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Stream Daylightling: The Effect of Restoring Culverted Streams on Ecosystem Functioning in Oslo, Norway

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Master of Science in Ecology
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Abstract

Culverted streams generally have compromised ecological integrity due to issues with low light, extreme habitat modifications and increased pollution loadings. Restoring culverts back to aboveground streams has the potential to solve these issues. One of the main goals in restoration is restoring ecosystem services through renewed ecosystem functioning. Directly measuring ecosystem functions has the ability to serve as indicators for many, often hidden variables on a systems wide scale. This study compared the ecosystem functioning of restored stream stretches to reference stretches within the city of Oslo to assess the effectiveness of restoration efforts. Vegetation, hydromorphology and water chemistry are additionally analyzed to determine underlying causes for discrepancies. The measured ecosystem functions were total, microbial and invertebrate mediated decomposition, algae biomass accrual, algae primary production, and grazing intensity. Six daylighted sites were compared to twelve reference sites that had never before been culverted and covered a diverse array of habitats, water chemistries and degrees of urbanization. Decomposition was measured in late autumn using litter-pack methods with one-month exposure times, using two difference decay mediums: alder (*Alnus glutinosa*) leaves and Wettex cellulose sponge cloths. Algae and grazing metrics were measured using granite tiles as an artificial substrate placed in the stream for one month in spring. The data was analyzed using ANCOVA on mixed effect models and multiple linear regression. Regarding ecosystem functioning, it was found that restored stretches of stream responded the same to environmental variables as reference stretches, that had never before been culverted. The generally higher primary production and possibly lower rates of decomposition seen in restored stretches can be explained by the early-successional state of the newly constructed streams, with greater light intensity from more open canopies resulting in increased primary production and lower litter fall leading to lower capacity for decomposition. Restored streams had neither higher nor lower ecosystem functioning than natural streams of the same vegetation community, hydromorphology and water chemistry. This study concluded that stream daylighting efforts in Oslo have been successful at restoring ecosystem functioning to a level comparable to those of similar streams in the vicinity.

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Introduction

Almost all European rivers have been degraded by either a combination of land-use change, fragmentation, water stress, invasive species or pollution: 90% of the catchment area has been converted over to agriculture in central and step Europe, 50% of wetlands and 95% of the flood plains have been lost due to channelization and flood control, more than 6000 large dams highly fragment Europe's river systems and water pollution issues continue to be a concern even with improved water sanitation standards and runoff control (Tockner et al., 2009). This extensive habitat loss, major fragmentation of populations and degraded water quality has resulted in the majority of rivers, streams and lakes exhibiting "less than good" ecological status according to the European Water Framework Directive (Eea, 2012). It is thus imperative that ecological restoration and mitigation be of primary concern for European water managers.

Specifically, culverting (redirecting streams underground) has had a rather large reach of influence on European water ways and can lead to many environmental, social and economic problems. As much as 20% and 15% of the total length of Switzerland's and Denmark's streams are confined to culverts respectively (Iversen & Anderson, 1997; Kummert & Stumm, 1992). Culverted streams often have poor ecological standing due to issues relating to lower light levels, extreme habitat modification, increased sources of pollution and separation from the terrestrial environment (Wild, Bernet, Westling, & Lerner, 2011). They also provide no social benefits and pose as economic risks, with higher maintenance costs due to difficulty of access and increased flooding potential (Wild et al., 2011).

Stream daylighting, also known as deculverting, has the potential to restore the ecological integrity of streams, as well as provide other social and economic benefits. Stream daylighting is essentially the removing of the culvert and placing the stream back on the surface. It is generally accompanied by a restoration of the stream back to a seminatural state. It has been reported that daylighting has increased ecological integrity through restored wetlands and riparian vegetation and brought back habitat heterogeneity to the streams and the landscape (Wild et al., 2011). It has also been shown to provide economic and social benefits through recreation and education and reduced maintenance costs (Wild et al., 2011).

Unfortunately, due to lack of post-assessment and/or reporting, little is known of the effectiveness of many restoration methods, exacerbating an issue of too much "gut feeling" and lack of science-based management seen in restoration projects. In Germany it was found that in stream restoration, almost no parameters were measured, instead success was based on "gut feeling" and they were likely to report a success result regardless of if ecological parameters improved or declined

(Jähnig et al., 2011). This may have a regulatory component, as managers fear losing funding from reporting negative results; but nevertheless, the poor reporting is most likely preventing the adoption of better methods. Specifically regarding culvert removal projects, economic benefits are rarely reported and social and environmental benefits are often reported anecdotally, or the reports are severely lacking spatially and temporally (Wild et al., 2011). For example, the removal of culverts has been reported to restore fish migration ability, but generally only fish presence is recorded and no data on where the fish came from or how far they actually move was collected (Wild et al., 2011). In adaptive management, one regularly monitors the progress of a project and makes adjustments accordingly. Clear goals and post-assessments of the realization of said goals is a very integral part of the adaptive management process. In addition, reporting allows for collaboration and learning from each other.

One common goal in stream restoration projects is the enhancement of ecosystem services such as cleansing and resilience. The paradigm until recently for achieving this has been focused on water quality and biodiversity. Starting in the mid-1970's, the main form of water quality protection in Europe was point-source pollution measures through the use of standards for waste water discharge and industrial emissions (Eea, 2012). This methodology proved inadequate in the face of the increasing problem of eutrophication, so in the late 1980's they switched focus to non-point pollution sources such as agricultural and urban sewage runoff (Eea, 2012). Then in 2000, the European Water Framework Directive called for the creation of River Basin Districts, to facilitate the collaboration across political boundaries to solve the issues faced by European water managers (Eea, 2012). Despite European regulation taking an increasingly watershed-scaled perspective, the dominant ecological paradigm since the 1990s has been increasing biodiversity at the short-stream-stretch scale, through increasing niche partitioning by increased heterogeneity of the stream bed, despite the lack of evidence that this is effective for anything but salmonid species (Roni, Hanson, & Beechie, 2008). The focus on biodiversity has come from an assumption that ecosystem services such as water filtration and regulation are positively correlated with diversity; which is true on at least a larger scale (Benayas, Newton, Diaz, & Bullock, 2009).

High habitat heterogeneity and biodiversity is not always naturally found in all locations and managing for unnatural conditions can result in negative results. Pedersen, Kristensen, and Friberg (2014) found in a study in Denmark that increasing stream bed heterogeneity with coarse gravel actually lowered the species count. The river system was dominated by homogenous fine sandy substrate, so the catchment lacked the necessary source populations to colonize this new habitat. Restoration should aim to recreate natural systems as closely as possible to reestablish the natural

geomorphological processes, and for that you need well defined metrics, relating to the entire catchment area.

In addition to biodiversity not always being natural, no information is provided by a diversity measures on the state of functionally vital species. There is a consensus that there is a minimum number necessary to sustain ecosystem function and more is better for long term stability of the system (Loreau M. et al., 2001). However, from a functional point of view, species really only matter regarding their functional role in the system, thus a functional group metric may be a better measure for evaluating restoration projects (Loreau M. et al., 2001). Alternatively, one could just look at the ecosystem function directly, such as decay or primary production.

Measures of ecosystem functioning can serve as indicators of many, often hidden, variables. For example in one study, the function leaf litter decomposition showed response to pH change, where strictly the diversity within the invertebrate functional group leaf-shredder and bacteria and fungi showed no great response; however upon closer investigation it was found that certain pH sensitive species declined and the acidity had a strong negative effect on bacterial and fungal extracellular enzyme efficiency (Simon, Simon, & Benfield, 2009). Measures of ecosystem function can be used as an assay for more complex interactions within the whole ecosystem at small and larger scales that may be missed by just biodiversity measures (Ryder & Miller, 2005). But ecosystem function can also be very confounding, as for example, leaf litter break down rates can be accelerated by increased biological activity and increased flow velocity, so determining the underlying cause may be impossible from just one measure of function (Paul, Meyer, & Couch, 2006). It is therefore important to measure a variety of variables in addition to ecosystem functioning such as diversity, functional groups, and abiotic environmental factors. Together they all interdepend on each other and will provide deeper insight into the specific situation (Loreau M. et al., 2001).

In the city of Oslo, Norway there is an initiative to daylight as many of the culverted streams as possible, in order to reestablish lost ecosystem services and restore the ecological integrity of the Oslofjord system. Many of the streams have already been deculverted and renaturalized. It is now of interest to assess the progress of these works and see if they are meeting their desired goals. This will provide valuable insights for future improvements and further culvert daylighting in Oslo and elsewhere. One of the goals is to restore the ecological function of the previously culverted streams. To evaluate this, ecosystem functioning of the newly restored daylighted streams needs to be compared to reference streams that had never been culverted and are similar in nature and in proximity to the restored stretches.

In this study the ecosystem functions of decomposition and primary production will be analyzed within 6 restored stretches and 12 reference stretches. The reference sites cover a wide range of habitats, degrees of urbanization and water quality levels found within the city of Oslo. Specifically, this study will look at total, microbial and invertebrate mediated decomposition, algae accrual, algae primary production and grazing pressure, compared to a variety of measured hydromorphological and other environmental factors. The comparability of ecosystem functioning in restored stretches to reference stretches will be assessed and possible causes for discrepancies will be evaluated. This will facilitate future science-based decision making within the stream daylighting process.

Methods

Study Area

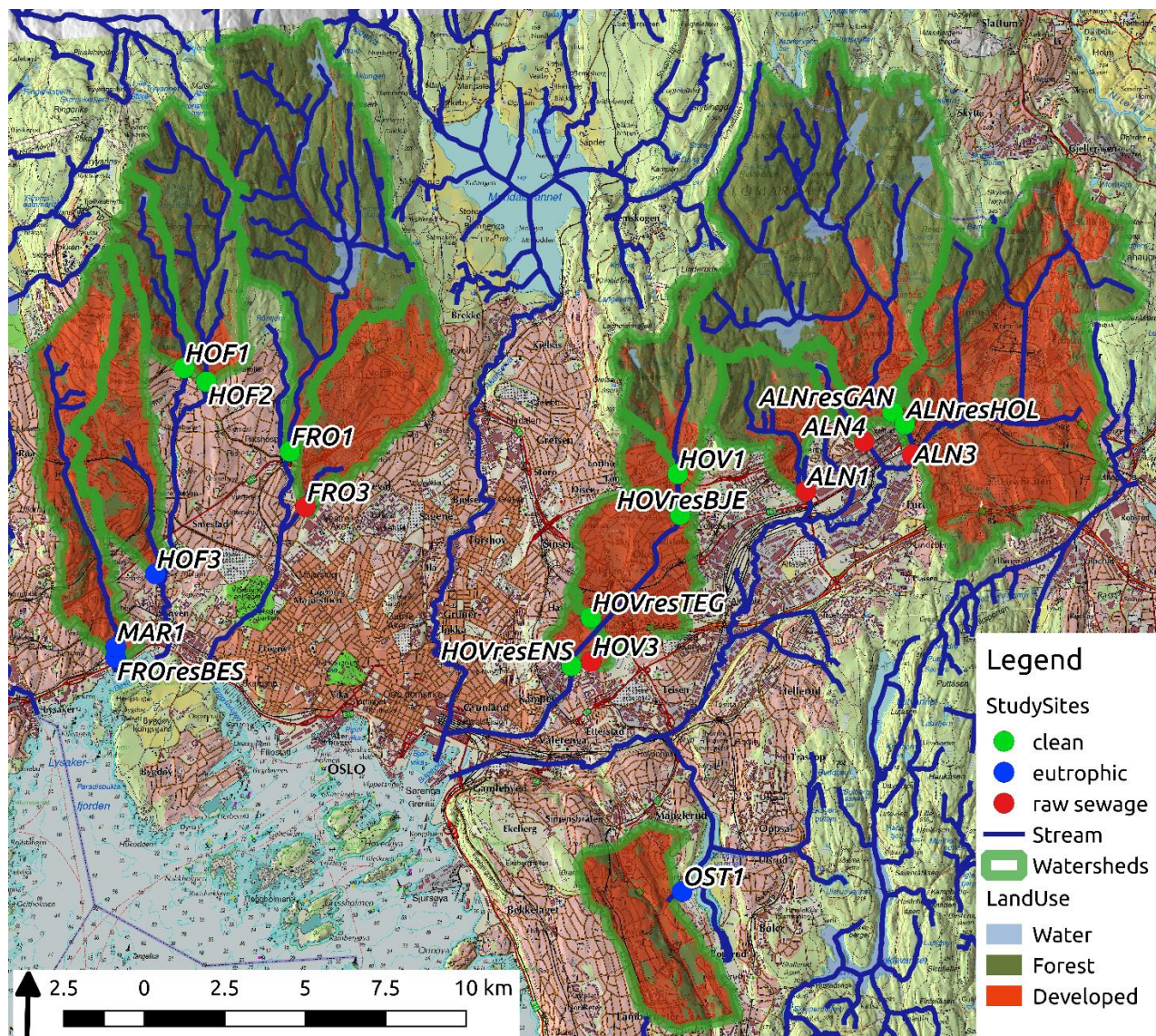


Figure 1: Map of study area with study sites classified by pollution level, study sites' watersheds and dominant land uses. Restored sites have a "res" amidst the name.

18 sample reaches were selected in streams within the municipality of Oslo; 12 reference reaches and 6 restored reaches. The restored sites were originally culverts that had been opened, placed back on the surface, and renaturalized. Sample reaches were defined as a length of stream of similar character that stopped at any culverts or bridges, which would significantly separate the below and above stream stretches. The sample sites were located in the suburbs and industrial parks around the city center and were selected to cover as diverse a spectrum as possible; they ranged from natural streams in mature forests to fully embanked channels along the road (see Fig. 1 for a study area map, Table 1 for summary statistics and Appendix 1 and 4 for detailed statistics and pictures.)

The city of Oslo, Norway is located at 59.92°N 10.73°E. As per the beginning of 2017, there were 666,759 residents in the municipality (Statistics Norway, 2017). It is located at the transition

between the nemoral and boreal biomes (Metzger, Bunce, Jongman, Mücher, & Watkins, 2005). The average temperatures for July and January are 16.4°C and -4.3°C respectively (NRK & Meteorologisk Institutt, 2018). The geology of the city is dominated mostly by sedimentary limestone, clay slate and marlstone while the surrounding hills are predominately igneous syenite, quartz syenite and sedimentary mica shale (Geological Survey of Norway, 2015).

