



Aquatic Invertebrate Monitoring at Agate Fossil Beds National Monument

2010 Annual Report

Natural Resource Technical Report NPS/NGPN/NRTR—2012/654



ON THE COVER

Niobrara River, Agate Fossil Beds National Monument

Photograph by: Lusha Tronstad, Wyoming Natural Diversity Database

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Introduction

Aquatic invertebrates are excellent indicators of ecosystem quality, and these animals have been used to monitor ecosystems since the 1870s (Cairns and Pratt 1993). Growth, survival, and reproduction of aquatic invertebrates are sensitive to declines in environmental quality allowing analysis of assemblage structure to monitor lakes, streams, and wetlands (Johnson et al. 1993). Aquatic invertebrates are ideal indicator species, because they are ubiquitous, abundant, relatively long-lived, diverse, and typically sedentary (Rosenberg and Resh 1993b). In contrast, water samples can be directly analyzed for suspected pollutants, but such samples are snapshots of water quality only and can easily miss discrete discharges of pollutants or other undesirable conditions. In addition, analyzing water samples can be costly, making aquatic invertebrates a cost-effective alternative. Aquatic invertebrates integrate ecosystem quality throughout their lives, and much research has focused on how pollution alters assemblages (e.g., Rosenberg and Resh 1993a).

Choosing the best method of collecting aquatic invertebrates can be difficult, especially because there are many different devices available (Merritt et al. 2008). The ecosystem type (e.g., stream, lake or wetland), substrate type (e.g., gravel, clay, bedrock), vegetation (e.g., rooted macrophytes, floating macrophytes, periphyton), type of aquatic invertebrates targeted (e.g., benthic, hyporheic, neustic, emerging adults), and study question should all be considered when deciding on what sampler to use. Whether to collect data quantitatively (statistically rigorous information; e.g., Hess sampler), semi-quantitatively (sampling area defined; e.g., kick net), or qualitatively (estimate presence and absence of taxa; e.g., dip net; Merritt et al. 2008) depends upon the research question. All sampling devices have advantages and disadvantages; therefore, choosing a sampler that minimizes sampling bias in a given ecosystem is vital.

The National Park Service has been monitoring fish at Agate Fossil Beds National Monument since 1979 (Stasiak 1990). Over that time, ecosystem quality declined. One explanation for the decline is the invasion of yellow flag iris (*Iris pseudacorus*) (Bowles 2010; Stasiak et al. in prep). Yellow flag iris probably slows water velocity and increases organic matter in the stream leading to large daily swings in dissolved oxygen concentrations. Another explanation for the decline in ecosystem quality is the introduction of invasive northern pike (*Esox lucius*) in the Niobrara River (Stasiak et al. in prep). Pike are piscivores and likely reduced the fish assemblage from 11 species to 3 species between 1989 and 2011. Stasiak et al. (in prep) speculated that pike currently feed on crayfish, because other fish are scarce in the river. Introducing pike may have caused a trophic cascade that changed the abundance and assemblage of invertebrates in the Niobrara River.

Hester-Dendy multi-plate samplers have been used to monitor aquatic invertebrates in the Niobrara River since 1989; however, the National Park Service would like to consider other sampling methods. In the Niobrara River, the Hester-Dendy plates imitate rooted macrophyte leaves and aquatic invertebrates colonize the plates for 30 days before being collected. These samplers can be effective, but have limitations. In the Niobrara River, large debris dams form upstream of the samplers, bias the invertebrates collected and introduce greater variability (Figure 1). Therefore, my objective was to compare Hester-Dendy, Hess, and core samples collected at 3 sites along the Niobrara River at Agate Fossil Beds National Monument. My specific questions were: 1) How does the number of individuals collected from each sampler

compare? 2) How do the bioassessment metrics compare among samplers? and 3) Do these samplers collect similar taxa?

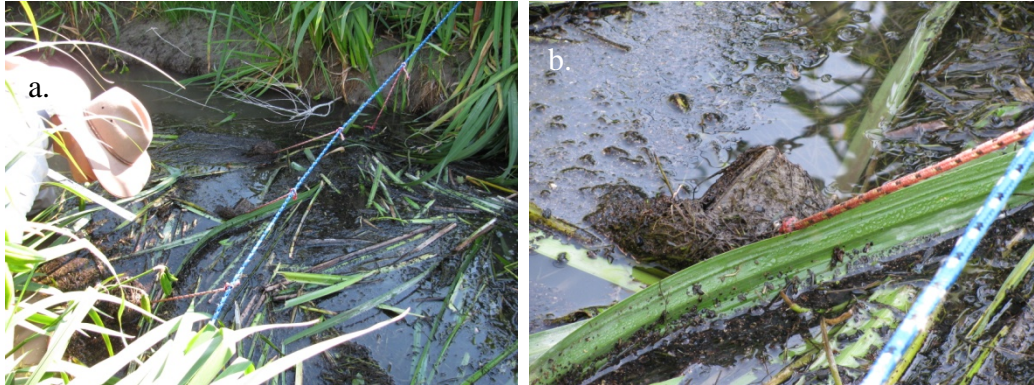


Figure 1. a. Debris dam caused by the Hester-Dendy samplers. b. The debris dam likely increases variability in samples, such as the sampler shown that was pushed out of the water by the debris dam. Photos by Marcia Wilson.

