RECLAMATION

Managing Water in the West

IS DETECTION OF LONG-TERM IMPACTS USING AQUATIC MACROINVERTEBRATES SEASONALLY DEPENDENT?

Research and Development Office
Science and Technology Program
Final Report 2014-7851

Sampling in Las Vegas Wash, Nevada.

S.M. Nelson

Mission Statements

The U.S. Department of the Interior protects America's natural resources and heritage, honors our cultures and tribal communities, and supplies the energy to power our future.

The mission of the Bureau of Reclamation is to manage, develop, and protect water and related resources in an environmentally and economically sound manner in the interest of the American public.

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Aquatic invertebrates have been utilized as ecological indicators of short and long-term (pulse and press) disturbances to streams for many years. Pulse disturbance recovery of communities is rapid (< 18 months). Press disturbances, such as altered flows below a dam, are repetitive and persistent in nature, resulting in a permanent non-normative community adapted to these enduring environmental modifications. Theoretically, community sampling at any time of the year should allow for press impact detection. However, research has been inadequate in addressing this assumption. Seasonal sampling variability resulting in non-detection of impacts could be particularly important when data are used to determine whether expensive alterations are needed for environmental mitigation. The question is whether biomonitoring can detect anthropogenic environmental alterations despite community seasonality. This paper addresses these issues through literature review and direct analysis of a multi-year macroinvertebrate data base collected during different seasons.

Literature indicated that elimination of seasonality effects was analysis dependent. Ordination methods were seasonally sensitive, while metrics and predictive modeling were often robust to seasonal effects. Data analysis also indicated that sampling in a single season is sufficient for detection of long-term impacts such as those associated with stream restoration structures and hydrological alterations related to dams.

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Executive Summary

Aquatic invertebrates have been utilized as ecological indicators of impacts to streams for many years. Aquatic macroinvertebrate communities integrate stresses and provide a “biological memory” of a particular environment. Short-term impacts, because of the presence of rapid response sensitive taxa are dynamic, while long-term effects on macroinvertebrates may be represented by a fixed invariable community. Short and long-term disturbances are commonly termed pulse and press, where a pulse disturbance is of limited duration (e.g., a chemical spill), while press disturbances are longer in duration and often involve changes in the watershed or stream channel. Community recovery from pulse disturbances is typically rapid (< 18 months). Press disturbances, such as a wastewater inflow or altered flows below a dam, are of a continuous or repetitive nature and persist for a number of years, resulting in a stressor-adapted community. Recovery to a normative biota does not take place because environmental modifications are enduring. Theoretically, sampling the community at any time of the year should allow for detection of these press impacts. However, life histories of invertebrates that result in absence of taxa during certain seasons could make impact detection unpredictable and it may be that some sampling seasons are superior to others for monitoring. There has been inadequate research and discussion addressing the validity of relating macroinvertebrate community data collected on a single occasion to conditions at stream sites during different times of year. Seasonal sampling variability resulting in non-detection of differences could be particularly important when bioassessment data is used to decide the success of expensive environmental restoration projects or to determine whether re-operation of dams is needed for flow mitigation. Seasonal changes in communities could be a confounding factor that results in erroneous conclusions. The question is whether biomonitoring data can detect anthropogenic alterations to the environment despite community seasonality. This paper addresses these issues through a combination of literature review and analysis of a long-term macroinvertebrate data base collected during four different seasons over several years.

Literature indicated that elimination of seasonality effects was dependent upon the analysis used. Ordination methods were sensitive to seasonality while metrics and predictive modeling were often robust to seasonality effects. Data analyses indicated that sampling in a single season is sufficient for detection of long-term impacts such as those associated with stream restoration structures and hydrological alterations related to dams.

METHODS and RESULTS

Methods and results are presented in Appendix A as Technical Memorandum No. 86-68220-14-03.

Appendix A: Technical Memorandum No. 86-68220-14-03.

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Mission Statements

The mission of the Department of the Interior is to protect and provide access to our Nation's natural and cultural heritage and honor our trust responsibilities to Indian Tribes and our commitments to island communities.

The mission of the Bureau of Reclamation is to manage, develop, and protect water and related resources in an environmentally and economically sound manner in the interest of the American public.

Technical Memorandum No. 86-68220-14-03

IS DETECTION OF LONG-TERM IMPACTS USING AQUATIC MACROINVERTEBRATES SEASONALLY DEPENDENT?

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Abstract

Literature was reviewed for examination of the effect of sampling season on the ability of aquatic invertebrate communities to detect long-term impacts. An analysis of a multi-season and multi-year data base was also undertaken to determine whether detection of stream restoration activities was related to seasonality of aquatic invertebrate communities.

Elimination of seasonality effects was dependent upon the analysis used. Ordination methods were sensitive to seasonality while metrics and predictive modeling were often robust to seasonality effects.

Results indicate that sampling in a single season is sufficient for detection of long-term impacts such as those associated with stream restoration structures and hydrological alterations related to dams.

Introduction

Aquatic macroinvertebrate communities integrate stresses (Barbour et al., 1999) and provide a "biological memory" of a particular environment. Short-term impacts, because of the presence of rapid response sensitive taxa (e.g., Barbour et al., 1999; Fritz & Dodds, 2005) are dynamic, while long-term effects on macroinvertebrates may be represented by a fixed invariable community. Short and long-term disturbances are commonly termed pulse and press, where a pulse disturbance is of limited duration (e.g., a chemical spill), while press disturbances are longer in duration and often involve changes in the watershed or stream channel (Niemi et al., 1990). Community recovery from pulse disturbances is typically rapid (< 18 months) (Niemi et al., 1990). Press disturbances, such as a wastewater inflow or altered flows below a dam, are of a continuous or repetitive nature and persist for a number of years, resulting in a stressor-adapted community. Recovery to a normative biota does not take place because environmental modifications are enduring. Theoretically, sampling the community at any time of the year should allow for detection of these press impacts. However, life histories of invertebrates that result in absence of taxa during certain seasons could make impact detection unpredictable. Alvarez-Cabria et al. (2010) suggest that some sampling seasons are superior to others for monitoring. Cao & Hawkins (2011) also advise that biological assessments are affected by sampling season and deal with this variability by aggregating samples across seasons. There has been inadequate discussion addressing the validity of relating macroinvertebrate community data collected on a single occasion to conditions at stream sites during different times of year (e.g., Carlson et al. 2012). Seasonal sampling variability resulting in non-detection of differences could be particularly important when bioassessment data is used to decide the success of expensive environmental restoration projects or to determine whether re-operation of dams is needed for environmental purposes (e.g, Richter and Thomas, 2007). Seasonal changes in communities could be a confounding factor that results in erroneous conclusions (Clarke & Hering, 2006). The question is whether biomonitoring data can detect anthropogenic alterations to the environment despite community seasonality.