Table 1: Summary table of water chemistry, hydromorphology and vegetation by study area, restored and reference sites. Five day biological oxygen demand (BOD₅), Ammonium (NH₄⁺) and phosphate (PO₄³⁻)

	Total				Restored sites				Reference sites				
	unite	avg	min	max	Stdev	avg	min	max	Stdev	avg	min	max	Stdev
Number of sample sites				18.00				6				12	
Low water BOD₅	mg/l	2.04	0.57	8.84	2.47	0.89	0.60	1.21	0.20	2.62	0.57	8.84	2.88
Med water BOD₅	mg/l	2.45	1.37	8.93	1.80	1.87	1.43	2.63	0.45	2.74	1.37	8.93	2.16
High water BOD₅	mg/l	1.51	0.41	3.36	0.75	1.26	0.95	1.60	0.23	1.64	0.41	3.36	0.89
NH₄⁺	µg/l	142	7	470	168	52	14	153	53	187	7	470	190
PO₄³⁻	µg/l	41	2	130	42	14	2	51	19	54	3	130	45
Temp	mid May to mid June daily avg in deg.	11.14	8.65	14.98	1.82	12.27	9.55	14.98	1.97	10.58	8.65	13.35	1.51
	late November daily avg in deg.	3.95	1.61	8.18	1.76	3.29	1.99	4.76	1.16	4.28	1.61	8.18	1.96
Length	m	97	11	270	63	76	36	140	50	107	11	270	68
Max Depth	cm	49	10	130	29	57	20	130	39	45	10	90	25
Depth Range	cm	40	6	120	29	47	13	120	38	37	6	84	24
Depth Variability	4 pt. scale	3	1	4	1	4	2	4	1	3	1	4	1
Width	m	2.7	0.8	4.9	1.3	2.9	1.6	4.1	1.0	2.6	0.8	4.9	1.4
Width Variability	4 pt. scale	3	1	4	1	3	1	4	1	3	1	4	1
Sed. Mode	Weightworth size class	pebble	silt clay	boulder	pebble	pebble	sand	pebble	pebble	pebble	silt clay	boulder	
	φ	-3	4	-8	4	-3	2	-4	2	-4	4	-8	5
Sed. Sorting	φ	2.8	4.4	0.0	1.2	3.2	4.2	2.0	0.9	2.6	4.4	0.0	1.3
Bank Gradient	deg.	58	10	90	24	53	20	90	27	61	10	90	23
Bank Height	cm	104	30	450	97	54	30	95	22	129	40	450	110
Bank Protection	% reach length	18	0	100	34	27	0	100	40	13	0	100	31
Bank Protection	height cm	42	0	290	81	23	0	60	28	51	0	290	98
Flow Character		swirled	slow	turbulent	swirled	swirled	uniform	turbulent	swirled	swirled	slow	turbulent	
Bank Veg. Cover	%	85	0	100	28	84	50	100	23	86	0	100	31
Canopy Cover	%	39	0	94	39	5	0	12	4	56	0	94	37
Riparian Forest Width		single row interrupted	isolated/absent	>15m	single row interrupted	single row interrupted	isolated/absent	single row	single row	single row	isolated/absent	>15m	
Bank Vegetation	n Mixed forest						0				4		
	n Deciduous forest						0				4		
Vegetation Community	n Meadow-Early successional riparian scrub						4				1		
	n Meadow						0				3		
	n Pioneer meadow						2				0		
Embanked	% of length	10	0	58	14	18	0	58	21	6	0	21	8
Emb. 2 Sided	% of length	2	0	10	4	3	0	10	4	2	0	10	3
Culverted	% of length	25	0	80	29	19	0	80	31	28	0	74	28
Bridge	% of length	2	0	7	2	3	0	7	3	1	0	2	1
Weirs	numb	3	0	32	8	8	0	32	12	1	0	4	1
Dams	% of length	1	0	9	2	0	0	0	0	1	0	9	3
% of Watershed Developed		50	9	88	28	49	16	80	29	51	9	88	28

Environmental variables

Water chemistry and temperature

Ammonium and phosphate

Water samples were taken at each reference site using the hand dip method between the 26th and the 29th of September, 2016 and at each restored site on the 18th and 19th of September, 2017. The samples were processed by an accredited lab at the Norwegian Institute for Water Research, in Oslo, using Norwegian Standard NS 4746:1975 for ammonium and Norwegian Standard NS 4724:1984 for phosphate.

BOD₅

The biological oxygen demand after 5 days (BOD₅) was measured in mg O₂/l at each of the sites, once by low flow on Sep 4th, once by medium flow on May 25th and once by high flow on Aug 10th in 2017. Each time, all the sites were sampled and processed the same day. The BOD₅ was measured with standard method 5210B (APHA, AWWA, & WEF, 2001). Only one sample was taken at each site per sampling day. The samples were taken in plastic containers using the hand dip method and analyzed in 250 ml glass bottles with ground glass stoppers. In order to characterize in-situ chemistry, pure stream water was analyzed without dilution or the addition of nutrients, bacterial seed or reagents to the samples. After coming in from the field, samples were poured into the 250 ml bottles, placed in a 20°C water bath and bubbled with an air-diffusion-stone for about 15 minutes as they warm up to 20°C±3°C as recommended in the USGS National Field Manual for the Collection of Water-Quality Data to prevent supersaturation as the water warms (Delzer & McKenzie, 2015). The air-diffusion-stones were cleaned with disinfectant Vircon S produced by Vircon, rinsed in DI water and soaked for multiple hours in DI water before each use. The oxygen-probe used was the Fibox 4 with the oxygen dipping probe DP-PSt3 produced by PreSens.

Temperature

Water temperature was measured at 30-minute intervals using HOBO and TinnyTag temperature loggers. They were anchored to the stream floor by rebar, either just above or below the substrate. Some of the sites were in city parks, so the temperature loggers were buried just below the substrate to prevent them from being stolen. Degree days (DD) used for the ecosystem function statistics were calculated based on the average temperature for each day.

Due to technical malfunctions and loss of the temperature loggers in flood events, some water temperature data was missing during the two ecosystem function experiments for sites ALN3, MAR1 and HOF1 from Nov. 7th to Dec. 6th, 2016, for all sites from May 16th to May 29th, 2017 and site ALN3 from May 16th to Jun 14th, 2017. The missing temperatures were modeled based on the water temperatures at other sites and air temperatures where necessary. Where the data permitted, it was

preferred to use the water temperature of the sites, to estimate the missing values, because water temperature is affected by more than just air temperature. The air temperatures used were collected at the Oslo (Blindern) observation station (NRK & Meteorologisk Institutt, 2018). The statistics were performed in R version 3.2.3 (The R Foundation for Statistical Computing, 2015). (See Appendix 3 for the models and results)

The temperatures missing from the 16th to 29th of May 2017 were from the first half of the ecosystem function experiment on algae growth. The experiment ran in total from May 16th to Jun 16th, 2017. The average air temperature for the second half of the treatment was 14.40°C. The air temperature difference between the first half and the second half of the experiment was 2.37°C and the air temperature difference was 2.05°C between the second half of the experiment and an equal time interval after the experiment from Jun 17th to Jun 30th, 2017. These differences in air temperature were not significantly different based on a t-test between two linear models ($t = 0.26$, $df = 78$, $p\text{-value} = 0.60$). The test compared the slopes of two linear models using reference level parameterization. One calculated air temperature vs a factor of the 1st half of the experiment over the 2nd half. The other linear model calculated air temperature vs a factor of the 2nd half of the experiment over an equal interval of time after the experiment. Thus, the average water temperature was estimated for the first half of the experiment by finding the difference in water temperature between the second half and the equal time interval after the experiment. This difference was then subtracted from the average water temperature of the second half, to get an estimated average water temperature for the first half. This procedure was done for each constituent site. The value acquired from this equation was then converted to degree-days by multiplying this average daily temperature by the number of days in the first half of the experiment, which was then added to the degree-days that were actually measured in the second half.

The above method did not work for two sites because data was missing from site ALN3 until the end of the experiment on Jun 16th and data was also missing from site ALN1 after Jun 16th. Instead, the missing water temperature data for site ALN1 was estimated with a linear model based on the water temperature of another site, HOF3, that most closely correlated to the missing site. Additionally, the missing water temperature data for ALN3 was modeled from its closest correlated site, FRO3. Pearson correlation coefficients were calculated either based on water temperature data during the second half of the experiment for ALN1 from May 30th to Jun 16th or during an equal time interval immediately after the experiment for ALN3 from Jun 17th to Jun 30th. The model for ALN1 had an R^2 of 0.80. The model for ALN3 had an R^2 of 0.80.

Water temperatures were missing from three sites in autumn 2016, but there was no data missing in autumn 2017. The missing water temperature data was estimated with a linear model based

on the water temperature of another site that most closely correlated to the missing site. The correlation and modelling were based on 2017 water temperature data during the same time of year as the missing 2016 data. The correlation was analyzed according to Pearson correlation coefficients. MAR1 correlated best with HOF3 and the model had an R^2 of 0.95. ALN3 correlated best with ALN1 and the model had an R^2 of 0.906. HOF1 correlated best with FRO1 and the model had an R^2 of 0.96. The models were then applied to 2016 water temperature data to get an estimate for the missing water temperatures. (See Appendix 3 for the models and results)

Hydromorphology

The hydromorphology assessment was adapted from the Restoring Rivers for Effective Management (REFORM) project protocols (Poppe et al., 2012). The hydromorphology was assessed over a two-week period at the end of the field-season. All the streams were measured during medium flow. Reach length was measured to the nearest meter either using aerial photos or GPS. The maximum depth at the shallowest cross section and the maximum depth at the deepest cross section were measured to the nearest centimeter. The depth variability was rated qualitatively as “none”, “low”, “medium” or “high” as compared to the other studied stretches. The stream width was measured at five equally spaced transects to the nearest tenth of a meter. The width variability was rated qualitatively as “none”, “low”, “medium” or “high” as compared to the other studied reaches. The sediment was sampled along five equally spaced transects with 20 random point samples per transect making a total of 100 point-samples. The sediment was classified according to the Wentworth size classes with groupings boulder, cobble, pebble, granule, sand and silt/clay and the addition of Xylal (coarse woody), coarse particulate organic matter (CPOM) and fine particulate organic matter (FPOM) (Wentworth, 1922). The sediment mode, sorting and sediment skewedness was calculated from the Wentworth size classes in the unit Phi (ϕ), according to the methods from Folk and Lord (1957): $\text{sorting} = (\phi_{84} - \phi_{16})/4 + (\phi_{95} - \phi_5)/6.6$, $\text{skew} = (\phi_{16} + \phi_{84} + 2\phi_{50})/2(\phi_{84} - \phi_{16}) + (\phi_5 + \phi_{95} - 2\phi_{50})/2(\phi_{95} - \phi_5)$. The Krumbein Phi scale was originally developed to simplify statistics and graphical representations with the linearization of particle size distributions across size classes, by defining the unit ϕ as $-\log_2 D$ where D is the diameter in millimeters (Krumbein, 1936). The degree of observed sediment movement during the field season was recorded qualitatively as either significant or insignificant. Sediment load alterations were assessed and all possible signs of an alteration were noted down. The flow character was qualitatively classified as “slow”, “uniform”, “heterogeneous/swirled” or “turbulent”. It was noted if there was important woody debris accumulation and/or important bed-load accumulation. It was noted if the stretch had gravel, sand and/or silt banks. The average bank gradient was recorded in degrees for the left and right bank separately. The average bank height was recorded in centimeters for the left and right bank separately.

If bank protection was present, the height in centimeters, type (biologic engineering measures, combined, pilotage, riprap, stone pitching facing, stone pitching tightly packed, concrete and grass) and percent of stretch length covered for the left and right banks separately was recorded.

Channel alterations were assessed within the sampled stretch and 1km upstream using Oslo Kommune 2017 leaf-off-aerial-photos and/or walking (Kartverket, NIBIO, & Statens Vegvesen, 2017). Since many of the streams in the city are below the surface, the stream network Elvnett – ELVIS from the Norwegian Water Resources and Energy Directorate was used as a reference to map the stream courses (NVE, 2017). Embankments, culverts, bridges, weirs dams and slabs were recorded at the site. Length was defined as distance along the stream and width as distance across the stream. It was recorded if embankments were on one or both sides of the stream, embankment height in centimeters and length in meters, length of bridges and dams, height and number of weirs, and width and length of culverts and slabs. For the 1km stream stretch above the sample reach, only length or number of features was recorded.

Vegetation

Bank vegetation

The percent vegetation coverage of the left and right banks were recorded separately. The percent canopy coverage of the river was recorded in mid to late summer, averaged from 7 point-samples at each reach evenly spaced down the length. The width of the riparian forest was recorded as “>15m”, “5-15m”, “single row”, “single row interrupted”, or “isolated/absent” for the left and right banks separately. The percent bank coverage of the riparian forest was recorded for the left and right banks separately.

Vegetation community

Vegetation communities were originally delineated within a 20m and 50m buffer around the sampled reach. The categories were adapted from the REFORM project (Peppe et al., 2012): herbaceous pioneer vegetation, cane brake, tall herbaceous fringe, nitrophilous fringe, invasive herbaceous species, woody pioneer, deciduous forest, mixed forest, coniferous forest, pasture, fallow land, grassland extensive, grassland intensive, lawn, field, soft wood floodplain forest, hard wood floodplain forest, wetland/bog, invasive woody species, and no vegetation/sealing. Vegetation communities were sketched in the field onto printed Oslo Kommune 2017 leaf-off-aerial-photos (Kartverket et al., 2017). It was observed that the vegetation communities did not differ significantly regarding species assemblages, and actually laid along a successional gradient; thus, they were simplified to mixed forest, deciduous forest, meadow-early successional riparian scrub, meadow and pioneer meadow for each sampled stream reach.

Watershed land-use

Watershed boundaries were calculated and edited from a 10x10 m resolution digital elevation model (DEM) and a 1:50,000 scale topographic raster map from the Norwegian Mapping Authority (Kartverket, 2013, 2017). A 1:50,000 scale vectorized stream network from the Norwegian Water Resources and Energy Directorate (NVE, 2017) was also used with the help of the software ArcGIS Desktop 10.4 and QGIS 2.14.20 with plugins from GDAL Tools Plugin 1.2.29, GRASS GIS 7.0.3 and SAGA 2.2.3 (Conrad et al., 2016; ESRI, 2016; GDAL Development Team, 2016; GRASS Development Team, 2015; QGIS Development Team, 2016). All spatial data was downloaded and worked with in projection EUREF89 UTM Zone 33. In QGIS, vector points were digitized at 1:25,000 scale by hand at the base of each sampled reach, using Google satellite imagery for reference (Google, 2017). The two DEM tiles were merged in QGIS, using the 'Merge' tool from GDAL. In ArcGIS, the 'Fill (Spatial Analyst)' tool was used on the merged DEM to remove depressions. In ArcGIS, the 'Flow Direction (Spatial Analyst)' tool was used on the filled-DEM, and thereafter the 'Flow Accumulation (Spatial Analyst)' tool was used on the flow-direction-raster to create a flow-accumulation-raster. In ArcGIS, the 'Snap Pour Point (Spatial Analyst)' tool was used on sampled-reach-points-vector and flow-accumulation-raster with snap distance = 80 m to get the points centered in the DEM's stream track, so as to avoid only getting half-watersheds. In ArcGIS, the 'Watershed (Spatial Analyst)' tool was used on the flow-direction-raster and snapped-sampled-reach-points-vector to automatically generate watershed boundaries. In ArcGIS, the 'Raster to Polygon (Conversion)' tool was used on the watershed-raster to create a watershed-polygon and thereafter the watershed-polygon was cleaned up by deleting all the polygons, not visible at full extent. The auto created watershed-polygon were then cross-checked with the Norwegian Water Resources and Energy Directorate stream network, the Oslo Kommune 2017 leaf-off-aerial-photos (Kartverket et al., 2017) and the 1:50,000 topographic maps. In QGIS, the watershed-polygons were adjusted by hand using the topographic lines as reference.