Study Area

The headwaters of the Niobrara River are located around Lusk, Wyoming, and flow eastward into Nebraska and eventually to the Missouri River near Niobrara, Nebraska. The Niobrara River Basin covers 32,600 km², of which the majority is grassland in northern Nebraska (Galat et al. 2005). Over 95% of the land within the basin is used for cattle and crop production. The Niobrara River flows through Agate Fossil Beds National Monument in western Nebraska about 23 km from the Wyoming border. At this point the Niobrara River is a low order stream flowing through grassland. Agate Fossil Beds National Monument includes about 1,100 ha in a valley bottom and about 18 km of river that flows through the 6 km wide monument (Figure 2). The riparian vegetation in the Monument is dominated by cattails (*Typha* sp.) and the invasive yellow flag iris. The substrate in the river consists of fine particles (e.g., sand). Pike, white suckers (*Catostomus commersonii*), and green sunfish (*Lepomis cyanellus*) currently inhabit the river within the Monument (Stasiak et al. in prep).

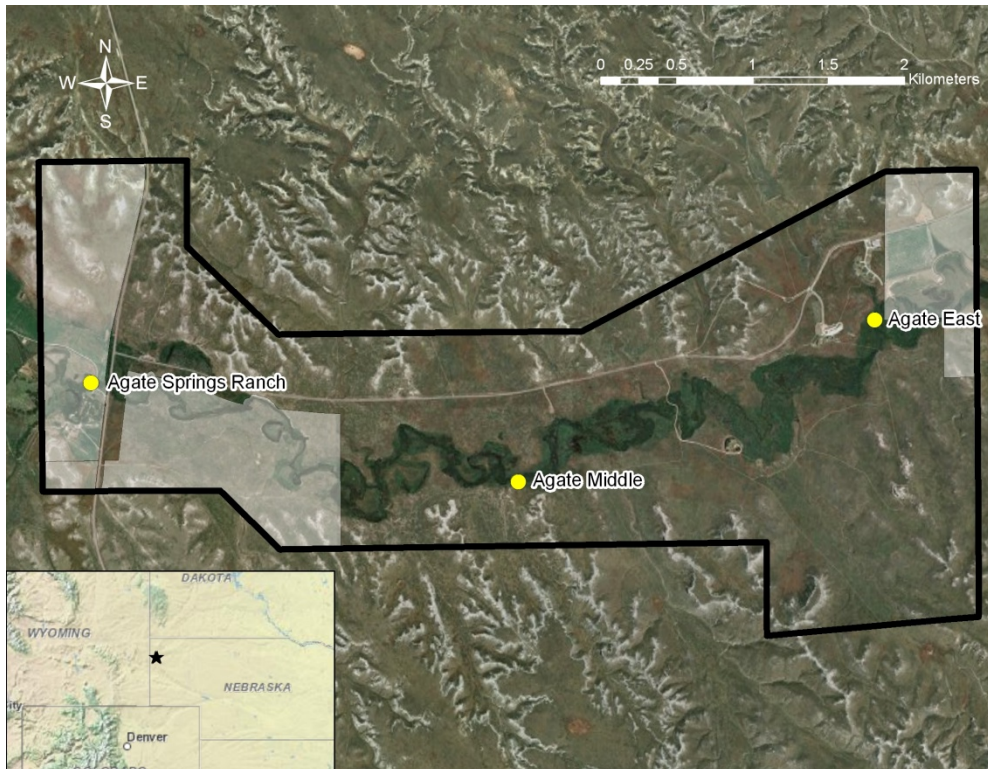


Figure 2. We sampled three sites along the Niobrara River at Agate Fossil Beds National Monument. The black line is the Monument boundary and the transparent white areas are private land within the Monument boundary. The inset map shows the location of Agate Fossil Beds National Monument (star).

I sampled 3 sites along the Niobrara River (Figure 2). The most upstream site (Agate Springs Ranch) is located near the west Monument boundary. Agate Springs Ranch has an overstory of plains cottonwood (*Populus deltoides*), and cattails are more abundant than iris (Figure 3a). The middle site, Agate Middle, is shallower and lacks an overstory (Figure 3b). Both iris and cattails are abundant here. Finally, Agate East, the site located before the Niobrara River flows out of the Monument, is the deepest site (Figure 3c). The vegetation is dominated by iris with a few willow (*Salix* spp.).