The Bureau of Reclamation (BR) uses aquatic macroinvertebrates for monitoring projects important to its mission. BR has interests in a large number of river restoration projects. These projects are typically related to water delivery, water salvage, or avoiding impacts to endangered species. Also of special interest to BR is the ability to detect environmental changes associated with hydrological alterations. Monitoring activities are largely used to ensure that BR can continue to supply users with water and power, often through a "trade" of restoration for water.

I examined issues of seasonality and detection of press impacts through a review of the relevant literature along with analyses of a macroinvertebrate data base from Las Vegas Wash (LW) that was collected for several years and in different seasons at variably altered stream restoration sites.

Methods

Literature review--This review was intended to summarize a body of literature and illustrate conclusions about the topic in question. Primary sources consisted of journal articles that were discovered using search terms like season, aquatic macroinvertebrates, variability, and impact. Articles that were obtained contained additional citations that were pertinent to the literature review. Literature was confined to temperate freshwater lotic systems.

Biomonitoring in Las Vegas Wash--Biomonitoring efforts using aquatic invertebrates have taken place in the LW watershed for several years to monitor on-going river restoration (Nelson, 2011) in this wastewater dominated system. Erosion control structures have been built, with 22 completed since 1999 for channel stabilization at headcut locations in LW. These structures are low height weirs used to help dissipate energy from large storm events. Along with these constructed weirs, stabilization of the channel bed has utilized bank protection and revegetation. Revegetation with woody plants included native species Fremont cottonwood (*Populus fremontii*) and willow (*Salix* spp.). Nelson (2011) used annually collected spring-time macroinvertebrates to verify effectiveness of restoration structures and determined that communities at reference, mainstem LW, and LW locations with structures differed from each other, with differences driven by hydrological, channel, and water quality characteristics.

Study area--Las Vegas Wash drains the Las Vegas metropolitan area and surrounding 1550 km^2 Las Vegas Valley, in the Mojave Desert of southern Nevada. Down-cutting and channelization of LW has occurred as wastewater discharges increased over time (Buckingham & Whitney, 2007).

Sampling at sites (Table 1) took place from 2004 to 2011 in March or April (spring), June (summer), August or September (roughly autumn), and December (winter) of each year. Sites included upstream reference sites (LW11.76 and LW11.1) above the influence of wastewater treatment plants (WWTP); mainstem sites below WWTP's where the channels were incised (mainstem-lotic) or broad with slow moving water (mainstem-lentic), and mainstem sites where structures were in place. Erosion control structures were located at sampling stations LW6.05, LW5.5, and LW3.85 (Table 1). While LW11.1 was sampled from the beginning of the monitoring program, LW11.76 was added in 2010 to gain additional spatial information. LW7.0 was also added in 2010 to provide an additional lentic water site. Along with biota, a variety of environmental variables were sampled.

Chemical, physical, and biological methods--Environmental variables measured for each site included water chemistry, physical parameters, and measurements of habitat qualities. Dissolved oxygen (DO), conductivity, pH, temperature, and turbidity were measured with portable meters. Water samples for alkalinity and hardness were analyzed using titration methods.

Size composition of the substrate was visually estimated at each site in the area where macroinvertebrates were collected. Categories were expressed as percent bedrock, boulders, cobble, coarse gravel, fine gravel, and sand/fines. Percentage categories were converted to a single substrate index (S.I.) value (e.g., Jowett & Richardson, 1990) using the formula S.I.=0.08* %bedrock + 0.07* %boulder + 0.06* %cobble +0.05* %gravel +0.04* %fine gravel + 0.03* %sand and fines. Stream wet width was measured with a measuring tape or a range finder. Depth was measured with a calibrated rod.

Water velocity at 10 cm above the substrate was measured at three discrete points in the channel cross-section within the invertebrate collection area. The average of these three measurements was used in analysis.

Habitat disturbance was estimated with Pfankuch's Index (Pfankuch, 1975). This subjective, composite index involves scoring 15 stream channel variables along the upper bank, lower bank, and stream bottom. Variables include estimates of plant density on the upper banks, the frequency of raw banks, and how much of the bottom is affected by scouring and deposition. High scores represent unstable channels at the reach scale. This index has been use to measure stream disturbance in other studies (Townsend et al., 1997).

Invertebrate sampling used a 1-minute kick method with a D-frame net (700-800 micron mesh) along a ca. 10-meter reach at each sampling site. Samples were preserved in 70% propanol. In the laboratory, samples were washed in a 600-micron mesh sieve to remove alcohol, invertebrates were picked from the substrate with the aid of an illuminated 10X magnifier, and then the entire sample was enumerated and identified under a binocular dissecting scope. Insect taxa were mostly identified to genus.

Mass of coarse particulate organic matter (CPOM) and plant matter related to autotrophic production (periphyton) was obtained from the macroinvertebrate sample. These samples were dried at 60^{o}C for 48 hrs and weighed to the nearest hundredth of a gram.

Data analysis--Experimental sites were not allocated to treatments and were not randomly interspersed throughout the area of consideration. Because landscape treatments were not assigned, these comparisons may be detecting something besides a difference in habitat and may be biased in a way that limits inferences. "Replicates" used in the site-type analysis were from different years and different sites. Perhaps the best description of this study is the "quasi-experiment" of Hargrove & Pickering (1992) where some level of pseudoreplication is acceptable in exchange for realism.

Ordination was used to examine patterns in macroinvertebrate data, and to identify variables associated with invertebrate distributions. Initial analyses of the macroinvertebrate data sets used detrended correspondence analysis (DCA), and revealed a data gradient length > 3, suggesting appropriateness of unimodal models for analysis. Therefore, canonical correspondence analysis (CCA) was used for direct gradient analyses. Faunal data were transformed (square root transformation) before analysis. Forward selection of environmental variables and Monte Carlo permutations were used to determine which and to what extent environmental variables exerted a significant ($P \leq 0.05$) effect on invertebrate distributions. If environmental variables were strongly correlated (Pearson correlation, $r \geq 0.6$), only a single variable was selected for use in CCA to avoid problems with multicollinearity. Environmental variables were normalized [(ln (X+1)) or arcsin squareroot transformation for percentage data] if the Shapiro-Wilks Test indicated non-normality. In the ordination diagram, taxa and sites are represented by points and the environmental variables by arrows. Arrows roughly orient in the direction of maximum variation of the given variable.

Factorial ANOVA was used to test for differences in invertebrate abundance [(ln (X+1)) transformed], taxa richness, and ETO (Ephemeroptera, Trichoptera, and Odonata) richness between site-types (reference, lentic, lotic, and sites restored with structures) and seasons. Differences between years were not examined and were not the focus of this study, but likely added variability to the analyses. I assumed that samples were independent, given that the most frequent sampling at any sites was seasonal. ETO taxa, along with Plecoptera, are often used in metrics used to describe water quality impacts in river systems (e.g., Ode et al., 2005; Smith et al., 2007). I omitted Plecoptera from the metric because of its absence, due to life history characteristics, from LW. The ETO metric is a tolerance metric, sensitive to riverine stressors. Significance was set at a P value of 0.05. If ANOVA detected a difference, Tukey’s test was used to compare means. Factorial ANOVA followed by Tukey’s test was also used to test for differences in environmental variables at different site-types. Data were transformed, if needed to normalize distributions, using ln (X+1). Data analyses with ANOVA, in this case, are limited in interpretation by pseudoreplication (Hurlbert, 1984).