The land-use classifications were derived from the color coding in the 1:50,000 scale topographic raster map from the Norwegian Mapping Authority (Kartverket, 2017). The software QGIS 2.14.20 was used with plugins from GDAL Tools Plugin 1.2.29, GRASS GIS 7.0.3 and SAGA 2.2.3 (Conrad et al., 2016; GDAL Development Team, 2016; QGIS Development Team, 2016). In QGIS with the GDAL tool 'Merge', The topographic raster map tiles were merged together. Then with the GDAL tool 'Polygonize', the merged topographic raster map was converted into a vector layer. The vectorized-topographic-map was then overlaid with a watershed-polygon created in QGIS with the SAGA tool 'Intersect'. An area field was also added to the nearest square meter with the field calculator in QGIS. Color codes were classified into 'water', 'forested', and 'developed'. Black

(outlines) and topographic lines were omitted. Odd colors were reclassified by hand at 1:25,000 scale to their corresponding land-use. Swamps were classified as 'forested' as they are vegetated and were found exclusively in forests. 'Forested' consisted solely of forests and swamps. Agricultural fields made up such a small proportion of the area within the watersheds that they were included in 'developed'. 'Developed' consisted of industrial, urban, suburban, parks, roads and agricultural fields. 'Water' consisted of reservoirs, ponds and streams. The final ESRI Shape file watershed-polygon was exported to a table in .csv format. The resulting land-use-classifications were cross-checked with Google satellite imagery (Google, 2017). The result was a table with the area in meters of land-use patches, within the watersheds feeding the sampled reaches. The final polygon of land-use by watershed, along with the topographic raster map from which it was created, can be seen in Figure 1.

Ecosystem functions

Algae growth

Algae growth was measured on 10x10 cm black granite tiles with the rough side facing up to mimic the natural substrate. There were three different treatments of the granite tiles: granite tiles on the stream bed, granite tiles with a Vaseline bead around the rim on the stream bed, and granite tiles on a raised table with a Vaseline bead around the edge. The Vaseline bead served the purpose of hindering the crawling macroinvertebrate grazers' access to the tiles. This was done to get an estimate of grazing on algae and primary production. One set of the Vaseline treated tiles was placed on a 5 cm raised table to further hinder grazer access and prevent the Vaseline bead from becoming



Figure 2: Photograph of tile arrangement for algae growth and grazing experiment: Photograph taken facing upstream. The bottom set of tiles are tied atop corrugated roofing metal. The top left set and the bottom set of tiles have a bead of Vaseline around the edge.

compromised by debris. The Vaseline was pure white petrolatum produced by Sanivo Pharma and distributed by Apotekene, Norway. The tiles in the Vaseline treatments had a 1 mm thick by 1 cm wide band of Vaseline around the edges and on top of the granite tile. For the Vaseline table treatment, the three tiles were tied atop corrugated roofing metal. The stream bed treatments were tied to plastic mesh 8 cm apart in two rows of three, lengthwise following the stream flow with the Vaseline treatment always being placed towards the right stream bank. It was chosen not to randomize the placement of the tile treatments firstly because it was impossible to do so with the raised table treatment, and secondly due to the difficulty of applying the Vaseline and transporting the tiles without

contaminating the non-Vaseline treatments. This should not be of consequence since the tiles were placed very close together and differential shading should not be an issue since all the streams had similar slope aspects and care was taken to place the tiles away from banks or large rocks.

The tiles were placed in at least 15 cm of water, or in the deepest pool to prevent drying out during low flow, but not in an area of sedimentation or significant turbulence. They were affixed to the stream bed with rebar and heavy stones, holding down the plastic mesh on top and along the sides to minimize tile movement. Care was taken not to create an eddy on the tiles. The table treatment was placed either next to or downstream from the stream bed treatments to avoid any interference that the paint on the roofing metal may have for algae growth. (See Fig. 2 for a photograph of the tile arrangement, and Fig. 3 for a close-up picture of the plastic mesh and fixation method). The tiles were placed out on the 16th and 18th of May and retrieved four weeks later on the 14th and 16th of June, 2017. The tiles were stolen at one restored (HOVresENS) site and one reference site (HOV1), so they were placed out again on May 29th and retrieved with the rest of the sites.

The tiles were checked periodically during their exposure time to remove excess debris buildup, re-expose buried tiles and replace stolen tiles. When they were removed, it was noted if the Vaseline on the stream bed or the Vaseline on the table treatments were untrustworthy for various reasons. There could be too much sediment or debris buildup on the Vaseline to effectively hinder crawling macroinvertebrates or the table tiles could be significantly closer to the water surface than the other treatments. These notes were used for evaluating which method to use for the statistical analysis. Using a plastic template and a razor, a 5x5 cm square was scraped from the center of the damp tile, and placed everything, except large macroinvertebrates, into a preweighed aluminum tray (see Fig. 3).

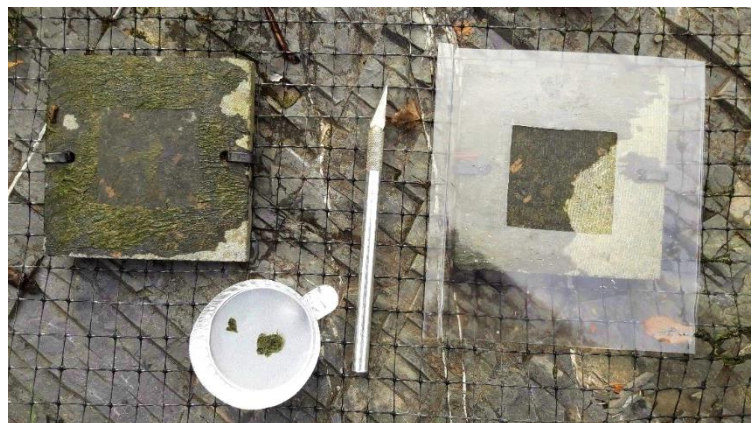


Figure 3: Photograph of tools for removing algae off of the tiles surface after exposure: From left to right: a tile that has had the algae already removed, preweighed aluminum tray, razorblade for scraping, 5x5cm plastic stencil.

The samples were stored by freezing. The frozen samples were placed into the drying oven at 50°C for 48 hours and weighed in grams to two decimal places to get dry weight. Then the samples were burned at 550°C for 2 hours to get ash free dry mass (AFDM) in grams to two decimal places. Due to possible mineral accumulations, dry mass was not used in the analysis; instead the final accumulated mass for each tile was recorded as AFDM in $\mu\text{g}/\text{degree day}$ to compensate for temperature differences between the different sites. The tiles on the ground without Vaseline were

used for algae biomass accrual, the tiles with Vaseline for reducing grazing were used for primary production and the difference between the plain treatment and the reduced grazing treatment was used as grazing pressure.

Leaf litter decay

Two identical litter bag experiments were performed with separate decay mediums to estimate leaf litter decay, one with real leaves and one with cellulose/cotton sponge cloth. The test mediums were specifically *Alnus glutinosa* leaves and cellulose fiber Wettex brand sponge cloths made from 30 percent cotton and 70 percent cellulose. These two treatments methods were



Figure 4: Photograph of setup for litter decay experiment: On top is the leaf litter with the small meshed bags inside the large meshed bags, and on bottom is the Wettex sponge cloth with the small and large meshed bags alternating down the chain.

performed to see if the Wettex sponge cloth could be used as a more convenient and easily standardized alternative to natural leaf litter. The leaves were all collected from the same tree at the same time in October 2016 and air dried before use. The leaves used in 2017 had been stored dry at room temperature. Two different sizes of plastic mesh were used in the litter bags. One with approximately 1cm openings to allow macroinvertebrate access to the litter medium and one with 315 μ m openings to exclude macroinvertebrate access and determine microbial breakdown. The leaf treatment used 15x10cm large meshed bags and 12x5cm small meshed bags. The Wettex sponge-cloth treatment used large meshed bags made of mesh wine-bottle-sleeves, tied shut at the top and bottom, and 12x7cm small mesh bags. Approximately 2 grams of leaves or four 2.5x8.5 cm Wettex sponge-cloths were placed in each bag. The leaves were weighed in grams to two decimal places and the sponge-cloth was weighed in grams to four decimal places. The leaves were soaked in water for 48 hours before being placed in the mesh bags. For the leaf treatment, plastic tags were placed inside the different sized bags for identification purposes; 7x4cm plastic tags for the large meshed bags with the leaves and 2x2cm for the small meshed bags with the leaves. The small meshed bag was then placed inside the large meshed bag before sealing the large meshed bag. Six replicates (one replicate being a large meshed leaf litter pack with a small meshed leaf litter pack inside) were then tied evenly spaced to an 80cm length of stainless-steel chain. For the Wettex sponge-cloth, the plastic identification tags were fastened to the outside of the mesh bags, but the bags were not placed inside each other. Six replicates of the Wettex sponge-cloth litter bags (six large meshed and six small

meshed bags) were then fastened, evenly spaced, to an 80cm length of stainless steel chain alternating between small meshed and large meshed bags, starting with the small meshed bag (see Fig. 4 for a photograph of the litter pack setup).

At each sampled reach, one chain of leaf litter packs and one chain of Wettex sponge-cloth litter packs was fastened to the stream bed by rebar at the top of the chain. They were placed in a pool to prevent drying out in a place where theft would be mitigated, and natural leaf accumulation could occur. Leaf litter packs were placed at all the reference sites in 2016 and all the restored sites in 2017. The Wettex sponge-cloth litter packs were set out in all the sites in 2017. The setout dates were the 7th and 8th of November, 2016 and 9th and 10th of November, 2017. The packages were retrieved four weeks later on the 5th and 6th of December, 2016 and 7th and 8th of December, 2017. In both years, immediately the day after being set out, two sets of fine and large meshed leaf litter packs were taken out of the stream to calculate loss on handling. After removing the litter packs from the stream, they were stored in the freezer until processing.

After thawing the samples in warm water, the leaves were gently rinsed and the sponge-cloth were gently cleaned in warm running water over a sieve to remove excess growth and accumulated debris. Leaves and Wettex sponge-cloth were then placed in preweighed aluminum trays and dried at 50°C for 48 hours. The dry weight was measured in grams to four decimal places. The leaves and Wettex sponge-cloths were burned at 550°C for 2 hours to get Ash-Free-Dry-Mass (AFDM). AFDM was measured in grams to four decimals. Six extra trays of sponge-cloths were also dried and burned to calculate the mineral content of fresh cloth strips, which compensated for loss on handling and air humidity.

The final results were recorded per litter bag as the exponential decay coefficient k according to the following formula: $(M_t/M_0) = e^{(-kt)}$ (Graca, Bärlocher, & Gessner, 2005). M_t is the final AFDM at time t , M_0 is the initial AFDM and t is time in degree days. The mass was recorded as AFDM due to mineral accumulations and time was recorded as degree days to account for temperature differences between the sites. M_0 was calculated as the initial-airdried-weight times the correction factor D . The correction factor D was calculated from the samples that were not put out for full exposure in the streams and was calculated as the average of AFDM/initial-airdried-weight. The correction factor D converts the initial-airdried-weight to AFDM while correcting for air humidity and loss on handling. The correction factor D was calculated for Wettex, leaf litter in small meshed bags and leaf litter in large meshed bags. The correction factor was not calculated separately with the Wettex for the small and large meshed bags because the sponge cloth is much less brittle than the leaves. Therefore, loss on handling was not a concern in this case.

Statistical analysis

The statistics were performed in R version 3.2.3 using packages lme4 1.1-17 for performing mixed effects modelling with the 'lme' and 'anova.lme' functions, multcomp 1.4-8 for Tukey's HSD tests on mixed effects models with the 'glht' function and vegan 2.4-6 for cluster analysis with the 'vegdist' function (Bates et al., 2018; Hothorn et al., 2017; Oksanen et al., 2018; The R Foundation for Statistical Computing, 2015).

There were two methods used for hindering macroinvertebrate grazers' access to the tiles (Vaseline and raised tiles with Vaseline) and two methods used for determining litter decay (leaf litter and Wettex sponge cloth). Paired t-tests between the site averages and Tukey-mean difference plots were used to evaluate which methods to use for the analysis.

The sites were first grouped into three pollution levels across which the data could be compared to assess if pollution had an effect on ecosystem function and if restored sites responded differently. The pollution level groupings were established using hierarchical clustering on Jaccard similarity coefficients from the water chemistry variables ammonium, phosphate and BOD₅. Jaccard similarity coefficients were used due to the non-normal distribution of the data. The validity of the groupings was assessed using ANOVA and Tukey's HSD tests.

ANCOVA on linear mixed effects models was used to assess whether there were differences in ecosystem function between these pollution groupings as well as between reference and restored sites. Site and sampling year were random effects. Pollution grouping and reference/restored were combined as an interaction with no main effects in the model. The ecosystem function was log transformed to normalize the data. If that ecosystem function variable had negative values, then 1.1 times the minimum value was added to all the values to make them all positive, for the log transformation to work.

To assess if other environmental variables besides pollution and restoration were influencing ecosystem function, linear models of the ecosystem functions were built using all the measured variables. The set of variables with which to do the model selection was determined by drawing a concept map of how the different variables should fit into the model and rejecting all the variables that either did not have enough variability or data of interest or those that correlated too strongly with other variables. The correlation was based on Pearson correlation coefficients. Due to the large number of variables, the criteria for rejection from use in the model selection was kept low, at $r = 0.50$, if the variables would logically be covariates. All variables correlating over $r = 0.50$ happened to be in theory, logical covariates. All the values used in the models were averages per site. To normalize the data, the model selection was performed on log transformed ecosystem functions. In

order to log transform ecosystem function variables with negative values, 1.1 times the minimum value was added to all the values. The data set was too small to perform automated AIC model selection, so stepwise regression with bidirectional elimination was performed. Both adjusted R^2 and variable significance values were considered in the selection. The modelling goal was maximum parsimony, so only variables with a p-value around 0.05 or less, were kept in the model. Statistically significant variables with dubious relevancies, determined from the graphical representations, were removed.

Results

Determining groupings for levels of pollution in the sampled reaches

Based on water chemistry, the sampled sites were grouped into three pollution levels, for the purpose of evaluating the effect of pollution on ecosystem functions for reference sites and restored sites. The clustering was based on phosphate (PO_4^{3-}), ammonium (NH_4^+) and BOD_5 by low water level and BOD_5 by medium water level. The BOD_5 at high water level didn't show any consistent patterns across the sites so it was left out of the evaluation process. BOD_5 at medium level only showed noticeably greater values at 2 sites, which happened to be both placed into pollution group 3 (see Appendix 1 for measured values by site). Figure 5 shows the clustering of the sampled sites using average linkage. The same exact groupings were also made when single and complete linkage was used.