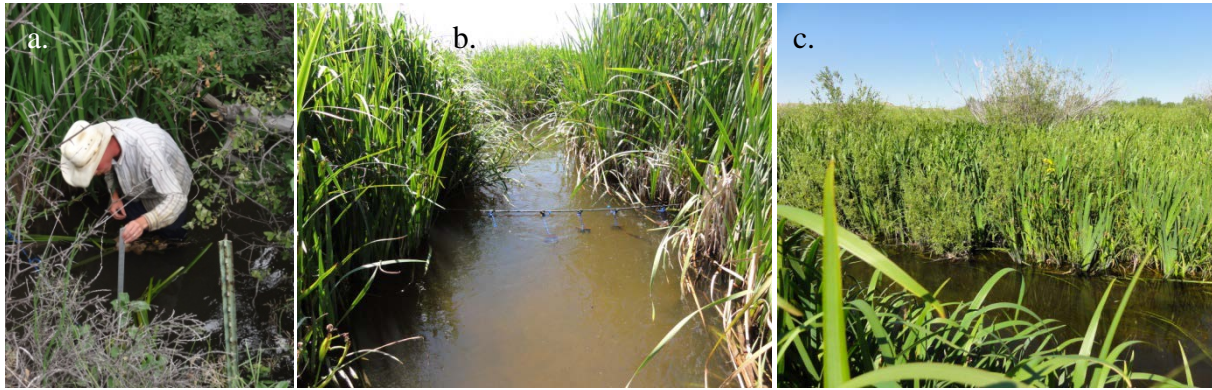


Figure 3. Photos of a. Agate Springs Ranch, b. Agate Middle, and c. Agate East. Location information located in Table 1.

Methods

General Measurements

To assess the general habitat characteristics of the Niobrara River I measured several features including water quality, turbidity, sediment composition, and depth. I measured dissolved oxygen (percent saturation and mg/L), pH, water temperature, specific conductivity, and oxidation-reduction potential using a Yellow Springs Instruments Professional Plus. The probe was calibrated on site before use. I measured turbidity by estimating the depth at which a Secchi disk disappeared from sight. The composition of sediment was estimated by sampling sediment across the width of the stream and selecting the percent class for clay, silt, sand, gravel, cobble, boulder, bedrock, and hardpan/shale on a scale of 0 -7 (0 = none, 1 = trace, 2 = 1-5%, 3 = 5-25%, 4 = 25-50%, 5 = 50-75%, 6 = 75-95% and 7 = 95-100%; Peterson et al. 1999). Clay was defined as fine particles forming a ribbon after removing water, whereas silt did not form a ribbon. Sand was particles 0.06-2 mm in diameter, gravel was 2-64 mm in diameter, cobble was 64-256 mm in diameter, boulder was 256-4000 mm in diameter, bedrock was >4000 mm in diameter, and hardpan/shale was firm, consolidated fine substrate. I measured the depth of the water with a meter stick (3.2 mm width) parallel and perpendicular to flow across the width of the stream. By subtracting the two measurements, I calculated an estimate of water velocity (vertical displacement); the greater the vertical displacement of the water, the higher the water velocity. Finally, I recorded the location and elevation of each site using a global positioning system (GPS; Garmin eTrex Vista HCx).

Hester-Dendy Samples

I deployed seven Hester-Dendy multi-plate samplers (76 mm by 76 mm, 9 plates, Wildlife Supply Company) at each site on 15 July 2010. Five Hester-Dendy samplers were used for analysis and two extra samplers were deployed in case any were lost. I strung a rope across the stream between two permanent posts and tied seven loops to separate the Hester-Dendy samplers. From each loop, I tied another rope with the Hester-Dendy samplers hanging about 0.3 meters below to allow for a drop in water level. I retrieved the samplers on 16 and 17 August 2010 by approaching the site from downstream and placing a dip net (212 μ m mesh) under each sampler. Hester-Dendy samplers were immediately placed in a container with 80% ethanol, and any organisms in the dip net were removed and placed in the same container. After returning to the laboratory, I dismantled the Hester-Dendy samplers to remove invertebrates that colonized the plates, rinsed samples using a 212 μ m sieve, and preserved samples in 80% ethanol. Replicates two through six were typically used for analysis.

Hess Samples

To sample invertebrates that live in the emergent vegetation that is abundant along the Niobrara River, I collected three Hess samples (500 μ m mesh, 860 cm² sampling area, Wildlife Supply Company) from each site on 16 and 17 August 2010. I placed the Hess sampler over cattails and yellow flag iris to collect invertebrates that lived on the vegetation and in the surrounding sediment. The vegetation and sediment were vigorously agitated using our hands and a brush, and the samples were preserved in 80% ethanol.

Core Samples

To collect invertebrates that live in the sediment of the Niobrara River, I took three core samples (Hand corer, 16.6 cm² sampling area, Wildlife Supply Company) from each site. Sediment was sieved through 212 µm mesh to remove fine particles and the samples were preserved in 80% ethanol.

Invertebrate Analysis

Invertebrates were sorted from the debris and identified to genus (Insecta, Turbellaria, Isopoda, and Amphipoda), family (Decapoda, Pelecypoda (Bivalvia), Gastropoda), class (Annelida, Acarina) or phylum (Nematoda) with two exceptions (order: Collembola, family: Diptera) according to Peterson et al. (1999). If invertebrates were extremely numerous (>500 individuals) in any sample, I subsampled as follows. First, I removed the large and rare invertebrates, then I subsampled the remaining invertebrates. Invertebrates were identified under a dissecting microscope using Merritt et al. (2008) for insects, and Thorp and Covich (2010) and Smith (2001) for non-insect invertebrates.

