Further research is required to conclude whether lower denitrification potential beneath *M. vimineum* is the result of *M. vimineum* actively altering N cycling processes within the soil through allelopathy or by aggressive uptake of water and N or if it is simply colonizing soils that have inherently low denitrification potentials due to dryness and low soil  $NO_3$  pools. A controlled experiment could be used to evaluate how *M. vimineum*, compared to both *A. petiolata* an *S. foetidus*, alters microbial communities within soil and in effect alters N cycling processes.

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#### APPENDIX

**TABLE 1.** Ratio of soil:stream concentrations of chloride, nitrate and sulfate and stream nitrate concentrations for each of the four riparian sites.

Site	Connected	Chloride Ratio	Nitrate Ratio	Sulfate Ratio	Stream Nitrate Concentration (mg-N/L)
HHB	Yes	1.32 <u>+</u> 0.35	0.40 <u>+</u> 0.18	0.37 <u>+</u> 0.07	4.2
HHB	No	0.73 <u>+</u> 0.12	0.59 <u>+</u> 0.26	1.37 <u>+</u> 0.25	1.1
DR2	Yes	0.41 <u>+</u> 0.07	3.04 <u>+</u> 1.50	1.11 <u>+</u> 0.18	1.8
DR5	No	1.43 <u>+</u> 0.01	0.79 <u>+</u> 0.47	1.00 <u>+</u> 0.26	2.1



**FIGURE 1.** Stream to soil chloride ratio as a function of stream width/ depth ratio (m). Increasing width/ depth ratio indicates increasing connectivity. N= 8 for each sample.

















**FIGURE 2.** Denitrification potential (A), microbial respiration (B), Soil organic matter (C) soil moisture (D) potential net nitrification (E) potential net mineralization (F) soil NH4+ pools (G), soil NO3- pools (H) in surface soils (0 - 10 cm) of four riparian sites in Baltimore, MD sampled in June 2014. Different letter indicate significant differences among sites. N=2 for each site.

















**FIGURE 3.** Denitrification potential (A), soil moisture (B), soil NO3- pools (C), soil NH4+ pools (D) potential net mineralization (E) potential net nitrification (F) total soil inorganic N pools (G) and soil organic matter (H) at four depths in riparian soils in Baltimore, MD sampled in June 2014. Values are means of two samples per each of four sites within either the Horsehead Branch or Dead Run watersheds. Different letters indicate significant difference between depths. N=32 for each depth.







**FIGURE 4.** Correlations of denitrification potential (DEA) and soil nitrate pools (A) organic matter content (B) and soil moisture (C) across all sites. Correlation coefficient (r) and probability (p) indicated on graphs. N=32.

















**FIGURE 5.** Denitrification potential (A), potential net mineralization (B), potential net nitrification (C), soil  $NO_3^-$  pools (D), soil  $NH_4^+$  pools (E), microbial respiration (F), soil moisture (G) and soil organic matter (H) for soils beneath three different plant species. Different letters indicate significant difference between species. N=2 for *A. petiolata* and *M. vimineum*, N=4 for *S. foetidus*.



**FIGURE 6**. Correlations between denitrification potential (DEA) and soil NO<sub>3</sub><sup>-</sup> pools (A), soil organic matter (B) and soil moisture (C) across all plant species. N=8.