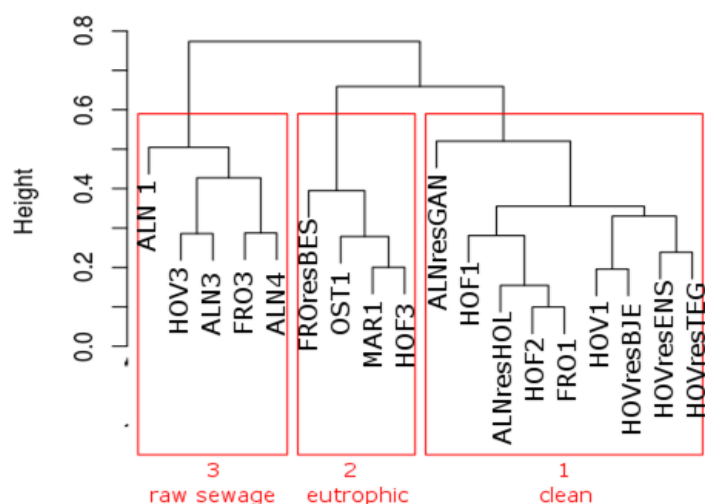
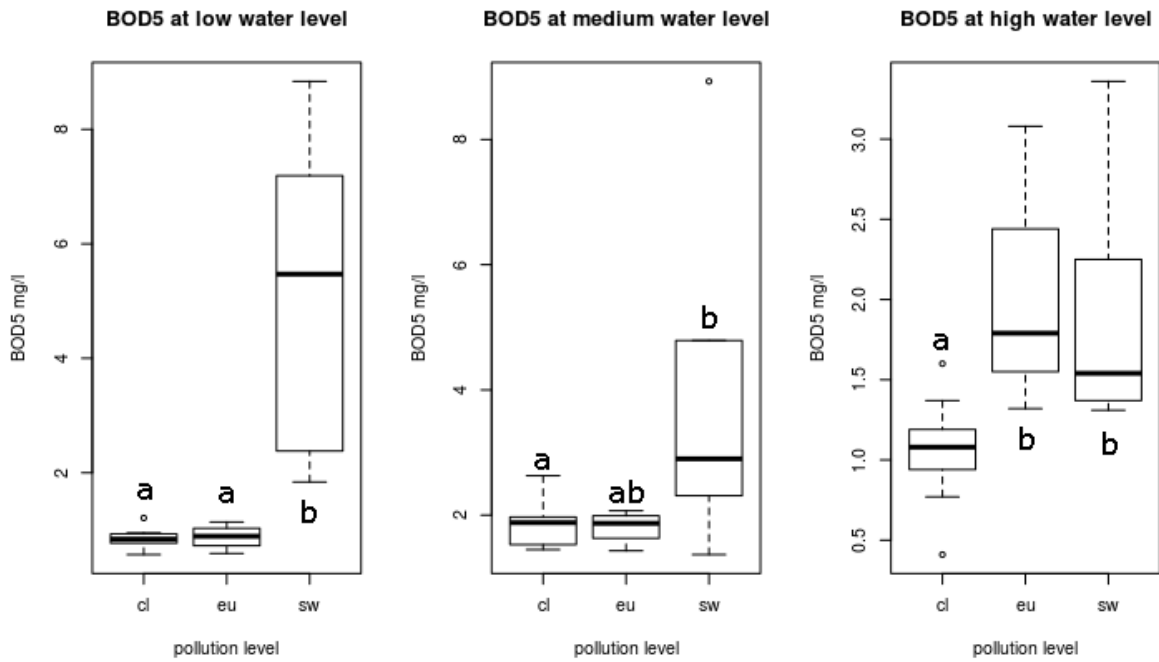


Figure 5: Cluster dendrogram of sampled stream reaches cut to three pollution groupings: Based on water pollution indices: PO_4^{3-} , NH_4^+ and BOD_5 by low water level and BOD_5 by medium water level. From average linkage of Jaccard indices.

The proposed groupings for the pollution levels were significantly different, as seen in the boxplots and Tukey test (see Figs. 6 and 7, and Table 2). However, BOD_5 was increasingly less significant with increased water level, but a trend was still apparent (see Fig. 6). The proposed pollution levels could be classified as level 1 being relatively clean, level 2 as eutrophic with elevated phosphate and level 3 as raw sewage with elevated levels of both phosphate, ammonium and BOD_5 (see Figs. 6 and 7, and Appendix 1 for summary table by site).



Figure

6: Boxplots of BOD₅ for all reaches by pollution level for low, medium and high water level: letters show Tukey's HSD groupings at $\alpha = 0.05$. Pollution level "cl" is clean, "eu" is eutrophic and "sw" is raw sewage.

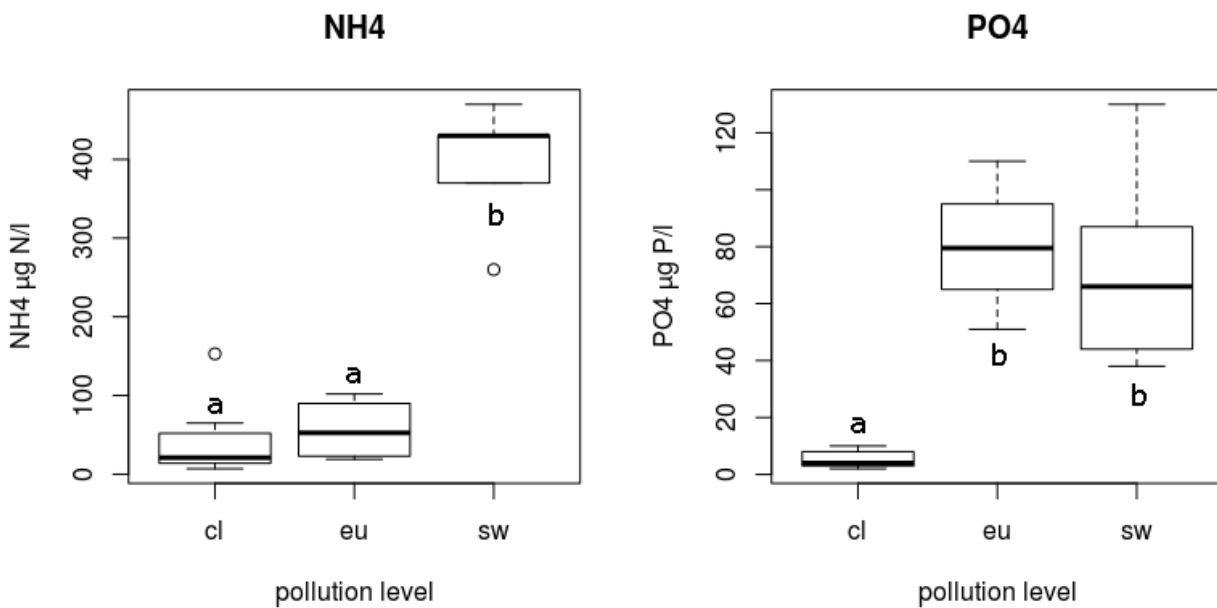


Figure 7: Boxplots of NH₄⁺ and PO₄³⁻ levels for all reaches by pollution level: letters show Tukey's HSD groupings at $\alpha = 0.05$. Pollution level "cl" is clean, "eu" is eutrophic and "sw" is raw sewage.

Table 2: Summary of ANOVA and Tukey's HSD test outputs: Pollution level "cl" is clean, "eu" is eutrophic and "sw" is raw sewage.

Variable	Tukey's HSD P-values by grouping combinations			ANOVA P-values
	cl-eu	cl-sw	eu-sw	
BOD ₅ low water	0.98	0.00*	0.00*	0.00*
BOD ₅ medium water	1.00	0.04*	0.09	0.04*
BOD ₅ high water	0.03*	0.03*	0.99	0.01*
NH ₄ ⁺	0.90	0.00*	0.00*	0.00*
PO ₄ ³⁻	0.00*	0.00*	0.90	0.00*

* indicate significant p-values with $\alpha = 0.05$.

Algae growth and grazing

Methods testing between two different procedures for reduced grazing on tiles:

Two different methods were used for inhibiting grazing on the tiles to measure grazing pressure: tiles on the ground with a Vaseline bead around the edge, and tiles raised up on a small table with a Vaseline bead around the edge. The grazing was then calculated as the difference between the tiles with inhibited grazing, and the simple tiles on the ground. According to the paired t-test, there was no significant difference in the calculated grazing ($t = 0.20$, $df = 15$, $p = 0.84$). As made apparent in the scatter plot of the calculated grazing for ground vs raised tiles, there was no correlation between the values of the two methods, but the raised tiles had much more dispersion (see Fig. 8). According to the Tukey mean difference plots, there was a proportional difference (see Fig. 9), and it was consistent regardless of the magnitude of growth; however this was most likely an artifact of the large discrepancy in dispersion between the raised and ground tile treatments. Most notable from the Tukey mean difference plots, was that the effect was consistent across pollution groupings and reference vs restored sites. The tiles were placed in the field on uneven rocky surfaces, so the table tiles usually were not closer to the water surface. However, 6 out of 16 table tile treatments were still noted as untrustworthy for being either too close to the water surface or having significantly different levels of sediment accumulation as compared to the ground tiles. Only 2 out of 17 ground tile Vaseline treatments were noted as untrustworthy due to significant sediment accumulation on the Vaseline. Also, during flood events, both the Vaseline treated and untreated tiles on the ground were swept over and occasionally buried by stones, while the raised tiles were always free from this scoring and never buried. This difference in conditions experienced by the ground tiles and the raised tiles, in combination with the apparent lack of correlation between the two methods, lead to the conclusion that the raised-grazer-inhibited tiles could not be confidently compared to the ground-grazer-uninhibited tiles to calculate grazing. The raised tiles could still serve the purpose of measuring primary production, since they were not as scoured by stones as the other two treatments (see Fig. 10 for boxplots of algae accrual by treatment method). However, paradoxically at some of the sites there was substantially less growth on the raised tiles where grazing was inhibited than on the ground tiles where grazing was not, as seen by the large negative grazing values (see Fig. 8). Thus, the ground tiles with Vaseline were used to calculate grazing and primary production for all further results.

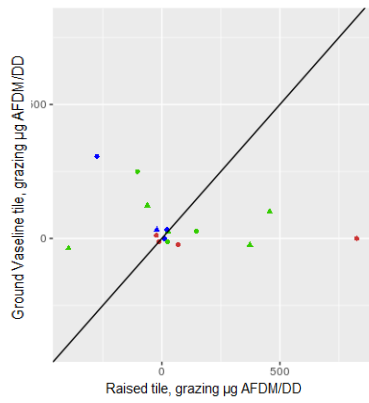


Figure 8: Grazing calculated from raised tile treatment vs ground tile treatment, both with Vaseline: The black line is for reference and has a slope of 1.

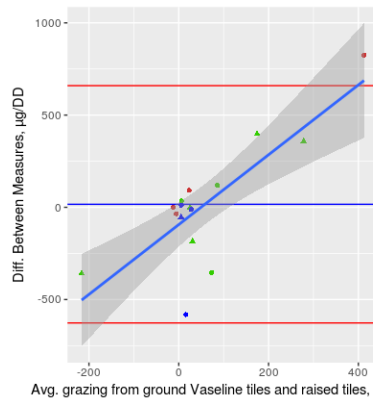


Figure 9: Tukey mean-difference plot of grazing calculated from raised tile treatment vs ground tile treatment, both with Vaseline: The x-axis is the average of the calculated grazing from both methods by site, and the y-axis is the difference between the calculated grazing from the two methods expressed either in $\mu\text{g/degree day}$ or percent of the mean. The thick blue line is the best fit line and the shaded area is the standard error. The three horizontal lines correspond to the upper and lower 95% limits of agreement (red) and the mean (blue).

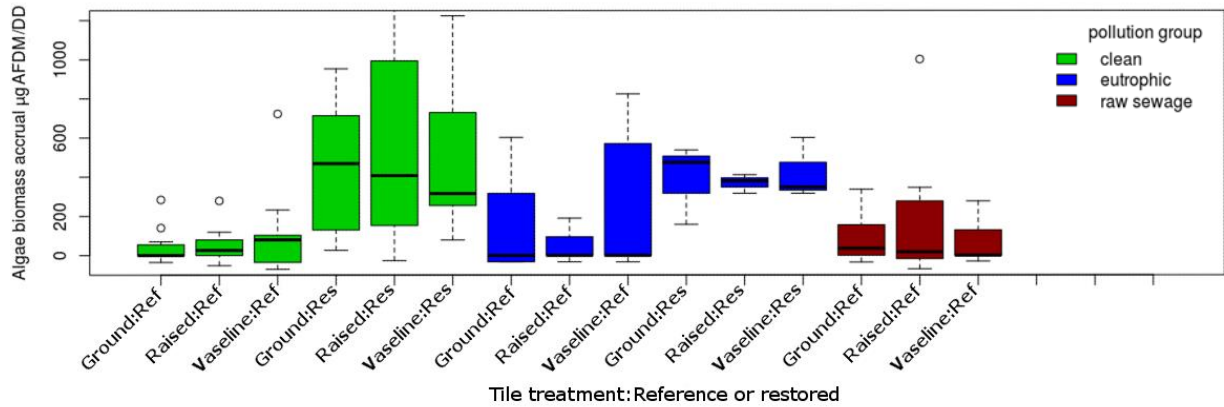
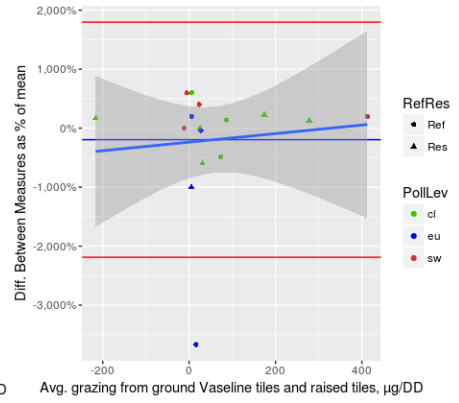


Figure 10: Boxplots of algae mass accrual by tile treatment, reference vs restored and pollution level: Tiles treated as ground were simply placed on the stream bed, Vaseline treatment had a bead of Vaseline around the edge to inhibit grazer access, and raised treatments were placed on a platform and had a Vaseline bead to inhibit grazer access.

Algae growth and grazing:

With an $\alpha=0.05$, according to a Tukey's HSD test on a log transformed mixed effect model with site as a random effect, the only significant difference for algae growth was between the reference and restored sites of the unpolluted streams ($p=0.04$) (see Fig. 11). There were no statistically significant differences between groupings for primary production (ANOVA, $p=0.12$). However, in general, restored sites showed more algae growth compared to the non-restored reference sites in both primary production and net algae growth. There was no observable pattern for grazing pressure by pollution level or restored vs non-restored (ANOVA, $p=0.59$).

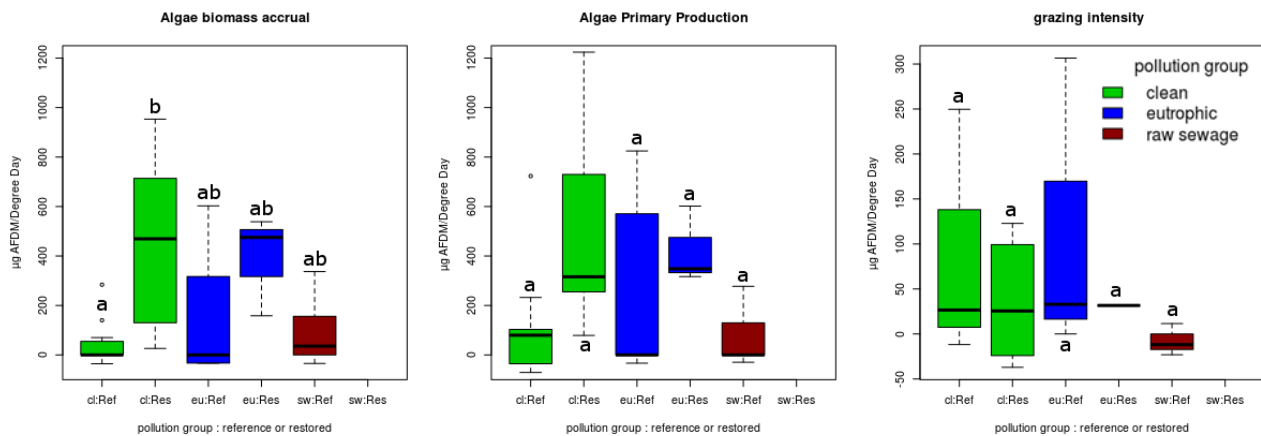


Figure 11: Boxplots of algae accrual, primary production and grazing of algae on granite tiles: The letters show Tukey's HSD groupings at $\alpha = 0.05$.