Several bioassessment metrics have been calculated since 1989 to estimate stream quality based on the invertebrates collected: Hilsenhoff Biotic Index (HBI), Ephemeroptera, Plecoptera, and Trichoptera (EPT) richness, number of EPT taxa divided by the total number of taxa collected, taxa diversity (Shannon index), taxa richness, and taxa evenness (Bowles 2010). To distinguish among sites, I used ANOVA to compare abundance (per sampler) and bioassessment metrics for each sampler (DataDesk6.1). To evaluate sampling devices, I used ANOVA to compare abundance (per sampler) and each of these six bioassessment metrics among sampler types. Differences among samplers and sites were distinguished using Bonferroni multiple comparison tests. Differences were significant when $p < 0.017$ ($0.05/3$; where I had three samplers and three sites). Finally, I used cluster analysis to estimate the degree to which the taxonomic composition of samples varied by sampler type (PC-ORD version 6).

Results

Water quality was similar between deploying and retrieving the Hester-Dendy samplers. In general, Agate Springs Ranch had the highest dissolved oxygen concentrations and Agate East had the lowest concentrations (Tables 1 and 2). All dissolved oxygen concentrations were collected mid-morning, except Agate Springs Ranch and Agate Middle on 16 August 2010, which were collected in the afternoon. pH was highest at Agate Springs Ranch on both dates. Specific conductivity increased from Agate Springs Ranch to Agate East. Reducing conditions (<200 mV; oxidation-reduction potential) appeared to dominate in the Niobrara River. At all sites, I could view the Secchi disk at the bottom of the stream. Stream width was consistent among sites; however, the Niobrara River was widest at Agate Springs Ranch and narrowest at Agate Middle. The water reached into the wetland vegetation at both Agate Middle and Agate East when I collected samples. Agate East was the deepest site and Agate Middle was the shallowest (Table 3). Stream discharge was highest at Agate Middle and lowest at Agate East. Overall, the substrate in the Niobrara River at each site was dominated by sand. At Agate Springs Ranch, I estimated that 1-5% was silt and 95-100% was sand. At Agate Middle, 25-50% was sand, 50-75% was gravel, and there was a trace of silt. Agate East was composed of 5-25% silt and 75-95% sand.

Table 1. Location (NAD83) and elevation of each site along the Niobrara River. Water quality data collected on 15 July 2010 when the Hester-Dendy samplers were deployed.

Site description	Agate Springs Ranch	Agate Middle	Agate East
Easting	599323	602143	604495
Northing	4697497	4696844	4697913
Elevation (m)	1354	1350	1343
DO (% saturation)	90	67	57
DO (mg/L)	6.8	5.2	4.5
pH	8.18	7.78	7.6
Water temperature (°C)	21.2	19.9	19
Specific conductivity (µS/cm)	417.1	451.2	455.6
ORP (mV)	182.1	171.6	219.8

Table 2. Water quality, air temperature, and stream width collected on 16-17 August 2010 when all the invertebrate samples were collected.

Site description	Agate Springs Ranch	Agate Middle	Agate East
DO (% saturation)	98	93	81
DO (mg/L)	7.6	7.3	6.7
pH	8.15	7.88	7.99
Water temperature (°C)	19.2	19.1	16.9
Specific conductivity (µS/cm)	419.5	423.7	428.5
ORP (mV)	188.7	45	107.6
Secchi disk depth (cm)	>64	>60	>97
Air temperature (°C)	27.5	19	28
Stream width (m)			
Open water	2.18	1.9	2.1
In vegetation, south	0	0.36	0.23
In vegetation, north	0	0.74	0.33

Table 3. Stream depth (cm) behind each Hester-Dendy sampler. Sampler 1 was on the south side of the Niobrara River and sampler 7 was on the north side of the river. Parallel depth is the actual water depth. Vertical displacement is an index of stream discharge, where larger numbers indicate higher flows. See methods for more details.

Sampler #	1	2	3	4	5	6	7	Mean
Agate Springs Ranch								
Parallel depth	53	60	64	63	62	59	54	59
Perpendicular depth	53	62	65	70	72	59	54	62
Verticle displacement	0	2	1	7	10	0	0	3
Agate Middle								
Parallel depth	56	58	60	59	56	52	40	54
Perpendicular depth	56	76	82	77	70	55	41	65
Verticle displacement	0	18	22	18	14	3	1	11
Agate East								
Parallel depth	77	89	93	97	96	99	96	92
Perpendicular depth	77	91	93	100	103	101	97	95
Verticle displacement	0	2	0	3	7	2	1	2

I identified 37 invertebrate taxa using Hester-Dendy multi-plate samplers in the Niobrara River, from four phylum (Nematoda, Annelida, Mollusca, and Arthropoda). Overall, Diptera, Ephemeroptera, Crustacea, and Mollusca were the most numerous groups collected in order of decreasing abundance. Hester-Dendy samplers from Agate East (1,987 ind/sampler) contained the most invertebrates and Agate Springs Ranch (420 ind/sampler) had the fewest ($F = 5.6$, $df = 2$, $p = 0.019$). Taxa diversity was lowest at Agate Springs Ranch (Bonferroni: $p < 0.005$), and

similar between Agate Middle and Agate East (Figure 4a; $F = 11.9$, $df = 2$, $p = 0.0014$). Agate East had the highest taxa richness (Figure 4b; $F = 12.57$, $df = 2$, $p = 0.0011$; Bonferroni: $p < 0.004$), and Agate Middle had higher taxa evenness than Agate Springs Ranch ($F = 12.3$, $df = 2$, $p = 0.0025$; Bonferroni: $p = 0.002$). I collected three EPT taxa at both Agate Springs Ranch and Agate East, and three EPT taxa at Agate Middle. Agate Springs Ranch contained the highest proportion of EPT taxa. The average tolerance value for invertebrates was higher at Agate East and Agate Middle, and much lower at Agate Springs Ranch ($F = 105.6$, $df = 2$, $p < 0.0001$; Bonferroni: $p < 0.0001$).