Correspondence of metrics between seasons was also examined using Pearson correlations. Observations were grouped by site for the various seasons for correlation analysis.

Results

Literature review--Literature indicated that seasonal effects were quantified in a variety of ways. The majority of studies used ordination or metrics for analyses. A smaller number of papers examined the effects of season on predictive modeling (Table 2). Predictive modeling efforts often present biological data as Observed/Expected (O/E) taxa.

Las Vegas Wash variables--Environmental variables differed significantly between sites (Table 3). Reference sites differed in water quality from other sites and had significantly higher conductivity and lower water temperatures, along with higher hardness and alkalinity values. Sites below WWTPs had native water diluted by the addition of WWTP water which also increased water temperature. Significantly higher velocities and wider channels were associated with sites that had been physically restored (Table 3). High velocities were also measured at lotic sites. These sites differed from other site-types in having incised channels and significantly greater depths (Table 3). Food resources (CPOM and periphyton) for macroinvertebrates tended to be significantly higher at reference and structure sites (Table 3). Significant differences between seasons were mostly observed for water quality parameters; however, periphyton also differed between seasons (Table 3). Water temperature had a strong seasonal component and temperatures were lower in the winter compared to other seasons (5.0-23.1^{o}C in winter and 8.9-34.9^{o}C in other seasons).

Macroinvertebrate communities-Las Vegas Wash--A total of 91 taxa were detected in LW with most individuals' belonging to the Chironomidae family (Table 4). In the CCA ordination, sites were separated along Axis I by site-types and by season along Axis II (Figure 1). Eigenvalues for the first two axes were 0.408 and 0.191 with 51% of species-environment relation explained. Permutation tests (1000 permutations) for all canonical axes were significant (F-ratio = 5.248, *P*-value = 0.0010). Sites were separated into three main groupings with reference sites to the far right on Axis 1 and lotic restored and unrestored sites to the left on the first Axis. Unimproved lentic sites were between these two groupings. Reference sites were characterized by high conductivites (n=34, 3609 $\pm$ 60 µS/cm) when compared to restored sites with structures (n=84, 2397 $\pm$ 15 µS/cm) (Table 3). Some of the taxa in this portion of the diagram such as *Chironomus* and *Hyalella* are known to be tolerant of relatively high salinities (Galat et al., 1988). Substrate size, velocity, stream width, and temperature all tended to increase towards the negative portion of Axis 1. Velocities (Table 3) at restoration sites (n=84, 0.70$\pm$ 0.03 m/S) were, on average, much higher than those recorded from reference sites (n=34, 0.33 $\pm$ 0.0304 m/S), and especially higher than mainstem lentic sites (n=34, 0.14 $\pm$ 0.02 m/S). Impacted lotic and lentic sites were less variable in community composition between seasons, with a much smaller range along Axis II. A greater variety of taxa occurred at reference and structure-restoration site communities.

Axis II seemed to be characterized by seasonal characteristics, including changes in periphyton biomass (more in spring) (Figure 1) and invertebrate community composition (Figure 2). *Simulium*, for example, was most abundant in spring, while the mayfly *Camelobaetidius musseri* was never detected in spring but was found in all other seasons. There also appeared to be some separation of impacted lotic and restored lotic sites along Axis II.

Macroinvertebrate metrics-Las Vegas Wash--Factorial ANOVA indicated that richness, abundance, and ETO richness differed significantly with site- type (Table 5). The interaction term was non-significant in all cases, while season was significant only for ETO richness (Table 5). The absence of *Camelobaetidius musseri* from samples collected in spring resulted in a significant difference between spring collected samples and all other seasons. None of the other seasons differed in ETO richness amongst themselves.

Differences in site-type patterns, with all seasonal data combined, were fairly consistent for taxa richness, abundance, and ETO richness, with unimproved lentic and lotic sites having metrics that were significantly lower than reference and structure sites (Figure 3). While abundance did not differ between reference and structure sites, richness metrics did (Figure 3). Simple taxa richness was significantly higher at reference sites while the tolerance metric ETO richness was significantly higher at structure sites when these measures were compared between reference and structure sites (Figure 3).

When metrics for the various sites and years were segregated by season and then compared, taxa richness (r=0.5266 to 0.7775, $P \leq 0.0001$), abundance (ln transformed) (r=0.5690 to 0.7457, $P<0.0001$), and ETO richness (r=0.4876 to 0.6523, $P \leq 0.0001$) were significantly correlated between seasons. An example of data for taxa richness is presented in Figure 4.

Discussion

Literature review—Of the 12 articles that were reviewed, several appeared to identify stream impacts using macroinvertebrates collected in a variety of seasons where metrics were largely unaffected by season (Helms et al., 2009; Clarke et al. 2002; Johnson et al., 2012). In some cases, a subset of metrics detected environmental degradation and were unaffected by seasons (Álvarez-Cabria et al., 2010; this study). Some studies also characterized the “best” season (Álvarez-Cabria et al., 2010) for biomonitoring**.** Johnson et al. (2012) compared impaired (urban) and non-impaired (rural) aquatic invertebrate communities on a monthly basis. They found that metrics such as taxon richness and EPT richness discriminated consistently, no matter the season, between different types of sites. Seasonality did not confound interpretation of biological metrics in New Zealand streams and the co-authors explicitly stated that seasonal variability did not need to be considered (Stark and Phillips, 2009).

Analyses with ordination often identified differences in seasonal communities. Ordination was useful in identifying natural temporal variation in macroinvertebrate communities between seasons (Álvarez-Cabria et. al., 2010; Boulton et al., 1992; Šporka et al., 2006). In some studies, less seasonal variation occurred in impacted communities because impacted taxa had non-seasonal life cycles (e.g., Carlson et al., 2012). Ordination may identify patterns that are ancillary to the question at hand, while metrics seem to provide consistent discrimination of impacts. Species identity, however, could be of importance in explaining community reaction, and ordination along with metrics may allow for improved explanation of macroinvertebrate response to impacts or restoration. Leunda et al. (2009) observed community succession by seasons with ordination; however, a biotic index provided consistent identifications of water quality classes, despite seasonal variation. In the case of Leunda et al. (2009), however, only high quality sites were sampled. Leunda et al. (2009) makes the point that many environmental variables vary seasonally and these variables may be shaping specific members of communities. Šporka et al. (2006) suggest that it is advantageous to collect more than one sample in a year and that a late autumn or winter sample had complementary value to a spring sample in their study.