Litter decay:

Methods testing between using Wettex and leaf litter as decay substrate:

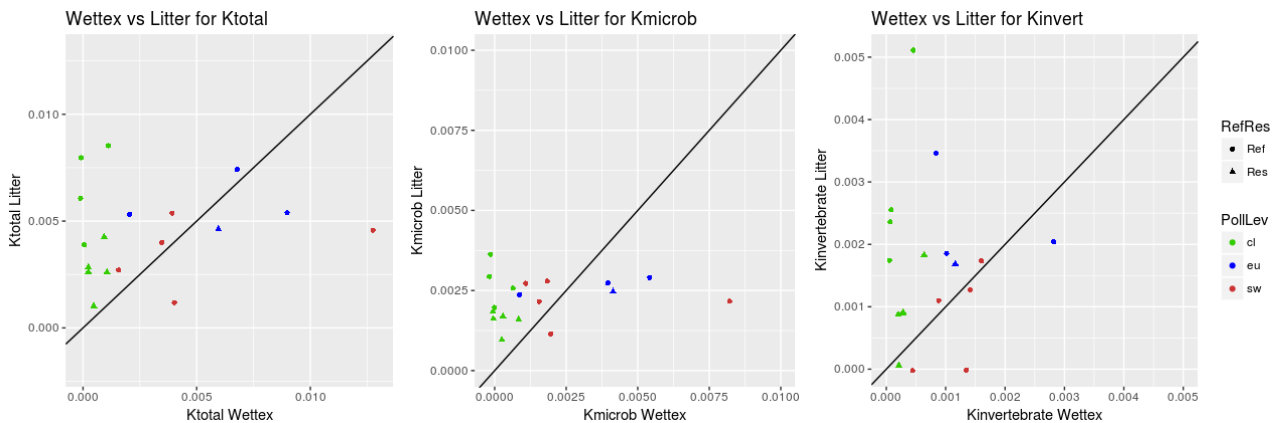


Figure 12: Wettex vs Litter for the three K decay constants (k_{total} , $k_{microbial}$, $k_{invertebrate}$): The black line has a slope of 1 and is for reference.

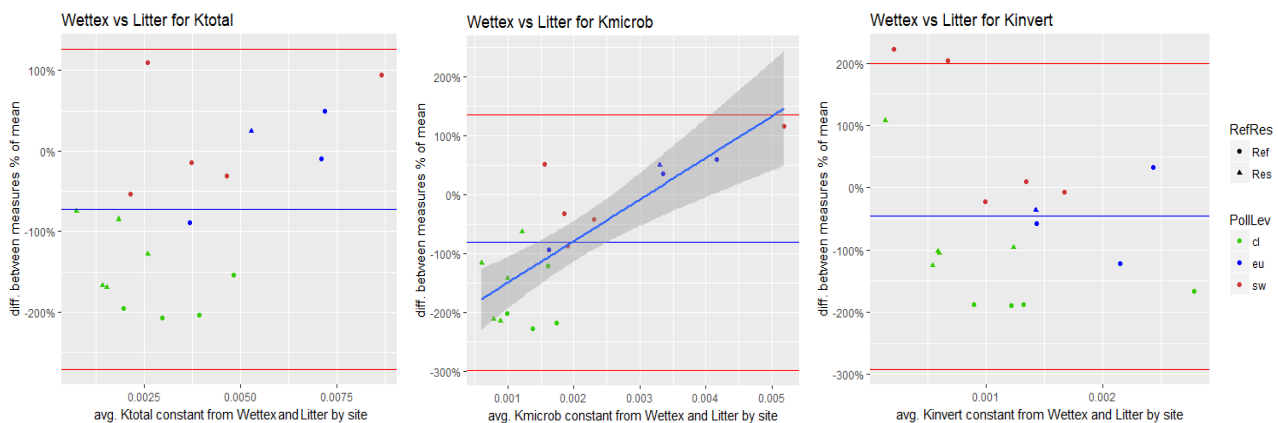


Figure 13: Tukey mean-difference plots of Wettex vs Litter for the three K decay constants (k_{total} , $k_{microbial}$, $k_{invertebrate}$): The x-axis is the average of the k decay constants from both methods by site, and the y-axis is the difference between the k decay constants from the two methods expressed in percent of the mean. The thick blue line is the best fit line and the shaded area is the standard error. The three horizontal lines correspond to the upper and lower 95% limits of agreement (red) and the mean (blue).

Litter decay was measured both with natural leaves and Wettex cotton-cellulose sponge-cloth. According to paired t-tests, there was no significant difference for decay factors k_{total} or $k_{microbial}$ between methods (k_{total} : $t=-1.62$, $df=17$, $p=0.12$, $k_{microbial}$: $t=-0.99$, $df=17$, $p=0.33$). However, there was a significant difference in $k_{invertebrate}$ ($t=-2.55$, $df=17$, $p=0.02$). When looking at the scatter plots, it is very clear that the Wettex method had much more dispersion and better distinction between pollution groupings for total and microbial induced decomposition (see Fig. 12). Nevertheless, the same general pattern still existed with the leaf litter method with the clean sites having the lowest decomposition and eutrophic sites having the highest, but there was very heavy overlap in pollution groupings (see Figs. 12 and 14). In the Tukey mean difference plots for k_{total} and $k_{microbial}$, there was an inconsistent proportional difference with the high decomposition rates having more decomposition in the Wettex and the low decomposition rates having less decomposition in the Wettex. This trend was very clear for $k_{microbial}$ where the samples were in small meshed bags, where there was nothing confounding from macroinvertebrate activity or samples breaking and washing away (see Fig. 13).

This trend was consistent with the observation that the Wettex at first was much less brittle than the leaves, but had the potential to decay to nearly nothing, where the leaves decayed to a hard skeleton. These trends were not observed for $k_{\text{invertebrate}}$. Instead according to the Tukey mean difference plot, there was a consistent proportional difference of $46\% \pm 62\%$ less macroinvertebrate mediated decay on the Wettex, as compared to the leaves (see Fig. 13). This difference was greater for the “clean” pollution group, where the other two pollution groupings had a difference closer to zero. Because of the inconsistent proportional difference observed between the two methods, the decay factor k could not be compared directly between the two methods. Also, the lower grazing values observed in the Wettex treatment, especially in the “clean” pollution grouping, should be interpreted with care. The cause of this disparity was not known, as it may have been due to the brittleness of leaves over Wettex at low decomposition levels and the extreme softness of Wettex when highly decomposed, or it may have been due to grazer selection preferences. Thus, it was chosen to analyze both the leaf litter and the Wettex cellulose-cotton sponge-cloth, despite the greater statistical power provided by the Wettex.

Leaf litter decay:

With an $\alpha = 0.05$, with an ANOVA on a log transformed mixed effect model with site and sample year being random effects, there were no significant differences between pollution groupings or restored vs unrestored (k_{total} : $p=0.11$, $k_{\text{microbial}}$: $p=0.26$, $k_{\text{invertebrate}}$: $p=0.20$). But graphically, the only noticeable difference was that nonrestored sites in the cleanest two pollution groupings had increased invertebrate mediated decomposition, which was reflected in the total decomposition rates (see Fig. 14). Otherwise, microbial decomposition was consistent across all pollution groupings and restored vs unrestored sites.

Wettex cotton-cellulose sponge cloth decay:

An ANOVA on a log transformed mixed effect model with site as the random effect, showed significant differences within all three decay factors (k_{total} : $p=0.00$, $k_{\text{microbial}}$: $p=0.00$, $k_{\text{invertebrate}}$: $p=0.00$). With an $\alpha = 0.05$, the Tukey’s HSD test only showed distinction between decomposition rates of the unpolluted and two polluted groupings. Graphically, the restored sites always lined up well within their respective pollution grouping (see Fig. 15).

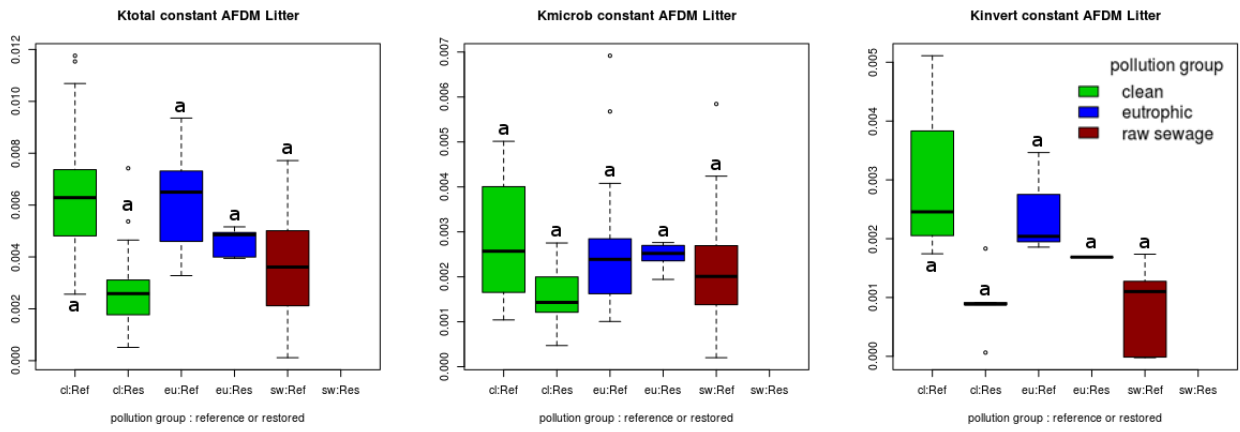


Figure 14: Boxplots of exponential decay constant k from leaf litter for total, microbial and invertebrate mediated decomposition: The letters show Tukey's HSD groupings at $\alpha = 0.05$.

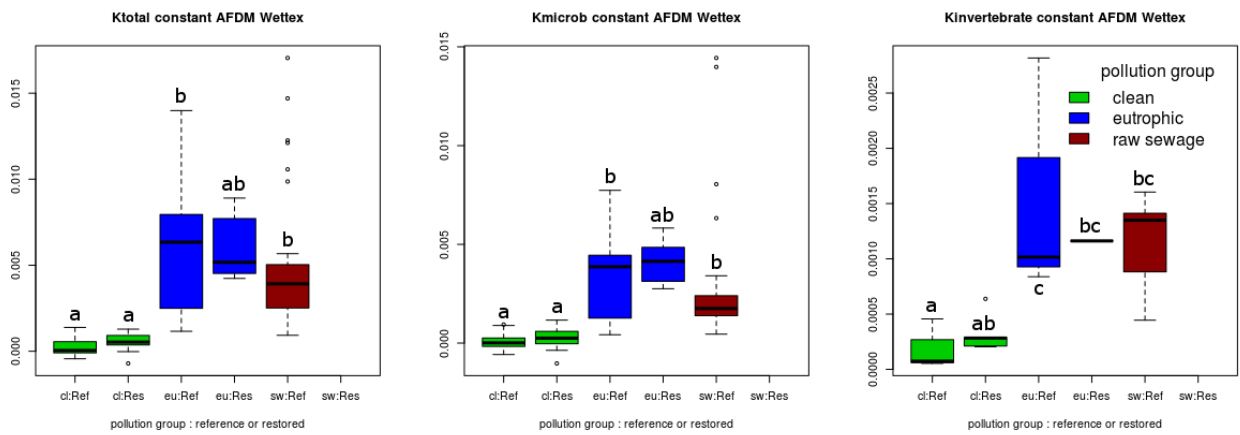


Figure 15: Boxplots of exponential decay constant k from Wettex sponge cloth for total, microbial and invertebrate mediated decomposition: The letters show Tukey's HSD groupings at $\alpha = 0.05$.

Modelling ecosystem function

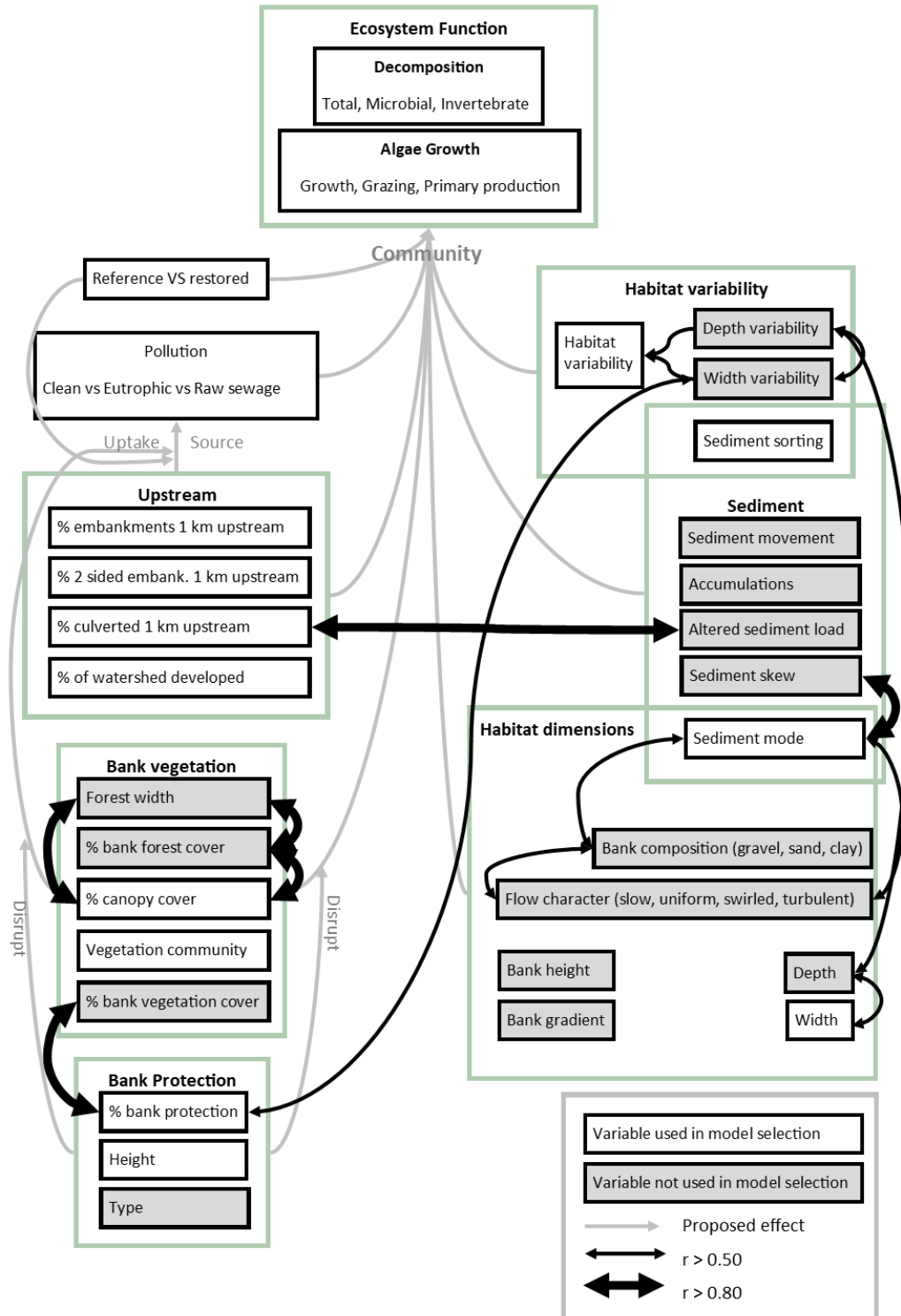


Figure 16: Diagram of proposed model for ecosystem function showing correlation between site specific environmental variables and which variables were used in the final model building: “r” is the person correlation coefficient. Variables were rejected from model building due to correlation with other explanatory variables or lack of data.