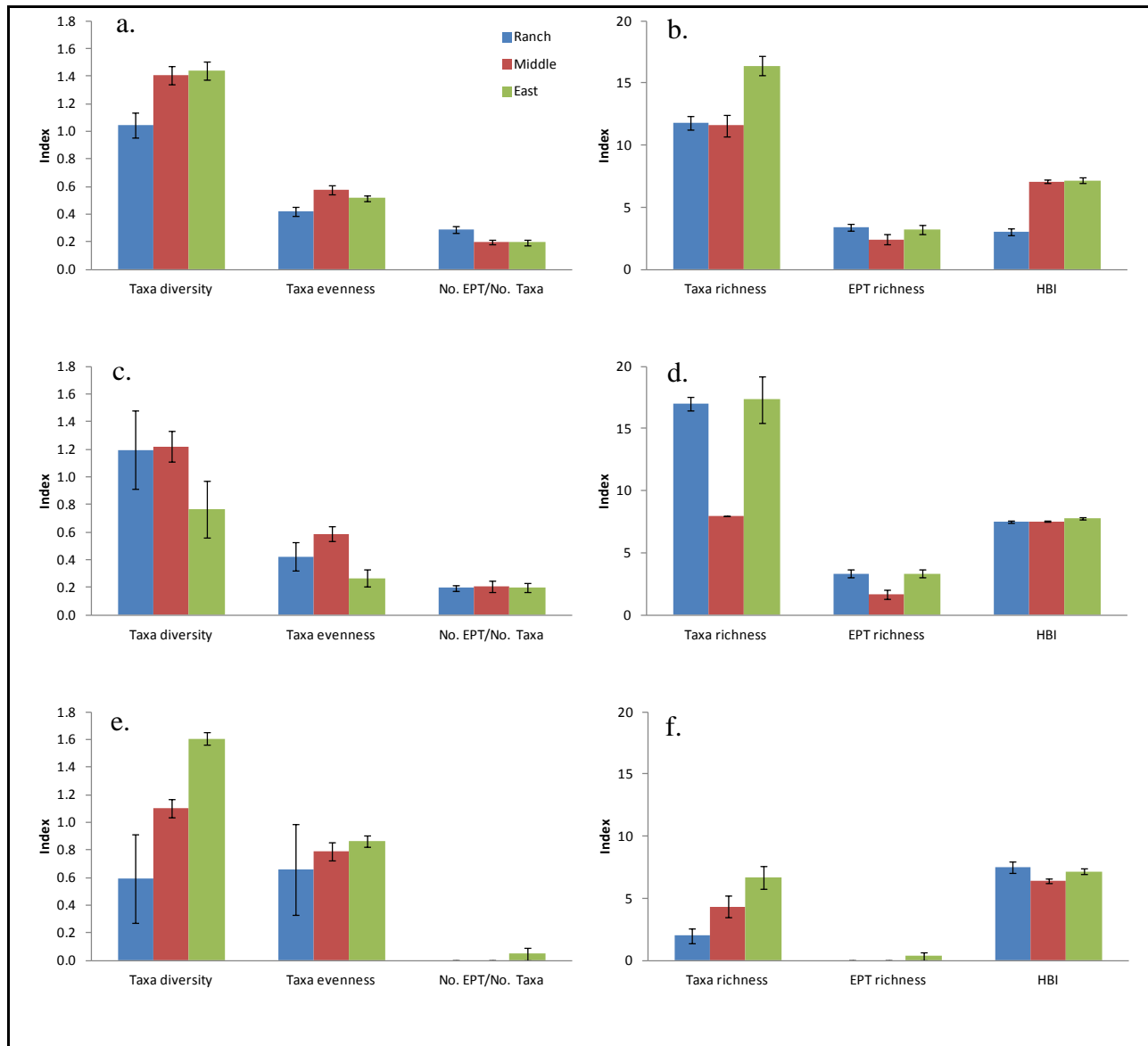


Figure 4. Invertebrate bioassessment metrics for three sites along the Niobrara River collected with Hester-Dendy multi-plate sampler (a, b), Hess sampler (c, d), and corer (e, f). Higher values for taxa diversity, taxa evenness, number of EPT taxa/number of taxa, taxa richness, and EPT richness indicate better ecosystem quality, while lower values of HBI indicate higher ecosystem quality. Error bars are standard errors.