Predictive modeling results were sometimes skewed when reference material was collected on a date different from that of the initial monitoring. However, this would have no effect when sites sampled at an off-time are compared amongst themselves. No loss of sensitivity with single season sampling was detected by Hawkins et al. (2000). However, sampling should be restricted to a standard period to minimize the influence of phenological shifts in taxonomic composition. Reference sites used in predictive modeling are therefore usually confined to a specific time period and additional testing is also limited to the same time period. Predictive modeling was especially useful when impacts were compared over wide geographical areas. While invertebrate communities may differ in composition geographically, “scores” are calculated on the same scale, allowing for comparisons.

Clarke & Hering (2006) suggest that ignoring natural seasonal variability can confound anthropogenic impact detection and that sampling in more than one season increases the ability to estimate ecological status. Linke et al. (1999) found that habitat variable differences between sites outweighed the importance of season as a predictor for taxa richness. They, however, believed a model that contained seasonality increased degradation detection and suggested that constraining the sampling time frame may miss important variation in communities. As a result, Linke et al. (1999) recommended monitoring programs that sample sites in at least two seasons. Šporka et al. (2006), using ordination, found clear separations of samples by season that were caused by macroinvertebrate life cycles. They also found that some bioassessment metrics may differ seasonally. Seasonal abundance of food, such as CPOM and periphyton, influence life cycles and may be incorporated into monitoring. Carlson et al. (2012) found that macroinvertebrate taxonomic composition, analyzed with ordination, was significantly related to agricultural land use in the spring, but not autumn in their system. They indicated that insect emergence was a likely explanation for this discrepancy.

Seasonal effects on Las Vegas Wash macroinvertebrates--Macroinvertebrate ordination variability was mostly explained by physical and chemical variables related to site-type (reference, lotic, lentic, and sites with structures). Ordination appeared to separate sites based on water quality (conductivity) and substrate size. Less variability was explained by restoration activities and seasonality. Main seasonal difference in taxon composition was between spring-collected samples and all other seasons. Reference sites were separated in ordination space from all other site-types which were clumped together to some degree, although lentic sites were separated from restored and unimproved lotic sites.

Biological metrics consistently separated site-types relative to restoration projects, demonstrating clear differences between reference sites, sites rehabilitated with structures, and unimproved lentic and lotic sites. The only biological metric significantly affected by season in LW was ETO richness due to life history characteristics of specific taxa. Taxa richness and invertebrate abundance at site-types did not differ between seasons. Unrestored lentic and lotic sites had lowest metric values while values at reference and structure sites were highest. In LW the seasonal variability in metrics appears to be small relative to differences between site-types. Biological site metrics were highly correlated between seasons, suggesting that the differences between site-types were consistent between seasons. This study provides evidence that macroinvertebrate community data collected in a single season and used to identify differences between unrestored and restored sites is relevant to other seasons, at least in the case of persistent environmental alterations. Similar to earlier analyses (Nelson, 2011), macroinvertebrates responded positively (increased richness and abundance) to restoration erosion control structures placed in LW.

Documentation of flow/dam effects on aquatic macroinvertebrates— Discrimination of flow effects that occurred up to 6 months antecedent to macroinvertebrate sample collection were documented across several geographic areas (Dunbar et al., 2010a; Dunbar et al., 2010b). Rehn (2009) found lower macroinvertebrate metric scores below dams that were strongly associated with altered hydrologic regime. Carlisle et al. (2012) found biological condition (EPT taxa richness and O/E) of samples collected in September were strongly and negatively related to the severity of depleted flows in mean (n=10 years) March flows. Armitage et al. (1987) used prediction to examine aquatic macroinvertebrate families' response to river regulation. Certain families, such as Heptageniidae, occurred at lower abundance than predicted while some groups of chironomids and oligochaetes occurred at higher abundances than predicted. Lowest degree of seasonality is associated with more disturbed streams (Helms et al., 2009). In the case of hydrological alterations these taxa are often largely non-insects that lack aerial stages and life history seasonality (Nelson, 2009). Declines in metric values were associated with increased dam height in a multiple watershed study by Nelson (2009). Dam height represents a multitude of impacts which includes those related to temperature and thermal regime modification, sediment transport, hydraulic residence time, and water quality.

Conclusions

While there is a general belief that more data is better, economic constraints along with a basic desire for efficiency suggest limits to data collection. While some have recommended that multiple collections of macroinvertebrates for impact detection are desirable, there are numerous examples where this level of effort has not been expended and where impact detection has still succeeded. It did not appear that there were any large differences in the ability of macroinvertebrates collected in a given season to detect effects in LW and this has been documented in other cases. Seasonality in LW did not affect the ability to detect restoration effects with macroinvertebrate assemblages.

It is also likely that studies of hydrological alteration using single season collections of aquatic macroinvertebrates will have success. Literature indicates that macroinvertebrates collected in a given season can detect flow impacts from other seasons. A variety of characteristics of hydrologically altered streams probably leads to this result and includes:

1) Non-insect taxa are often an important metric in studies of river regulation and this type of community is non-seasonal in character,
2) Hydrological alterations studied are typically press disturbances which do not allow for recovery and variability in communities,
3) Hydrological alterations may impact specific taxa in a near universal manner. Heptageniid mayflies, for example, are often reduced in abundance below dams (Rader and Ward, 1988; Carlisle et al, 2012) while other taxa seem to consistently increase in abundance such as *Ephemerella* and *Hydroptila*. This response has been demonstrated below dams in North America (Rader and Ward, 1988; Munn and Brusven, 1991), Europe (Armitage, 2006), and Japan (Takao et al., 2008).

Detection of impacts is dependent upon analysis selection. Metrics and predictive modeling are most successful in eliminating taxa seasonality effects from analyses.

Of course, other issues may arise such as year-to-year variation at reference sites. However, recurrent collections at a subset of reference sites will provide estimates of correction factors for temporally similar samples. A pulse disturbance at a reference site would also be detected in this manner and identified as an outlier. Pulse disturbances at an impacted site may be less of a concern. In many cases the community associated with a press disturbance will likely be non-responsive since most sensitive taxa are already absent.

Literature and analyses of seasonal macroinvertebrate data indicate that seasonally constrained aquatic macroinvertebrate collections may be used to examine press disturbances related to restoration and persistently hydrologically altered environments.

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Vegas Wash. Rich Durfee identified invertebrates in Las Vegas Wash samples. Daren Carlisle reviewed an early version of the manuscript.

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Table 1. Sites used for seasonal study of Las Vegas Wash macroinvertebrates.