The variables used in the final model selection process, along with the variables that they are correlated with, and the assumed effect on ecosystem function can be seen in Figure 16. It was assumed that the ecosystem functions are influenced by biotic communities which in turn would be affected by the variable groupings: bank vegetation, habitat dimensions, sediment character, habitat

variability, restoration status, upstream alterations, watershed characteristics and water pollution. The variable grouping bank protection could indirectly affect ecosystem functions by disrupting connectivity between the bank and the stream or by decreasing habitat variability. In addition, measured water pollution could be influenced by some of the same variables, such as the percent watershed developed as a pollution source, upstream alterations and restoration status as modulating factors and bank vegetation as possible uptake agents which could be disconnected from the stream by bank protection. Specific variables were not used either due to correlation with other variables or lack of data. Bank height and gradient were not used due to lack of variability between sites and sediment movement. Altered sediment load was not used due to the very low representation within the small sample size (see Appendix 1). All the forest related variables (% riparian forest bank cover, % canopy cover and riparian forest width) were strongly correlated with each other, so only percent canopy cover was used in the modelling since it is assumed to have the most direct effect on primary production. The percent bank protection and the percent bank vegetation cover were very highly correlated ($r = -0.90$) so only bank protection was used since that is generally the variable intentionally manipulated by river works. Width variability and depth variability were averaged together to a new variable called habitat variability, because they correlated both with each other and with bank protection and stream depth respectively. Stream depth correlated with both depth variability and stream width, so it was not used. Sediment mode, bank sediment composition and flow character were all correlated, so only sediment mode was used since it also correlated strongly with sediment skew. (see Appendix 2 for correlation matrixes)

Modelling pollution levels from upstream sources

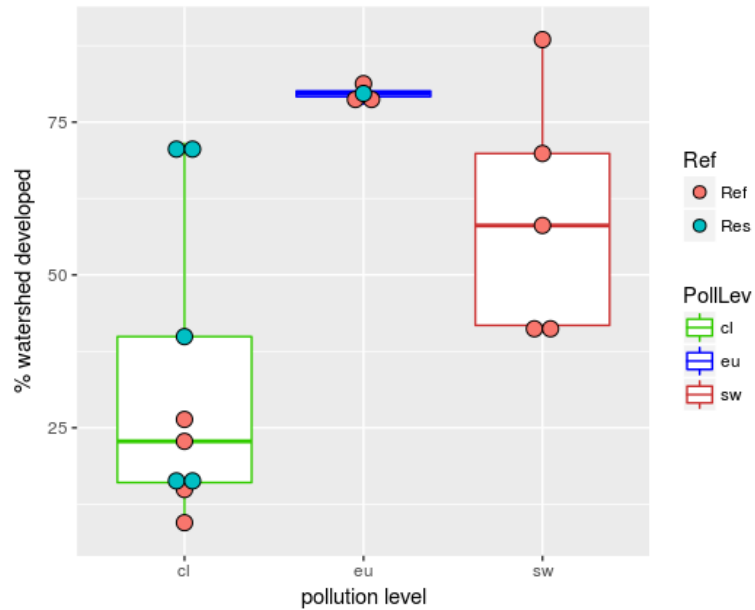


Figure 17: Final model for pollution level from upstream sources. % watershed developed by pollution level: Restored sites are color coded for reference but that was not a significant variable. The pollution levels are clean, eutrophic and raw sewage.

The first step taken to model the ecosystem functions was to model the pollution in the sampled reaches. Reference vs restored, bank protection, bank vegetation and upstream variables were used for the model selection (see Fig. 16). In the final model, it was found **pollution level was a function of percent watershed developed** and had an adjusted R^2 of 0.47 and an ANOVA p-value of 0.00 (see Fig. 17; see Appendix 2 for model parameters). Clean stretches had watersheds with a less developed land area than the polluted eutrophic and raw sewage stretches. The relative size of watersheds, the dominant land use and the pollution level of the study sites can be seen in Figure 1.

Modelling algae

The final models for the ecosystem functions regarding algae are in Appendix 2; in summary they were:

Algae biomass accrual ~ % canopy cover $R^2 = 0.70$

Canopy cover: decreased algae biomass accrual with an increase in % canopy cover

Algae primary production ~ % canopy cover $R^2 = 0.60$

Canopy cover: decreased algae biomass accrual with an increase in % canopy cover

Grazing intensity ~ pollution level + stream width Adj $R^2 = 0.67$

Stream width: stream width has a positive effect on grazing intensity for clean sites

Pollution level: significant as an interaction with stream width

(See Figs. 18 and 19 for graphs of significant variables)

In the modelling of biomass accrual and primary production, canopy cover was found to be the only significant variable in both the models. For grazing intensity, stream width was significant for the clean sites.

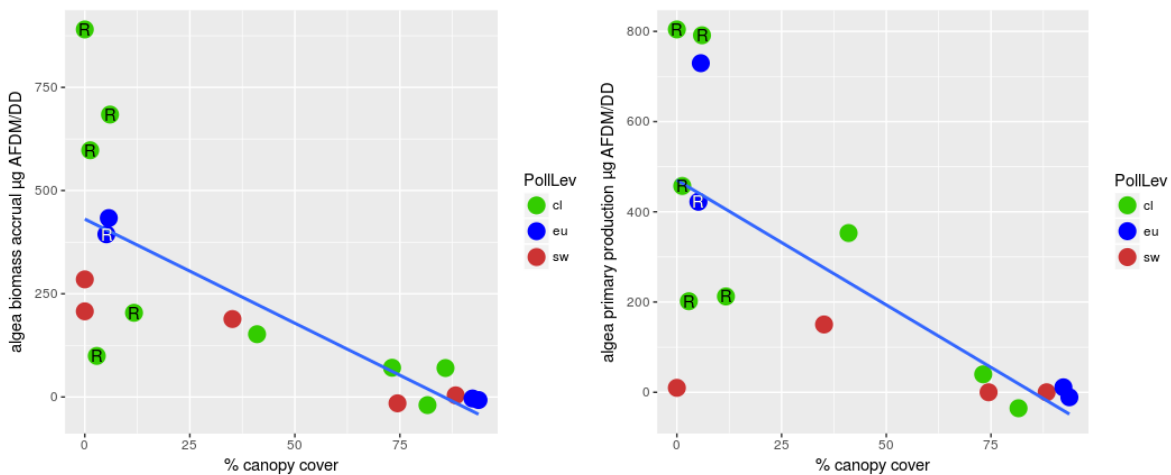


Figure 18: Graphs of algae biomass accrual and primary production by percent canopy cover: Pollution levels are clean, eutrophic and raw sewage. Restored sites are labeled with an R for reference but is not a significant variable.

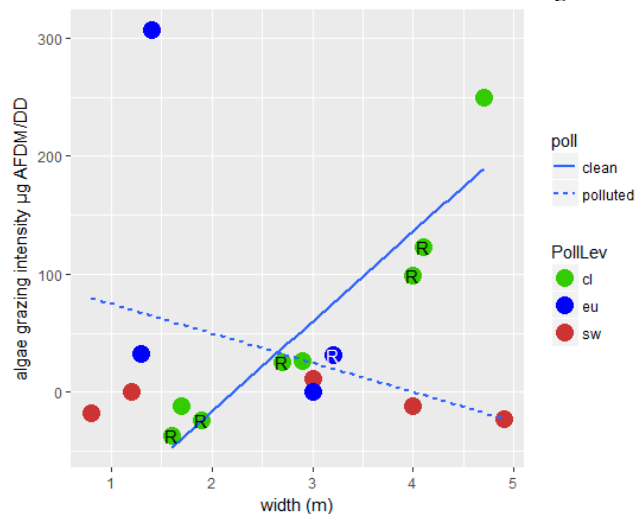


Figure 19: Graphs of algae grazing intensity by stream width: Pollution levels are clean, eutrophic and raw sewage. Restoration was not a significant variable but it is displayed for reference.

Modelling decomposition

The final models for ecosystem functions regarding decomposition are in Appendix 2; in summary, they were:

Wettex

Total decomposition ~ stream width + sediment mode + pollution level + vegetation community Adj R² = 0.94

Stream width: has a positive effect on total decomposition

Sediment mode: increase in sediment size has a negative effect on total decomposition

Pollution level: has an increasingly positive effect with increasing levels of pollution

Vegetation community: deciduous forest has the highest decomposition rates followed by pioneer meadow, then meadow riparian scrub, mixed forest, and lastly meadow

Microbial decomposition ~ sediment mode + pollution level + vegetation community Adj R² = 0.90

Sediment mode: increase in sediment size has a negative effect on total decomposition

Pollution level: has an increasingly positive effect with increasing levels of pollution

Vegetation community: deciduous forest and pioneer meadow have the highest decomposition rates followed by meadow riparian scrub, mixed forest, and lastly meadow

Invertebrate decomposition ~ habitat variability + pollution level Adj R² = 0.73

Habitat variability: has a positive effect when the streams were polluted

Pollution level: significant as an interaction with habitat variability

Leaf litter

Total decomposition ~ % culverted 1 km upstream + vegetation community Adj R² = 0.62

Culverted 1 km upstream: negative effect on total decomposition

Vegetation community: mixed forest and deciduous forest have the highest decomposition rates with pioneer meadow, meadow and meadow riparian scrub having the least

Microbial decomposition ~ % culverted 1 km upstream + vegetation community Adj R² = 0.68

Culverted 1 km upstream: negative effect on total decomposition

Vegetation community: mixed forest has the highest decomposition rates followed by deciduous forest and meadow, then pioneer meadow and meadow riparian scrub having the least

Invertebrate decomposition ~ pollution level + vegetation community Adj R² = 0.58

Pollution level: raw sewage affected streams have lower invertebrate decomposition

Vegetation community: mixed forest and deciduous forest have the highest decomposition rates followed by meadow riparian scrub and pioneer meadow and then meadow having the least

(See Figs. 20 to 24 for graphs of significant variables; see Appendix 2 for model parameters)

Nearly the same models were found for k_{total} and $k_{\text{microbial}}$. Vegetation community was significant in nearly all the models. The only decomposition factor that vegetation community was not significant for was $k_{\text{invertebrate}}$ for the Wettex sponge cloth. Wettex cellulose sponge cloth was additionally affected by sediment mode, stream width, habitat variability and pollution level. Leaf litter was additionally affected by percent culverted 1 km upstream and pollution level.

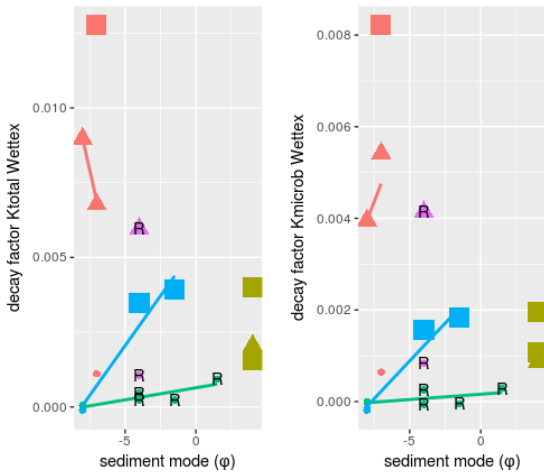


Figure 20: Graphs of exponential decay factors k_{total} and $k_{microbial}$ by sediment mode, vegetation community and pollution level for of Wettex sponge cloth: Pollution levels are clean, eutrophic and raw sewage. Restored sites are labeled with an R for reference but are actually not a significant variable in the model. Sediment mode is displayed in units of Phi (ϕ) so smaller values are larger stones

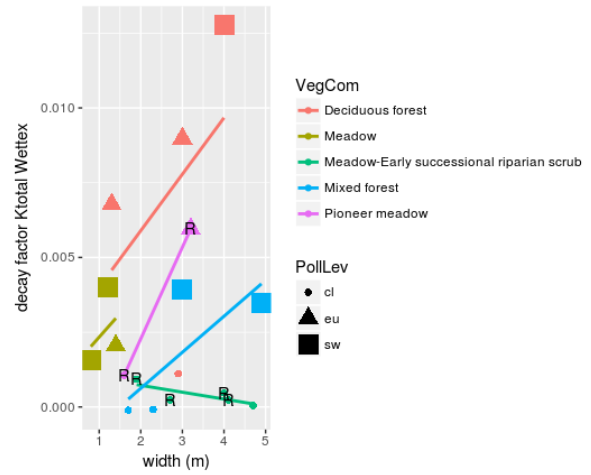


Figure 21: Graph of exponential decay factor k_{total} by stream width, vegetation community and pollution level for of Wettex sponge cloth: Pollution levels are clean, eutrophic and raw sewage. Restored sites are labeled with an R for reference but are actually not a significant variable in the model.

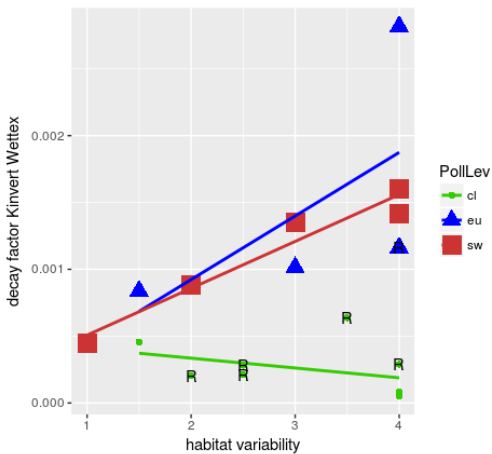


Figure 22: Graph of exponential decay factor $k_{invertebrate}$ by habitat variability and pollution level for of Wettex sponge cloth: Pollution levels are clean, eutrophic and raw sewage. Restored sites are labeled with an R for reference but are actually not a significant variable in the model. Habitat variability is a 4-point qualitative scale with 1 being the lowest.

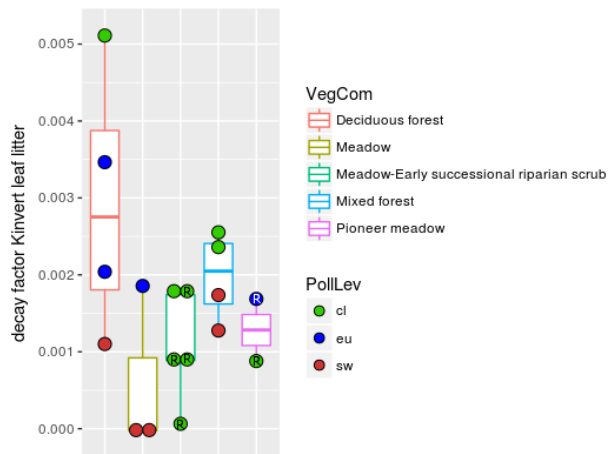


Figure 23: Graph of exponential decay factor $k_{invertebrate}$ by vegetation community and pollution level for of leaf litter: Pollution levels are clean, eutrophic and raw sewage. Restored sites are labeled with an R for reference but are actually not a significant variable in the model.

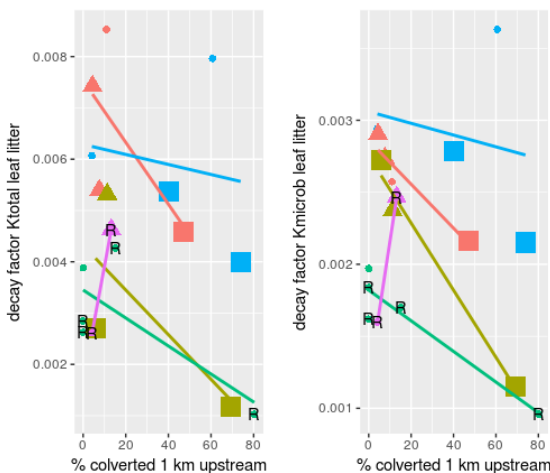


Figure 24: Graph of exponential decay factor k_{total} and $k_{microbial}$ by percent culverted 1 km upstream, vegetation community and pollution level for of leaf litter: Pollution levels are clean, eutrophic and raw sewage, but are not significant and only displayed for reference. Restored sites are labeled with an R for reference but are actually not a significant variable in the model.