I collected 34 taxa of invertebrates from four phylum using a Hess sampler in the Niobrara River. Overall, Diptera, Mollusca, Odonata, and Ephemeroptera were the most numerous invertebrates in decreasing order of abundance. Agate East had the highest density of invertebrates (2,710 ind/sampler) and Agate Middle had the lowest density (152 ind/sampler; $F = 6.4$, $df = 2$, $p = 0.033$; Bonferroni: $p > 0.05$). Taxa diversity was similar among sites (Figure 4c; $F = 1.4$, $df = 2$, $p = 0.31$). I collected the same number of taxa at Agate Springs Ranch and

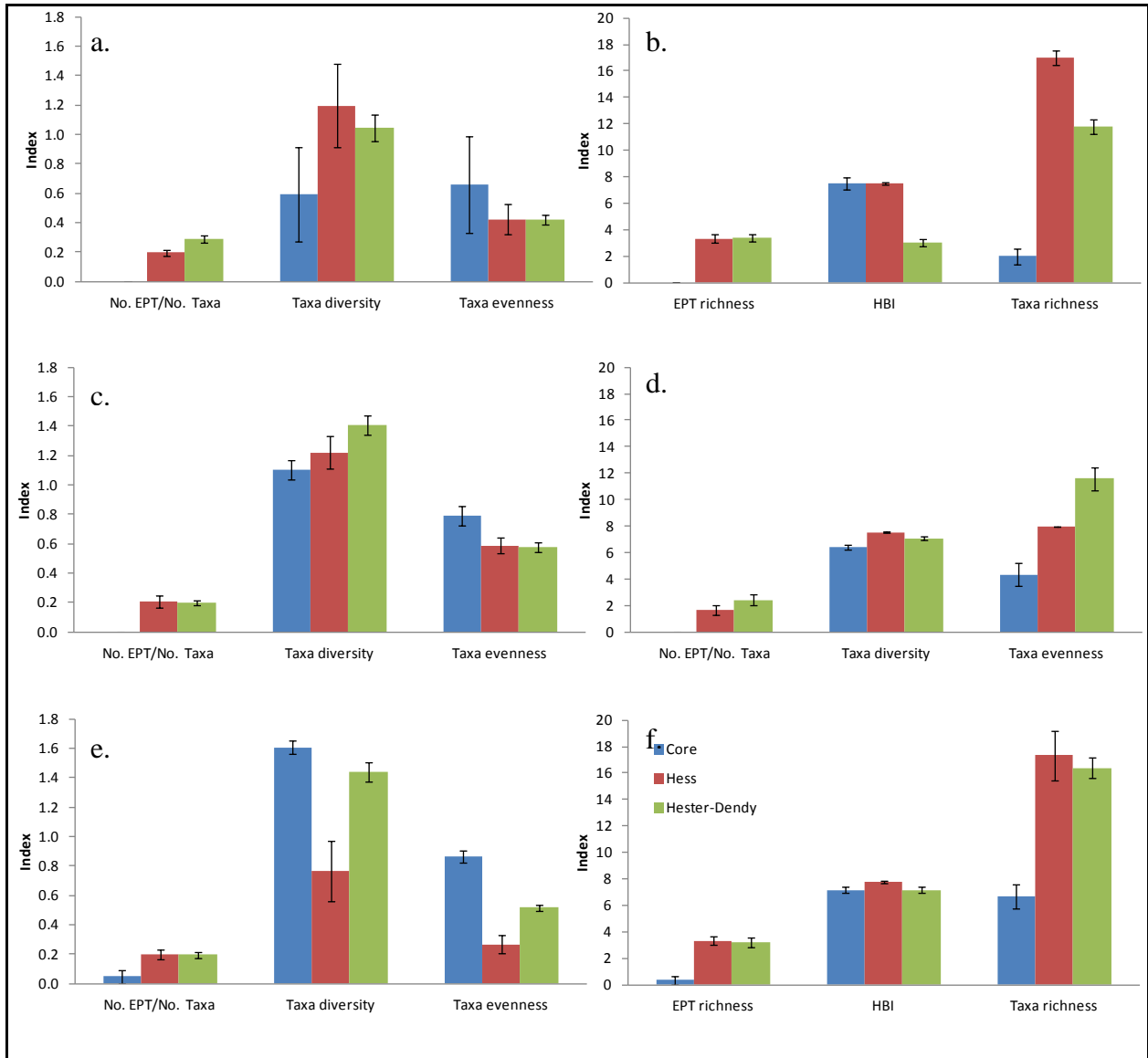


Figure 5. Invertebrate bioassessment metrics for three sites along the Niobrara River, Agate Springs Ranch (a, b), Agate Middle (c, d), and Agate East (e, f), collected with core, Hess sampler, and Hester-Dendy multi-plate sampler. Higher values for taxa diversity, taxa evenness, number of EPT taxa/number of taxa, taxa richness, and EPT richness indicate better ecosystem quality, while lower values of HBI indicate higher ecosystem quality. Error bars are standard errors.

Agate East, and half the number of taxa at Agate Middle (Figure 4d; $F = 22.3$, $df = 2$, $p = 0.0017$; Bonferroni: $p < 0.004$). Taxa evenness was highest at Agate Middle and lowest at Agate East, but values were not statistically significant among sites ($F = 4.5$, $df = 2$, $p = 0.064$). I collected three EPT taxa at Agate Springs Ranch and Agate East, and two EPT taxa at Agate Middle. The proportion of EPT taxa ($F = 0.047$, $df = 2$, $p = 0.95$) and mean tolerance values ($F = 4$, $df = 2$, $p = 0.079$) were similar among sites.