Site code	Description	Site-type
LW11.76	LV Wash above Vegas Valley Drive-sampling initiated in 2010-furthest upstream site	Reference
LW11.1	LV Wash below Vegas Valley Drive	Reference
LW9.1	LV Wash upstream of confluence with Clark County Advanced Wastewater Treatment Plant (CCAWTP)	Lotic
LW8.85	LV Wash at gage	Lotic
LW7.0	LV Wash-sampling initiated in 2010	Lentic
LW6.05	LV Wash at Pabco Road weir	Structure-restoration
LW5.5	LV Wash at Bostick weir	Structure-restoration
LW3.85	LV Wash at Demonstration weir	Structure-restoration
LW0.55	LV Wash downstream from the Northshore Road Bridge.	Lentic

Table 2. Results of literature review.

Literature citation	Biotic characterization			Seasonal change detected	Comments
	Ordination	Metrics	Predictive		
Álvarez-Cabria, M., J. Barquín, & J.A. Juanes. 2010.	X	X	--	X	Examined both hydromorphological and water quality stressors. Purpose was to identify the best season for biomonitoring with macroinvertebrate communities. Macroinvertebrate metrics correlated better with stress gradients (especially hydromorphological characteristics) during stable flow seasons. Many metrics did not show significant seasonal differences. Authors suggest that autumn is best season for sampling macroinvertebrate communities. No information on whether autumn sampling was predictive of insults occurring in other seasons. High flows occurred in winter and spring in this study.
Boulton, A.J., C.G. Peterson, N.B.	X	--	--	X	Ambient discharge in Sycamore Creek

Grimm, & S.G. Fisher. 1992.					constrained community structure, and, over longer periods, discharge extremes altered communities. A distinct seasonal change in communities was detected. Drought may have more of an impact on community structure than spates.
Clarke, R.T., M.T. Furse, R.J.M. Gunn, J.M. Winder, & J.F. Wright. 2002.	--	X	X	--	Samples collected in three seasons. Sampling variation was not greater in one season than another. Table 2 suggested clear differences between sites with different qualities regardless of season in which they were collected.
Dunbar, M.J., M. Warren, C. Extence, L. Baker, D. Cadman, D.J. Mould, J. Hall, & R. Chadd. 2010.	--	X LIFE index used as metric	--	--	Macroinvertebrate samples were collected in spring and autumn. Analyses were used to determine whether river discharge in the 6 months before sampling influence macroinvertebrate scores. Antecedent high and low flows influenced LIFE scores from spring and autumn samples. There was no test of ability of autumn

					samples to detect wintertime impacts.
Feio, M.J., T.B. Reynoldson, & MA. Graça. 2006.	--	--	X	X	How seasonal variability in macroinvertebrate communities affects the performance of a predictive model for assessing environmental quality was studied. Decided that samples should be collected in the same season as reference material was collected in.
Hawkins, C.P., R.H. Norris, J.N. Hogue, & J.W. Feminella. 2000.	--	--	X	--	Analysis indicated that one-time sampling resulted in a sensitive and accurate model. Sampling should be restricted to a standard index period. Mean Observed/Expected values where the probability of capture was $\geq$ 50% were similar between years.
Helms, B.S., J.E. Schoonover, J.W. Feminella. 2009.	X	X	--	X	Ordination distance among seasonal samples from the same sites increased as % forest cover increased. Communities were simpler and more consistent at impacted sites. Metrics were significantly related to

					% forest in all three seasons.
Johnson, R.C., M.M. Carreiro, H.-S. Jin, J.D. Jack. 2012.	X	X	--	X	Seasonal differences detected with NMS. Significant differences were detected between impaired and non-impaired streams for every month of the year when using taxon richness and EPT richness.
Linke, S., R.C. Bailey, & J. Schwindt. 1999.	Cluster analysis	X	X	?	Higher metric values were found in macroinvertebrate samples collected in winter relative to summer. Season, however, was not a significant predictor of taxon richness in the predictive model. Authors suggest that season should be taken into account in bioassessment and that 'temporal constraint' may miss important variation in the community at a site.
Reece, P.F. & J.S. Richardson. 2000.	Seasonal invertebrate data plotted in reference group ordination space.	--	X	X	Compared observed sites to expected (reference) sites. Main question in this study was whether samples collected in different seasons could be compared to reference

					sites that were only collected in early autumn. While results showed that comparisons using different seasons were robust, it was recommended that test samples should be collected during the same season as reference samples.
Šporka, F., H.E. Vlek, E. Bulánkova, & I. Krno. 2006.	X	X	--	Yes with ordination and some metrics.	Scores from macroinvertebrate indices varied naturally between seasons, confounding the detection of anthropogenic environmental change. Different seasons provided different resources and stressors. Spring was characterized by increased temperatures, discharge, light, and nutrient supplies which increased primary production and increased abundance of algophagous macroinvertebrates. High temperatures in summer resulted in high microbial activity and low DO's. In autumn and winter, temperature

					decreased along with illumination, and a large supply of allochthonous organic matter resulted in the development of detritophagous invertebrates. Sampling in certain seasons was considered inappropriate for logistical reasons.
Stark, J.D. and N. Phillips. 2009. Seasonal variability in the macroinvertebrate community index: are seasonal correction factors required? New Zealand Journal of Marine and Freshwater Research 43:867-882.	--	X	--	X	Seasonality detected but irrelevant to identification of impacted sites.

Table 3. Mean environmental variables associated with types of sites found in Las Vegas Wash. Standard error is in parentheses. Letters associated with rows indicate whether there was a statistically significant difference in variables between site-types. Those with the same lower-case letters indicate no significant difference (Tukey HSD test, P>0.05) between site-types.

Variable	Site-type			
	Lentic (n=34)	Lotic (n=56)	Reference (n=34)	Structure (n=84)
pH	8.28^{a} (0.04)	7.24^{b} (0.03)	8.18^{a} (0.05)	8.04^{c} (0.03)
DO* (mg/L)	7.92^{a} (0.14)	7.05^{b} (0.11)	9.86^{c} (0.34)	8.17^{a} (0.10)
Conductivity* (µS/cm)	2389^{a} (43)	2102^{b} (26)	3609^{c} (60)	2397^{a} (15)
Temperature* (^{o}C)	23.9^{a} (0.6)	24.1^{a} (0.5)	16.2^{b} (1.1)	23.9^{a} (0.4)
Alkalinity* (mg/L)	133^{a} (5)	121^{a} (2)	189^{b} (4)	125^{a} (2)
Hardness* (mg/L)	614^{a} (24)	556^{b} (11)	1514^{c} (27)	652^{a} (7)
Velocity (m/S)	0.14^{a} (0.02)	0.87^{b} (0.06)	0.33^{c} (0.03)	0.70^{d} (0.03)
Pfankuch index	100^{a} (2)	88^{b} (2)	97^{a} (2)	78^{c} (2)
Width (m)	15.9^{a} (0.4)	7.7^{b} (0.3)	$9.8^{a,b}$ (0.7)	62^{c} (2)
Depth(m)	0.96^{a} (0.03)	0.66^{b} (0.02)	0.33^{c} (0.02)	0.31^{c} (0.01)
Substrate index	4.9^{a} (0.2)	6.6^{b} (0.2)	4.8^{b} (0.1)	5.6^{c} (0.1)
% sand	46^{a} (4)	12^{b} (3)	25^{c} (3)	19^{c} (1)
Turbidity (NTU's)	7^{a} (1)	3^{b} (0)	4^{b} (2)	7^{a} (1)
CPOM (g)	0.08^{a} (0.04)	0.09^{a} (0.01)	0.80^{b} (0.15)	1.62^{b} (0.40)
Periphyton* (g)	$0.20^{a,b}$ (0.06)	0.05^{a} (0.03)	1.30^{c} (0.47)	$0.56^{b,c}$ (0.12)

*Indicates that these variables differed significantly between seasons.