Discussion

Pollution: Merits of the three levels and pollution sources

Using phosphorous (P), nitrogen (N) and five-day biological oxygen demand (BOD₅) for analyzing the effect of pollution on ecosystem functions is a credible method. The use of hierarchical cluster analysis to group similar sample sites has been shown to provide good results in water pollution assessments (Alberto et al., 2001; Singh, Malik, Mohan, & Sinha, 2004). BOD₅ is one of the few variables that is a reliable indicator of raw untreated sewage and can be directly linked to hypoxia events (Howard, Espigares, Lardelli, Martin, & Espigares, 2004; Mallin, Johnson, Ensign, & MacPherson, 2006). Nitrogen (N) and phosphorous (P) levels have been used in many studies since they are the two macronutrients needed by plants and thus are of particular interest regarding eutrophication (e.g., Dodds, Smith, & Lohman, 2002; Elser et al., 2007; Francoeur, 2001; Harpole et al., 2011; Mallin, Johnson, & Ensign, 2009; Mallin, Parsons, Johnson, McIver, & CoVan, 2004). Even over multiple ecoregions, most of the variation in algae biomass can be attributed to P and N (Dodds et al., 2002). It is often stated that according to Liebig's law of the minimum only the one nutrient that is in least supply relative to the organisms needs will be limiting, and thus of interest (as cited in Ågren, Wetterstedt, & Billberger, 2012). However in multispecies communities, it is very unlikely that a single nutrient is limiting and multiple nutrient additions will often be synergistic (Elser et al., 2007; Francoeur, 2001; Harpole et al., 2011). Therefore, this study analyzed both N and P simultaneously.

The three groupings that were created using the hierarchical cluster analysis have ecologically meaningful nutrient level ranges. The groupings created by the clustering were clean, eutrophic and raw sewage. Both the eutrophic and the raw sewage groupings had elevated levels of phosphorus and the raw sewage group had additionally elevated levels of nitrogen. (Dodds et al. (2002) found that across all North America encompassing multiple ecoregions there was a breaking point of 30 total µg P/l and 40 total µg N/l above which there was greater algae growth and these breaking points lined up perfectly with the separation between the clean and eutrophic clustered groups for P and the raw sewage group being significantly over this breaking point for N.

Additionally, the pattern seen in the groupings spatially and temporally regarding P, N and BOD₅ are consistent with other studies. By low water levels, only the raw sewage clustered pollution grouping had elevated BOD₅ levels; however, by high water levels, the raw sewage groupings levels lowered and the eutrophic grouping's BOD₅ levels increased to match that of the raw sewage grouping. This same pattern of highly polluted streams decreasing BOD levels during rain events and medium polluted streams increasing pollution levels was seen in a study by Mallin, Johnson, and Ensign (2009). This is likely a result of the raw sewage grouping always receiving sewage inputs, so,

by high water flows, it becomes diluted; however, the eutrophic sites become newly affected by sewage inputs from storm overflows during high water events. The same study also found that increased N was also associated with increased BOD, just as in this study. It was also found that increased development of the watershed resulted in streams with higher nutrient loadings and BOD₅ levels. Many studies have found relationships between development of a watershed and increased levels on N, P and BOD (Bannerman, Owens, Dodds, & Homewer, 1993; Holland et al., 2004; Line, Arnold, Jennings, & Wu, 1996; Mallin et al., 2009). As seen in Figure 17, there was not a clean relationship between pollution level and percent development of the watershed; the mid pollution level generally had the most developed watersheds and some of the clean sites had the same level of development in their watersheds as the heavily polluted raw sewage affected sites. This may be a result of differing land uses within the city; however, in other studies it has been found that residential and industrial areas do not differ significantly in their effect on stream pollution (Holland et al., 2004; Mallin et al., 2009). All the clean sites that had relatively large amounts of development in their watersheds were restored sites. It is very reasonable to assume that the municipality put in more effort to clean up the restored streams than the natural ones, resulting in this overlap seen in the restored streams and the sewage affected streams. Also, whether a sewage line is leaky, should not necessarily correlate with the development of the watershed. The exact relationship seen between the two polluted groupings may be happenstance, with the most highly polluted sites having less development in their watersheds than the mid polluted sites. Nevertheless, there was significantly more pollution in streams with highly developed watersheds, and this is in accordance with literature.

Algae

Analysis of methods

Regarding measuring biomass accrual, little could be done to improve the replication of natural conditions without switching from tiles over to natural stones. Tiles have been shown to decrease variability in measurements but the use of ceramic tiles has been cautioned against as it is not a natural substrate (Morin & Cattaneo, 1992; Shilling & Davis, 2005). Granite tiles as opposed to ceramic tiles were used in this study to mimic the natural substrates, but still maintain the uniformity not achievable by natural river stones. The tiles were only left out in the streams for a month and this is often not long enough for natural levels of algae ash-free dry mass accumulation, Lamberti & Resh (1985) determined that 3 months was necessary. However a full cycle of periphyton colonization, growth, and sloughing can happen within a month in highly nutrient enriched streams, and this sloughing can significantly reduce measured algae accrual (Pringle, 1987; Stelzer & Lamberti, 2001). Thus, the one month exposure time should be a good compromise for measuring algae accrual.

Many studies use chlorophyll-a as a measure of algae assay for algae biomass; however in this study, AFDM was used since chlorophyll-a and biomass are not necessarily correlated, especially when comparing across sites with significantly varying species compositions, light, and temperature (Baulch, Turner, Findlay, Vinebrooke, & Donahue, 2009). Between different current regimes there can be as much as 30 to 40 times more biomass in slower streams and significantly different species compositions and successional trajectories and physical structure (Poff, Voelz, & Ward, 1990). Concentration of chlorophyll is highly variable across different habitat conditions much more than dry mass and ash-free dry mass (Morin & Cattaneo, 1992; Shilling & Davis, 2005)

There is some criticism that the methods used to exclude grazers are not entirely effective, however some reduction in grazing pressure is often necessary to see an effect of nutrient or light levels on algae biomass. In this study, Vaseline and elevation of the tiles was used to prevent grazers from accessing the tiles, in order to be able to determine primary production and calculate grazing pressure. It has been criticized that neither Vaseline nor raising the tiles effectively excludes anything other than crawling grazers (Feminella, Power, & Resh, 1989; Kuhara, Nakano, & Miyasaka, 2000; McAuliffe, 1984). Macroinvertebrates were observed in this study in high numbers on some of the Vaseline and raised treatment. Grazing pressure can mask any effects of nutrient or light additions on algae biomass, but partially reducing grazing has shown significant increases in algae biomass accrual under nutrient enriched conditions (Elwood, Newbold, Trimble, & Stark, 1981; Feminella et al., 1989; Hawkins & Furnish, 1987; Stewart, 1987)

Of the two methods used for excluding grazers (Vaseline and raising the tiles), Vaseline should, in theory, most closely maintain natural conditions found on untreated tiles. Vaseline has the issue that it might become compromised by debris sticking to the Vaseline and forming a bridge. Scouring of the stream bed by tumbling rocks during flooding significantly alters algae species communities with endolithic species being selected for over filamentous ones and has the ability to reduce algae by more than threefold (Power & Stewart, 1987). It was observed that the raised tiles were not subjected to this same level of scouring; the tiles placed on the ground were often found with stones on them or completely buried, but the raised tiles were always completely free. Also, in this study, there was no correlation between algae mass accumulations on raised tiles and Vaseline tiles, further supporting the possibility of differing environmental conditions. Thus, the use of Vaseline for measuring grazing pressure and primary production is favored over raising the tiles. No reasons were found to not use Vaseline, even if the value for primary production is not absolute, given that some of the production is scoured off and grazed.

Discussing the effectiveness of excluding grazers and scouring during flooding for measuring primary production, begs the question, if one can even measure primary production through grazer

exclusion. Generally there is higher primary productivity with increased grazing pressure and lower autotrophic biomass up to a certain point (Cooper, 1973; Lamberti & Resh, 1983; Marker, 1976; McNaughton, 1976). However, the exact effect that grazers have on the algae can be highly varied where in some instances there is an increase in chlorophyll-a concentration but no change in biomass, while in other instances there is reduced biomass as high as 30% (Kehde & Wilhm, 1972; McIntire, 1973). The fact that primary production arguably cannot be measured simply by removing grazers, and that grazers cannot be entirely excluded with noninvasive means, simply brings up the point that the values for grazing pressure and primary production are not absolute values, but still may be very valuable in assessing ecosystem integrity.

Algae response to pollution levels

There were no significant differences between pollution levels regarding algae biomass accrual, primary production or grazing pressure despite countless publications citing a link between the algae and nutrients (Dodds et al., 2002; Elwood et al., 1981; Feminella et al., 1989; Hawkins & Furnish, 1987; Stockner & Shortreed, 1976, 1978). Dodds et al. (2002) when looking at all of North America found a breaking point of 30 $\mu\text{g P/l}$ and 40 $\mu\text{g N/l}$, above which there was greater algae growth, and these breaking points coincidentally line up with the separations between the pollution levels used in this study. They also found that as much as 40% of the biomass variation across the continent was attributed to P and N levels. However, Wuhrmann and Eichenberger (1975) found in their study, no increase in algae biomass with added macronutrients of P and N, but they did with added micronutrients, indicating that micronutrients can also be important limiting nutrients, even if they aren't given as much attention. The lack of correlation seen between algae and nutrients could be a sign that there are other factors masking the effect of macronutrients, such as micronutrients, light levels, sediment or other habitat characteristics.

Primary factors governing algae growth and grazing

When looking at all the measured environmental variables, the sole significant driving factor behind algae biomass accrual and primary productivity was percent canopy cover. Light levels had such a dominant effect on the algae that it masked the effects from all the other possible influential factors such as nutrients or habitat. Where nutrients were not limiting, a positive relationship between light levels and algae biomass has been observed in other studies (Stockner & Shortreed, 1976). These previous studies and the fact that even under open canopy conditions, there was no observable pattern between the pollution levels suggests that nutrients are not the limiting factor for algae in these streams.

Given the apparent importance of canopy cover, the open early successional nature of the restored sites explains their increased algae accrual and primary production. The difference between

the restored and reference sites was statistically insignificant; nevertheless, noticeably greater levels of algae accrual and primary production were seen in the restored sites. This can be explained very well by the fact that the restored sites were disproportionately situated in open, early-successional habitats as opposed to the reference sites. All the restored sites had canopy coverages of less than 15% with only two of twelve reference sites being less than 15%. Seeing that restored stretches are newly constructed streams, it should be a given that they are open, early-successional, and thus it would make sense that they exhibit greater propensity for algae accrual and primary production given the better lighting conditions.

This importance of canopy cover was not seen in the grazing intensity measurements, instead it was found that stream width for clean sites was the major driving factor behind grazing, possibly explained by grazer species assemblages. In Scandinavian streams, species richness has been found to strongly correlate with stream order, width and slope which are all intercorrelated (Brönmark, Herrmann, Malmqvist, Otto, & Sjöström, 1984; Malmqvist & Eriksson, 1995; Malmqvist & Mäki, 1994; Wiberg-Larsen, Brodersen, Birkholm, Grøn, & Skriver, 2000). This is in accordance with the theory that with more area, you will have increased species diversity due to increased habitat diversity from greater habitat size (Rosenzweig, 1995). At lower levels of diversity, increasing the species count should result in greater functional efficiency due to niche complementing, until the species become ecologically redundant (Setälä & McLean, 2004). At higher pollution levels, other factors may be influencing species assemblages such as reduced nutrient limitations, hypoxia and toxicity (Gammeter & Frutiger, 1990; Maltby, 1995; Moreno & Callisto, 2006).

Decomposition

Analysis of methods:

There was a relatively large range of temperatures experienced in the streams, and therefore the decomposition rates needed to be adjusted with degree days. The average daily temperature during the course of the experiment ranged from 1.52°C to 7.95°C. Decomposition has been found to increase with increased temperature due to greater fungal efficiency (Fernandes, Seena, Pascoal, & Cássio, 2014; Reice, 1974). Adjusting for temperature with degree days has been shown to work (Ruffo & Bollero, 2003).

The purpose of using natural leaves as a decomposition substrate is to mimic natural conditions; however, air drying a single species of leaf and placing them in litter bags, severely compromises them structurally and results in debatably unnatural conditions. Estimating dry-weight as opposed to drying the leaves beforehand, increases variability of measurements in an already highly variable substrate and is thus not ideal. However, drying the leaves before exposure results in

rapid leaching of soluble compounds which would naturally not happen, facilitating significantly faster colonization and decomposition of the leaves (Bärlocher, 1997; Boulton & Boon, 1991; Gessner, Chauvet, & Dobson, 1999). There also may be a shift in the type of colonization of the leaves, bacterial or fungal (Bärlocher, 1997). Many riparian and aquatic macrophytes don't abscise their leaves so drying them is totally unnatural (Bärlocher, 1997). Nevertheless, some leaves do dry on the stem or on the ground before entering the water, so this issue is not clear cut (Gessner et al., 1999). In this study *Alnus glutinosa* leaves were used, which could very well dry either in the branches or on the ground, prior to blowing into the stream. Another issue is that there is a wide variety of aquatic macrophytes, riparian vegetation and tree species that contribute to the litter load, but only one species, that may not even be present in that particular stretch is used (Bärlocher, 1997). This was definitely the case in this study with *Alnus glutinosa* generally only being found in restored stretches yet used as the decomposition medium at all the sites. Also, the act of placing the leaves in bags alters the physical forces that would be felt on natural leaves (Bärlocher, 1997; Boulton & Boon, 1991). Nevertheless, leaves are a widely used standard for measuring decomposition, and the same leaves need to be used at each location to make decomposition rates comparable between sites.

Leaves suffer from issues with high levels of variability, which may be alleviated by using pure cellulose mediums. Leaves from one tree species, but from different individuals has been shown to have significantly different decay rates either due to genotype or other factors such as browsing (Irons III, Bryant, & Oswood, 1991; LeRoy, Whitham, Keim, & Marks, 2006). There are even significant differences in decay seen in leaves from the same tree, but different shad levels in the canopy (Sariyildiz & Anderson, 2003). The Wettex cellulose sponge cloth would provide a much more consistent substrate for comparing decomposition rates.

Pure cellulose mediums have been shown to function well in microbial decay assays. There are no nutrients in the pure cellulose strips, so it is more ideal for looking at nutrient differences than leaves, which may mask small effects with their own nutrients (Newman, Kumpf, Laing, & Kennedy, 2001). However, pure cellulose lacks many of the compounds and structures of natural leaves so they will decompose fundamentally differently (Howard, 1988; Jenkins & Suberkropp, 1995). Nevertheless, it has been shown that aquatic fungi and bacteria can digest pure cellulose (Rabinovich, Melnik, & Bolobova, 2002; N. Singh, 1982). Shredder palatability of the pure cellulose has probably significantly to do with conditioning by fungi and bacteria (Graça, 2001). Unfortunately, the decay of pure cellulose cotton happens very fast so conditioning may not have time to happen properly (Gestel, Kruidenier, & Berg, 2003; Tiegs, Langhans, Tockner, & Gessner, 2007). In addition, shredders will selectively feed on different leaf species depending on factors such as toxicity, toughness, nutrients, so assuming that they will utilize cellulose sponge cloth the same as leaves is

unreasonable (Graça, 2001). Gestel et al. (2003) found that soil invertebrates did not correlate with cellulose decomposition, but did with other tested mediums. Thus, the Wettex cellulose sponge cloth decay should not be compared directly to the leaf litter decay, but could serve well for determining microbial decomposition.