I collected 14 taxa using a sediment corer in the Niobrara River. Overall, Diptera, Crustacea, and Mollusca were the most numerous groups in decreasing order of abundance. Agate East had the highest density of invertebrates (28 ind/sample) and Agate Springs Ranch had the lowest abundance (5 ind/sample). Taxa diversity and evenness were similar among sites (Figure 4e). Taxa richness, EPT richness, and the proportion of EPT taxa were highest at Agate East (Figure 4f), but not statistically different among sites ($p > 0.05$). Finally, mean tolerance values for taxa were similar among sites ($F = 3.5$, $df = 2$, $p = 0.10$).

In general, Hester-Dendy and Hess samplers collected similar samples, but core samples typically differed (Figure 5). The total number of individuals collected per sample was similar between Hester-Dendy and Hess samples ($F = 5.19$, $df = 2$, $p = 0.012$, Bonferroni: $p = 0.999$), but core samples collected fewer individuals than Hester-Dendy samples (Bonferroni: $p = 0.017$). All samplers collected a similar number of Mollusca, Crustacea, and Coleoptera ($p > 0.05$), but Hester-Dendy samplers collected more Ephemeroptera (mean values; Hester-Dendy = 146, Hess = 17, and core = 0.1, Bonferroni < 0.017) and Diptera (mean values; Hester-Dendy = 182, Hess = 50, and core = 6.4, Bonferroni < 0.017) per sample than Hess or core samples ($F = 9.38$ and 14.26 , $df = 2$, $p = 0.008$ and < 0.0001 , respectively). The assemblage of taxa did not vary by the sampler, as cluster analysis did not group samples by sampler type, with the exception of core samples (Figure 6). However, Hester-Dendy and Hess samplers were intermixed throughout the tree.

Bioassessment metrics were similar between Hester-Dendy and Hess samples, but values differed for core samples. Taxa diversity was similar among samplers, but taxa richness, taxa evenness, EPT richness, HBI, and EPT taxa/total taxa differed among samplers. Core samples produced different values for taxa richness ($F = 43.57$, $df = 2$, $p < 0.0001$), taxa evenness ($F = 8.2$, $df = 2$, $p = 0.0016$), EPT richness ($F = 58.56$, $df = 2$, $p < 0.0001$), and EPT taxa/total taxa ($F = 72.7$, $df = 2$, $p < 0.0001$) compared to Hester-Dendy and Hess samplers (Bonferroni: $p < 0.017$). However, HBI values differed between Hester-Dendy (mean = 5.5) and Hess (mean = 7.8) samplers ($F = 12.28$, $df = 2$, $p = 0.0001$, Bonferroni: $p < 0.017$). The lower values for Hester-Dendy samplers may be from a large number of *Paraleptophebia* (tolerance value = 1.2) collected with Hester-Dendy samplers at Agate Springs Ranch site.

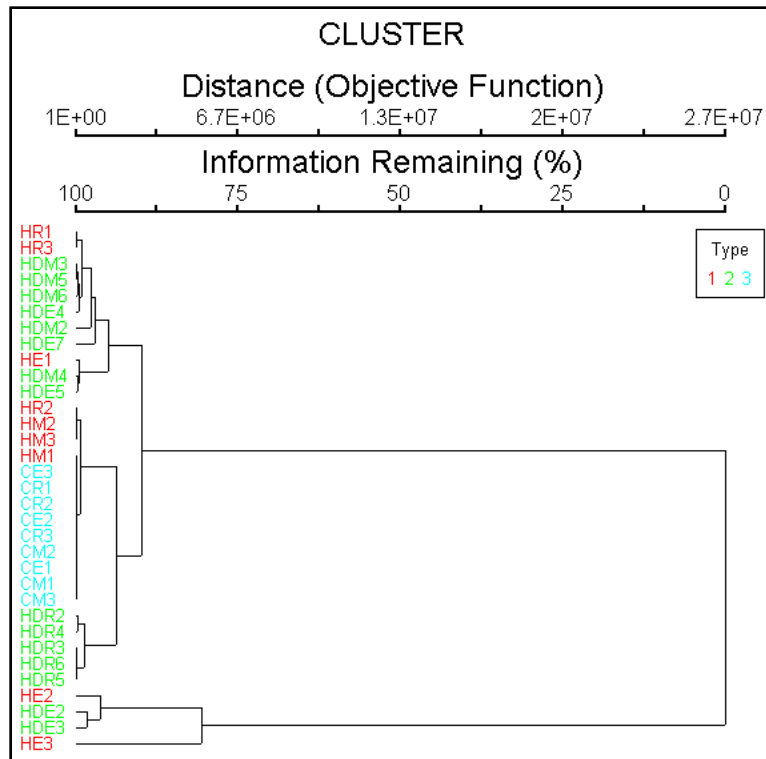


Figure 6. Cluster analysis of the taxonomic composition of invertebrates, as sampled by different devices. Hester-Dendy (green), Hess (red), and core (blue) samplers collected similar taxa.