Table 4. List of collected taxa and overall abundance in Las Vegas Wash.

Taxa	Abundance
COLLEMBOLA	2
EPHEMEROPTERA	
Baetidae	
Baetis sp.	23
Callibaetis sp.	44
Camelobaetidius musseri	1064
Fallceon quilleri	3291
Caenidae	
Caenis sp.	1
ODONATA	
Aeshnidae	
Aeshna sp.	1
Calopterygidae	
Hetaerina sp.	47
Coenagrionidae	18
Argia sp.	746
Ischnura sp.	2
Corduliidae	1
Gomphidae	10
Erpetogomphus sp.	1
Libellulidae	2
Brechmorhoga sp.	12
Erythemis sp.	1
Sympetrum sp.	1
HETEROPTERA	

Corixidae larvae	2
Trichocorixa calva	1
Mesoveliidae	
Mesovelia mulsanti	1
Veliidae	
Microvelia gerhardi	1
TRICHOPTERA	
Glossosomatidae	
Culoptila sp.	47
Hydropsychidae	
Smicridea sp.	1686
Hydroptilidae	
Hydroptila sp.	440
LEPIDOPTERA	
Pyralidae	
Petrophila sp.	793
COLEOPTERA	
Curculionidae	1
Dytiscidae	
Hydroporinae	2
Neoclypeodytes cinctellus	9
Heteroceridae	1
Hydrophilidae	
Enochrus sp.	2
Tropisternus sp.	1
DIPTERA	
Ceratopogonidae	3
Dasyhelea sp.	2
Chironomidae	1

Diamesinae	
Diamesa sp.	5
Orthocladiinae	
Cricotopus sp.	5132
Cricotopus / *Orthocladius* sp.	3
Eukiefferiella sp.	15
Limnophyes sp.	3
Orthocladius sp.	3
Parametriocnemus sp.	3
Thienemanniella sp.	17
Chironominae	
Apedilum sp.	254
Chironomus sp.	1096
Cladotanytarsus sp.	165
Cryptochironomus sp.	28
Dicrotendipes sp.	231
Endotribelos sp.	6
Glyptotendipes sp.	1
Goeldichironomus sp.	1
Micropsectra sp.	31
Nimbocera sp.	7
Paratanytarsus sp.	1
Phaenopsectra sp.	198
Polypedilum sp.	5334
Pseudochironomus sp.	45
Rheotanytarsus sp.	71
Tanytarsus sp.	33
Xestochironomus sp.	1

Tanypodinae	
Ablabesmyia sp.	3
Labrundinia sp.	1
Pentaneura sp.	71
Procladius sp.	3
Culicidae	
Culex sp.	1
Dolichopodidae	17
Empididae	
Hemerodromia sp.	42
Ephydridae	99
Muscidae	15
Psychodidae	3
Pericoma /*Telmatoscopus* sp.	1
Psychoda sp.	1
Sciomyzidae	2
Simuliidae	
Simulium sp.	882
Stratiomyidae	1
Caloparyphus sp.	114
Euparyphus sp.	125
Tipulidae	3
Erioptera sp.	2
Limonia sp.	17
TURBELLARIA	47
Dugesiidae	
Dugesia sp.	141
HIRUDINEA	

Erpodellidae	1
Glossiphoniidae	1
OLIGOCHAETA	
Enchytraeidae	7
Lumbricidae	3
Lumbriculidae	29
Naididae	95
Tubificidae	70
NEMERTEA	
Prostoma sp.	26
CLADOCERA	
Daphniidae	
Simocephalus sp.	2
OSTRACODA	12
AMPHIPODA	
Hyalellidae	
Hyalella azteca	323
DECAPODA	
Cambaridae	1
GASTROPODA	
Ancylidae	
Ferrissia sp.	2
Physidae	260
Thiaridae	
Melanoides sp.	26
BIVALVIA	
Corbiculidae	
Corbicula	149

Table 5. Results of 2-factor ANOVA for macroinvertebrate richness, abundance (ln X+1), and ETO richness for site-type during four seasons in Las Vegas Wash.

RICHNESS

Source	DF	SS	MS	F	P
SITE-TYPE	3	2785.00	928.334	88.93	0.0000
SEASON	3	25.99	8.664	0.83	0.4789
TYPE*SEASON	9	23.96	2.663	0.30	0.9853
Error	192	2004.38	10.439		
Total	207				

Grand Mean 7.2138 CV 44.79

ABUNDANCE

Source	DF	SS	MS	F	P
SITE-TYPE	3	331.850	110.617	79.23	0.0000
SEASON	3	1.821	0.607	0.43	0.7284
TYPE*SEASON	9	15.040	1.671	1.20	0.2990
Error	192	268.070	1.396		
Total	207				

Grand Mean 3.3814 CV 34.94

ETO

Source	DF	SS	MS	F	P
SITE-TYPE	3	305.916	101.972	54.10	0.0000
SEASON	3	38.962	12.987	6.89	0.0002
TYPE*SEASON	9	23.544	2.616	1.39	0.1958
Error	192	361.875	1.885		
Total	207				

Grand Mean 2.4247 CV 56.62

Figure 1. Biplot from seasonal data collected from 2004-2011 based on a canonical correspondence analysis (CCA) of sites with respect to environmental variables. Environmental variables were related to community attributes as shown by arrows. Site samples are represented by geometric shapes as shown in the legend.