Discussion

Prairie streams can be difficult to sample for aquatic invertebrates. Prairie streams often have fine substrates, yet most quantitative aquatic invertebrate samplers are designed for gravel or cobble bottom streams (e.g., Surber sampler). Therefore, alternative sampling methods are necessary. Core samplers have the advantage of directly targeting fine substrates; however, cores typically sample a small area and the samples can be rather time intensive to sort. Additionally, fewer invertebrates may live in the sediment, depending on conditions. Others have used semi-quantitative box samplers in prairie wetlands that consist of placing a 4-sided rectangle in the wetland and collecting invertebrates within the area using a dip net (Meyer and Whiles 2008). However, box samplers do not work well in deep habitats, streams, or areas with dense vegetation. Therefore, an appropriate sampling method often depends upon the conditions, habitat, and the question.

Another option for collecting aquatic invertebrates is using Hester-Dendy plates, which is an artificial substrate sampler. Hester-Dendy samplers provide solid substrate in habitats that may lack such areas. Additionally, these samplers may mimic snags or macrophytes which can be difficult to sample. Invertebrate density is typically calculated based on the surface area of the plates; however, surface area in natural habitats (e.g., surface area of macrophyte leaves) is seldom calculated and would be extremely labor intensive. Therefore, density or biomass of aquatic invertebrates collected with Hester-Dendy samplers can only be compared to aquatic invertebrates from other ecosystems that were collected using the same method. In the Niobrara River, macrophytes in the wetland area provide abundant substrate for aquatic invertebrates. The wetland area differs from the main channel in that the wetland area can be large (0.4 km wide in places) and water velocity is significantly slower. Hester-Dendy samplers were placed in the main channel of the stream where water velocities were higher causing large debris dams to form. Debris dams were periodically cleared from the Hester-Dendy samplers; however, I have observed debris dams that were >0.3 m (>1 ft) deep and >1.8 m (>6 ft) in length when I retrieved the samplers (Figure 1). Because of these large debris dams, I collected species that normally would not be collected with a Hester-Dendy sampler, such as large crayfish (Decapods). Also, debris dams may cause higher variability in the samples because either more (including debris) or fewer invertebrates (removing debris may displace individuals) may be collected depending on how the samplers are gathered. I have also observed Hester-Dendy samplers pushed out of the water by debris dams (Figure 1b).

Hess samplers collect natural densities of aquatic invertebrates that are comparable to other quantitative methods among ecosystems (per m² of stream bottom). Hess samplers are traditionally used similarly to Surber samplers, but they have the advantage of surrounding the sampling area. I chose to use a Hess sampler to collect aquatic invertebrates in the Niobrara River, because I could sample the macrophytes and sediments to estimate natural densities. I slipped the Hess sampler over the macrophytes at the edge of the main channel, and scoured the vegetation and sediment. Therefore, I sampled invertebrates that lived in both habitats (vegetation and sediment) and that had multiple habits (e.g., crawlers, clingers, etc.). Hess samplers have shortfalls too; for example, Hess samplers cannot be used in deep water. However, modifications can be made to minimize bias.

In general, I collected similar aquatic invertebrates using Hester-Dendy and Hess samplers, but core samples contained less diversity. Hester-Dendy samplers contained a few taxa that I did not collect in Hess samples. For example, I found low densities of Hirudinea (leeches), *Heptagenia* (mayfly), Anisoptera (dragonflies), and *Polycentropus* (caddisfly) in Hester-Dendy samples, but these taxa did not appear in Hess samples. Based on the habits of these taxa, I would expect Hess samples to collect them. However, these are rarer taxa and I collected fewer Hess samples than Hester-Dendy samples, which may explain why I collected greater diversity in the latter. Additionally, I collected ostracods in Hester-Dendy samples. Ostracods are small crustaceans that probably flowed through the 500 μm mesh of the Hess sampler, but were retained in the 212 μm mesh of the sieve for the Hester-Dendy samplers. Conversely, I collected Stratiomyidae (true fly) only in the Hess samples. Stratiomyidae appeared to be rare, which explains why I did not collect them in other samplers. Finally, I collected far less diversity in the core samples, but I only collected *Corbicula* using the core sampler at the East site.

Using core and Hess samplers together, the overall density of invertebrates in the Niobrara could be calculated given the available habitat in each area. In August, the stream channel was between 63 and 100% of the habitat available to aquatic insects, while only 0 to 37% of water was in the wetland vegetation. Therefore, invertebrates that can live in the sediments (collected with a core sampler) had the greatest area to inhabit. A much larger area of the riparian habitat is flooded earlier in the year before water is withdrawn for irrigation. Floodplains can provide large areas for invertebrates to colonize and produce higher biomass compared to other habitats within the ecosystem (Benke 2001).

I recommend using a Hess sampler to collect aquatic invertebrates in the Niobrara River. Collecting invertebrates with a Hess sampler compared to a Hester-Dendy sampler will reduce the number of visits to the sites along the Niobrara River from potentially two (deploying, three visits to clear debris dams, and collecting) to only one (collecting). By collecting invertebrates on natural substrate there may be less variability in the samples, because of the difficulties using Hester-Dendy samplers in the Niobrara River. In general, Hester-Dendy and Hess samplers collected a similar number and assemblage of aquatic invertebrates in 2010. I do not recommend collecting core samples, because these samples contained significantly lower diversity and I collected the same taxa in Hess samples with the exception of rare *Corbicula*. Hess samples should be collected in early August, when water levels are slightly higher and more water extends into the riparian habitat, which will expedite sampling.

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