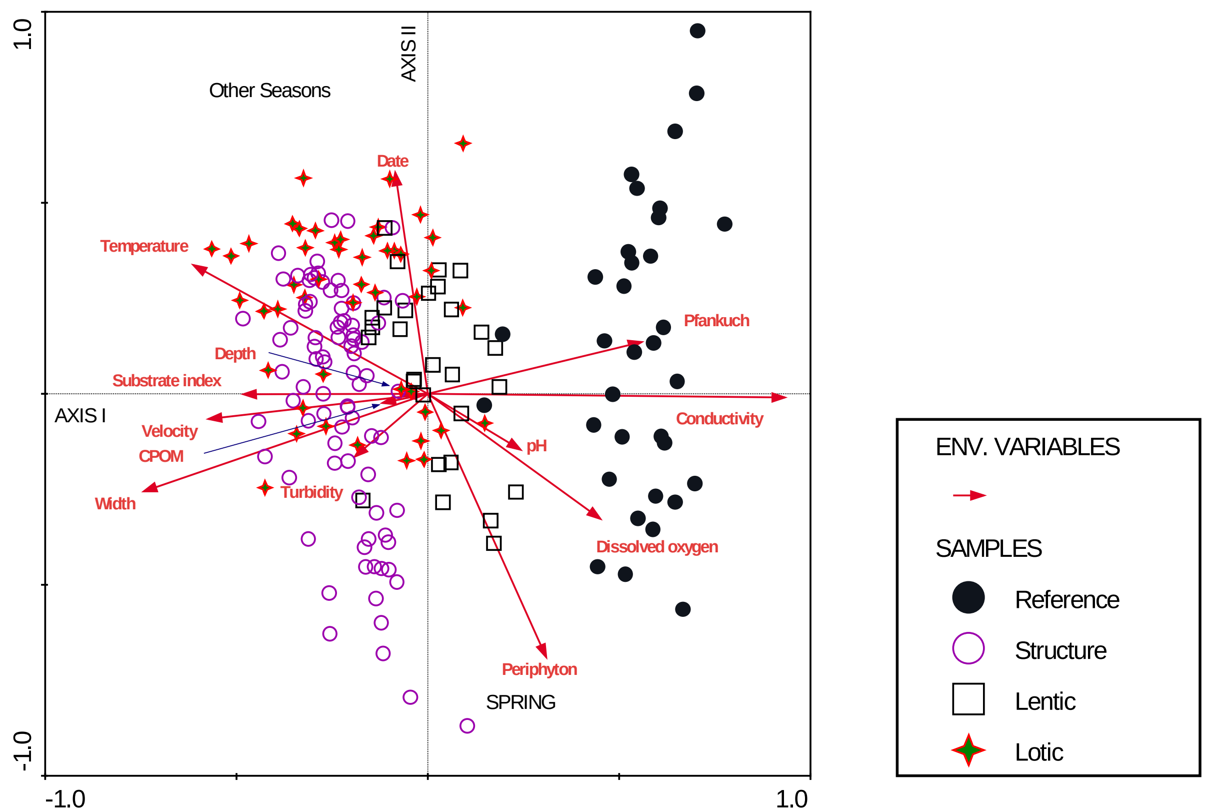


Figure 2. Biplot from data collected from 2004-2011 based on a canonical correspondence analysis (CCA) of macroinvertebrate taxa in association with environmental variables. Only those species that had a fit and weight ≥ 5% are shown in the figure.

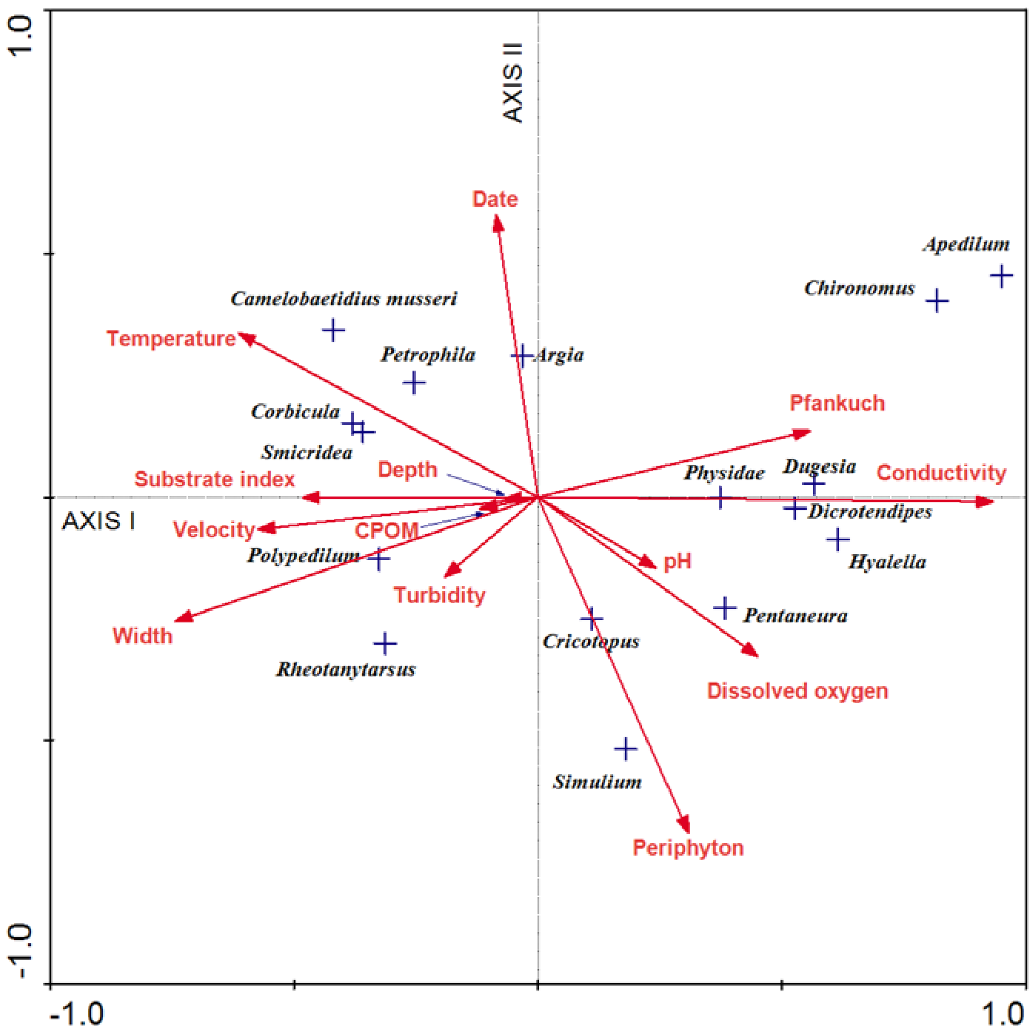


Figure 3. Mean taxa richness (a), mean invertebrate abundance (b), and Mean ETO richness (c) at different types of sites in the Las Vegas Wash drainage. Variability is presented as standard error. Bars with the same lower-case letters indicate no significant difference (Tukey HSD test, P>0.05).

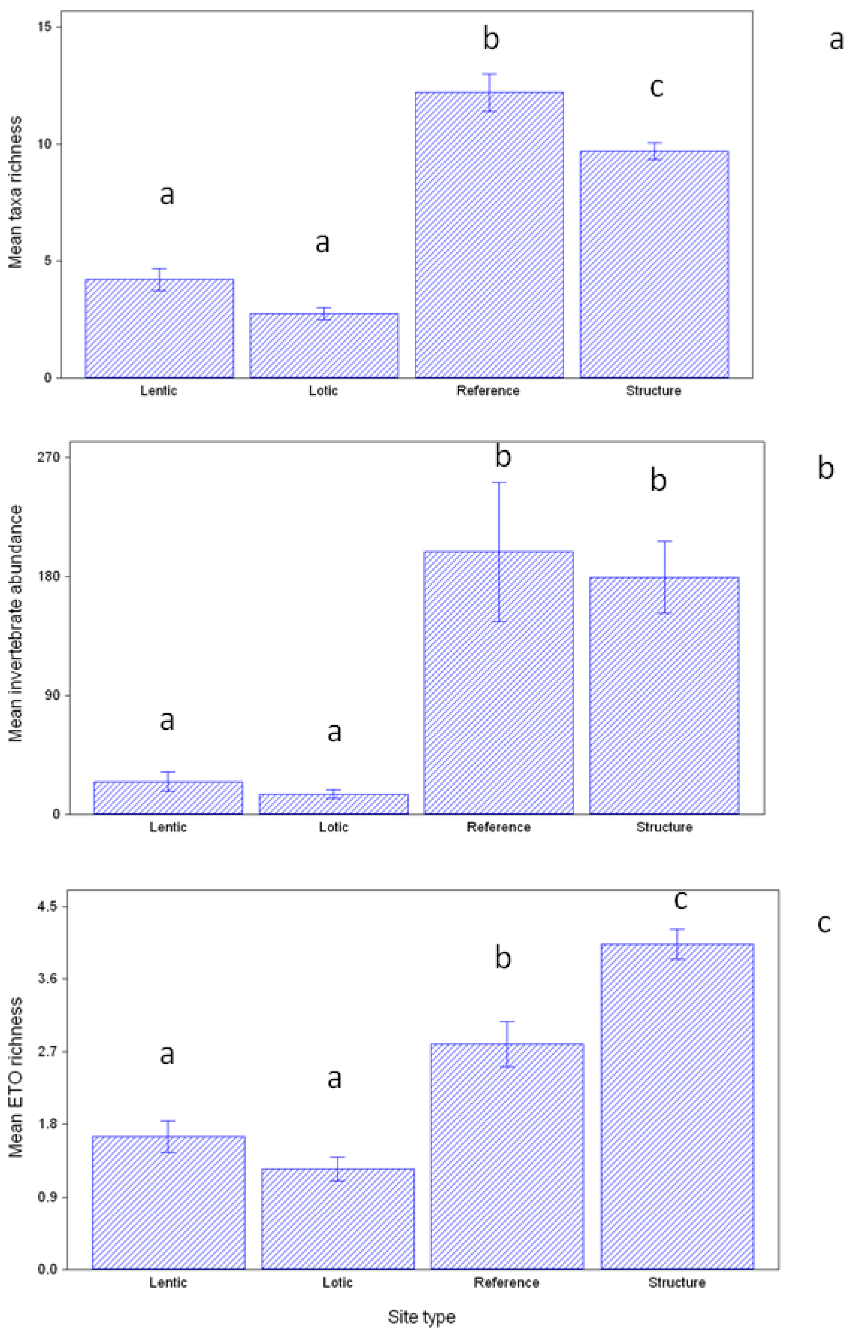
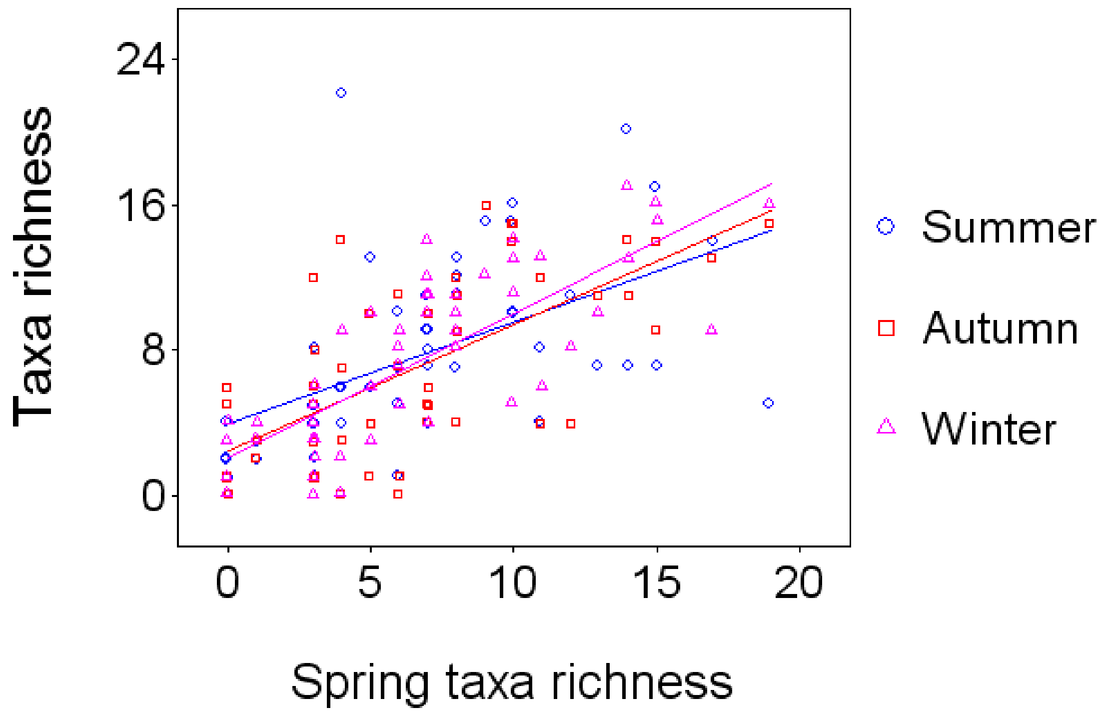


Figure 4. Correlation of spring taxa richness with summer, autumn, and winter taxa richness (*r*=0.5266 to 0.7775, *P*≤0.0001).



PEER REVIEW DOCUMENTATION

PROJECT AND DOCUMENT INFORMATION

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Document IS DETECTION OF LONG-TERM IMPACTS USING AQUATIC MACROINVERTEBRATES SEASONALLY DEPENDENT?

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Team Leader S. Mark Nelson

Leadership Team Member

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Peer Reviewer Daren Carlisle

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REVIEW REQUIREMENT

Part A: Document Does Not Require Peer Review

Explain

Part B: Document Requires Peer Review: SCOPE OF PEER REVIEW

Peer Review restricted to the following Items/Section(s): Reviewer:

REVIEW CERTIFICATION

Peer Reviewer - I have reviewed the assigned Items/Section(s) noted for the above document and believe them to be in accordance with the project requirements, standards of the profession, and Reclamation policy.

Reviewer: Daren Carlisle Review Date: 5/14/14
Signature (see acknowledgements)

Preparer - I have discussed the above document and review requirements with the Peer Reviewer and believe that this review is completed, and that the document will meet the requirements of the project.

Team Member: S. Mark Nelson Date: 6-13-14
Signature