The Wettex cellulose sponge cloth seemed to suffer from high rates of mechanical loss in the large mesh bags, rendering the utility of the calculated invertebrate mediated decomposition very poor. When taking all measured variables into account, there was increased invertebrate mediated decomposition with increased pollution level for Wettex, but for the leaves, the inverse was seen with invertebrate mediated decomposition having the lowest rate in the highest pollution level. Invertebrate mediated decomposition is a function of microbial growth, mechanical loss and grazing. Elwood et al. (1981) found that with leaves, increased respiration rates accounted for only 10 to 34% of the increased mass loss from nutrient enrichment treatments, suggesting increased mechanical breakdown of the leaves at higher nutrient levels. Cellulose cotton strips lost 95% of their tensile strength with only 30% mass loss, and in general they decomposed much faster than natural leaves (Tiegs et al., 2007). So, from the fact that leaves already lose most of their mass through mechanical means and pure cellulose breaks down much faster and loses a very significant amount of tensile strength very quickly, and that there was near entire mass losses in some of my sites, it can only be assumed that the invertebrate decomposition of the Wettex is severely confounded by mechanical loss of the severely softened cellulose sponges at higher decomposition rates. Disregarding any grazing preferences, calculated invertebrate mediated decomposition of the Wettex cellulose cloths should not be compared to that of the leaves. It is more an indication of microbial activity, resulting in increased mechanical loss of the Wettex, and not that of invertebrate shredding activities.

Decomposition's response to water pollution

Invertebrate activity was not a significant contributor to decay as seen by the identical models for total and microbial decay in the leaf and Wettex treatments. This pattern has been seen in many other studies (Benfield, Jones, & Patterson, 1977; Matews & Kowalczewski, 1969; Meyer, 1980; Reice, 1977).

When looking at total and microbial decay, Wettex cellulose sponge cloth showed increasing rates of decomposition with increasing levels of pollution. No pattern was discernable from the leaf litter treatments, which may be due to too high levels of variability between the leaves (Irons III et al., 1991; LeRoy et al., 2006; Sariyildiz & Anderson, 2003). The increased decomposition with increased nutrient loading has been observed in many studies (Gulis, Rosemond, Suberkropp, Weyers, & Benstead, 2004; Gulis & Suberkropp, 2003; Stelzer, Heffernan, & Likens, 2003; Suberkropp, Gulis, Rosemond, & Benstead, 2010).

When taking all the other variables into account, invertebrate mediated decomposition for the leaf litter was least in the highest pollution grouping. At different pollution levels, different invertebrate assemblages are observed in other studies and are to be expected due to reduced nutrient limitations, hypoxia and toxicity (Gammeter & Frutiger, 1990; Maltby, 1995; Moreno & Callisto, 2006; Woodcock & Huryn, 2005). Invertebrate densities are often greatest with mid-level nitrification (Elwood et al., 1981). In the literature there is often no decline in decomposition rates of microbes at high levels of nitrification (Elwood et al., 1981; Imberger, Walsh, & Grace, 2008), so this result from my study could very well be a result of invertebrate shredding on the leaf litter and not microbial, as was seen in the Wettex. Alone, pollution level was not a significant variable for leaf litter decomposition, since there were other more influential factors at play.

Primary factors governing decay rates

Vegetation community was the primary significant factor for all the forms of decomposition except invertebrate mediated decomposition of Wettex. Globally plant litter has proven to be a more important factor on decomposition rates than climate (Cornwell et al., 2008). Many studies have found increases in decomposition with increased litter quality from different vegetation types, however with very inconsistent explanatory mechanisms where fungi and invertebrates may or may not have increased in abundance and/or exhibited community changes (Bärlocher & Graça, 2002; Benfield et al., 1977; Benfield, Webster, Tank, & Hutchens, 2001; Cornwell et al., 2008; Kominoski, Marczak, & Richardson, 2011; Kreutzweiser, Good, Capell, & Holmes, 2008; Laitung & Chauvet, 2005; Lecerf, Dobson, Dang, & Chauvet, 2005; LeRoy & Marks, 2006; Mckie & Malmqvist, 2009; Stone & Wallace, 1998; Whiles & Wallace, 1997). Diverse leaf litter has the potential to increase microbe and invertebrate diversity (Bärlocher & Graça, 2002; Laitung & Chauvet, 2005; Lecerf et al., 2005; Stone & Wallace, 1998; Whiles & Wallace, 1997). However, it was frequently found that diversity did not correlate with decomposition levels, hinting to the importance of individual species (Bärlocher & Graça, 2002; Kominoski et al., 2011; Lecerf et al., 2005; Mckie & Malmqvist, 2009). One consistency in the literature was that litter quality and quantity was the most important variable in increasing litter decay rates (Benfield et al., 2001; Kominoski et al., 2011; Lecerf et al., 2005; LeRoy & Marks, 2006; Mckie & Malmqvist, 2009; Stone & Wallace, 1998; Whiles & Wallace, 1997). This pattern can be seen in this study where deciduous forests generally had the greatest rates of decomposition and meadows and meadow-early-successional-scrub the least. However, there were very few sample points in this study ($n = 18$) relative to the number of variables (5 levels for vegetation community and 3 levels for pollution), so these models suffered from overfitting with very little sampling overlap between the levels of the different factors. The model building was already done with strict criteria so further simplification would have been very dubious. However, despite

overfitting issues, there is confidence in the results given that the general pattern seen in this study coincides with that found in the literature; nevertheless, one should be critical towards the precise relationships between vegetation groupings.

For the invertebrate mediated decomposition of Wettex cellulose sponge cloths, vegetation community was not significant, only pollution level and habitat variability. As previously discussed, this measure is most likely a function of microbial decay and not invertebrates and microbial decay is very heavily influenced by nutrient loadings. However, when looking at just one pollution level at a time, the pattern of increased decomposition with increased habitat variability was very apparent (see Fig. 22). Studies have found breakdown rates to be highest on substrates with the greatest spatial heterogeneity and this couldn't be linked to invertebrate diversity and abundance or water velocity which suggests the importance of diversity in microbes (Reice, 1974, 1977).

Substrate mode was found to be a statistically significant factor for total and microbial decomposition rates for the Wettex cellulose sponge cloth, however this may be an artifact. It was found that decomposition decreased with an increase in stone size. This entirely contradicts the literature, which generally states that decomposition is greatest in larger stones where heterogeneity is greatest (Mackay & Kalf, 1969; Meyer, 1980; Reice, 1974, 1977). This discrepancy may well also be an artifact of an overfitted model. As seen in Figure 20, there was only a decrease in decomposition with larger stones for the mixed forest vegetation type, but this decrease in pollution was also accompanied by a decrease in pollution level; so, whether this decrease was due to substrate size or pollution level cannot be discerned. Furthermore, within the other vegetation communities, there was no correlation between substrate size and decomposition rate, further drawing the relationship into question.

Stream width as well as percent culverted one kilometer upstream were significant factors for microbial decomposition, possibly as a result of increased habitat area. Stream width was significant for the Wettex cellulose sponge cloth treatments and culverts were significant for the leaf litter. Faster break down rates have been observed in other studies in larger streams (Benfield & Webster, 1985; Triska, Sedell, & Buckley, 1975). In theory, with increased area you should have increased diversity (Rosenzweig, 1995), and species diversity has been shown to be strongly correlated with stream size and length (Brönmark et al., 1984; Malmqvist & Eriksson, 1995; Malmqvist & Mäki, 1994; Wiberg-Larsen et al., 2000). Functional efficiency of fungus increases at lower levels of diversity with an increase in diversity, probably due to niche complementing and increased resilience to disturbance, but after it plateaus signaling redundancy in the system (Setälä & McLean, 2004). Stream width as a surrogate for total upstream length and culverts as significant barriers and reduction of habitat size, should indicate very well the habitat area at each site.

Effectiveness of restoration of culverted stretches

Generally, it was found that ecosystem functions responded the same to the measured environmental variables in restored sites as in reference sites. The apparent increased algae accrual and primary production seen in restored sites (see Fig.11) can be fully explained by decreased canopy cover in the earlier successional habitats. When stratifying sites by pollution level, there was no statistical difference between restored and reference sites regarding decomposition, however it is graphically visible in the boxplots (see Fig. 14) that restored sites had lower decomposition of leaf litter than the reference sites. The issue is that the leaf decomposition for the reference and the restored sites were measured in different years, so it is impossible to conclude if these observed differences are due to variations between years or an effect of restoration. The Wettex cellulose sponge cloth decay for all sites and leaf litter decay for the restored sites were sampled in the same year. It is quite likely that the difference seen in leaf litter decay between the restored and reference sites is due to variations between years, given that there were nearly no differences observed in decomposition of Wettex cellulose sponge cloth between reference and restored sites (see Fig. 15), and the same general pattern of clean sites having less decomposition is seen in both Wettex cellulose sponge and leaf litter from the same year. However, even if there were a difference in leaf litter decomposition between restored and reference sites, it would have been impossible to decouple the effectiveness of restoration from vegetation community given that all the restored sites were found in early successional habitats with only one reference site in the meadow-early successional scrub category, and vegetation community came up as very significant at determining decomposition rates. Lower decomposition rates in habitats with less quantity and lower quality leaf inputs from the riparian vegetation was observed in other studies as well (Benfield et al., 2001; Kominoski et al., 2011; Lecerf et al., 2005; LeRoy & Marks, 2006; Mckie & Malmqvist, 2009; Stone & Wallace, 1998; Whiles & Wallace, 1997).

Conclusion

This study concluded that the ecosystem functions in previously culverted, restored stream reaches were very comparable to never before culverted, reference reaches. Thus, restoration of culverted streams in Oslo has been successful, at least with regards to the ecosystem functions of algae production and litter decay. Any observable differences in ecosystem functioning between the restored and the reference reaches, can be attributed to the early successional, open nature of the newly constructed streams, markedly the increase in primary production due to more open tree canopies. There was arguably no difference in decomposition rates between the restored and reference sites, but even a possibly lower decomposition rate in the restored sites could be explained by lower high-quality leaf inputs from riparian vegetation.

The question for the future is, given the constraints that cityscapes places on urban rivers specially, do the deculverted streams have the potential to develop along the successional gradient to mature forests, and is this even desirable? Through the course of succession, there are tradeoffs in ecosystem functions; greater litter imputes increase decomposition potential, but also decrease primary production through shading. In addition, ecosystem function is not the sole management objective, and other considerations need to be taken into account such as recreational value, water cleansing, maintenance costs, health and safety. However, it can be said from this study that restored culverted streams are neither better nor worse than their natural counterparts, at least regarding ecosystem functioning.

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Appendices

Appendix 1: Summary Statistics Table by Sampled Stream Reach

Table A2.1: Summary table of all measured variables by sampled stream reach

Site	Pollution Level	Ref/Res	Ecosystem Function Variables																											
			Water Chemistry						Algae Growth						Litter Decay factor k															
			low water BOD5 mg/l	medium water BOD5 mg/l	high water BOD5 mg/l	NH4 µg N/l	PO4 µg P/l	Exposure days	Alg Degree Days deg.	Alg Avg Temp deg.	Alg Growth AFDM µg/DD	sd	Grazing on Alg AFDM µg/DD	Alg Prim Prod AFDM µg/DD	sd	Exposure days	Wettex Degree Days deg.	Wettex Avg Temp deg.	Ktotal Wettex	sd	Kmicrob Wettex	sd	Kinvert Wettex	Leaf litter Degree Days deg.	Leaf litter Avg Temp deg.	Ktotal Leaf litter	sd	Kmicrob Leaf litter	sd	Kinvert Leaf litter
ALNresGAN	clean	restored	0.60	1.94	0.95	153	2	29	379.90	13.10	99	108	123	202	119	28	61.80	2.21	0.00023	0.00047	-0.00006	0.00032	0.00029	61.80	2.21	0.00285	0.00089	0.00184	0.00062	0.00089

Site	Pollution Level	Ref/Res	Riverbed																								
			Channel Dimensions						Percent Wentworth Size Classes								Sediment Statistics in Phi (φ)						Movement and Accumulations				Flow
			Reach Length m	Max Depth Shallowest Cross section cm	Max Depth Deepest Cross section cm	Depth Range cm	Depth Variability 4 pt. scale	Width m	Width Variability 4 pt. scale	Boulder %	Cobble %	Pebble %	Granule %	Sand %	SiltClay %	Xyal %	CPOM %	FPOM %	Sed Mode φ	Sed Sorting φ	Sed Skew φ	Sed Movement	Sed Load Alterations	Important Woody Debris	Bedload accumulation	Flow Character	
ALNresGAN	clean	restored	40	15	53	38	4	4.1	4	17	35	41	7	0	0	0	0	Pebble	-4	2.2	-0.13			yes		turbulent	

Site	Pollution Level	Ref/Res	Banks												Upstream and Watershed							
			Bank Dimensions and Composition				Bank Alterations				Bank Vegetation				1 km stretch upstream					Watershed		
			Gravel Banks	Sand Banks	Silt Banks	Bank Gradient deg.	Bank Height cm	Bank Protection %	Height Bank Protection cm	Dominant Type Bank Protection	Veg Bank Cover %	Canopy Cover %	Width of Riparian Forest	Riparian Forest Bank Cover %	Vegetation Community	Embanked % of length	Embanked 2 Sided % of length	Culverted % of length	Bridge % of length	Weirs num	Dams % of length	% of Watershed Developed
ALNresGAN	clean	restored	1	0	0	20	50	5	20	StonePitchingTight	95	3	isolated/absent	3.5	Meadow-Early successional riparian scrub	0	0	0	5	1	0	16

Appendix 2: Model Building

Upstream Effect on Pollution Levels

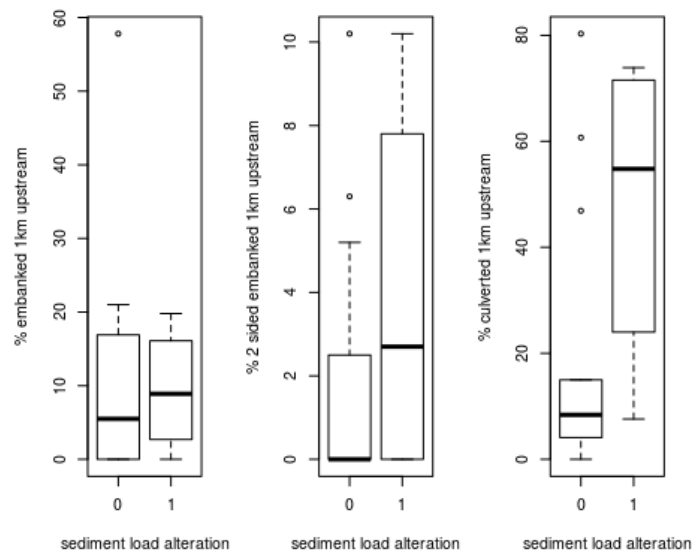


Figure A2.1: Boxplots of sediment load alteration vs stream modifications to 1 km stretch upstream: Streams with altered sediment loads had a marginally significantly greater percentage of culverts in the 1 km stretch upstream the sampled reach ($t = -1.7583$, $df = 4.2536$, $p\text{-value} = 0.1492$).

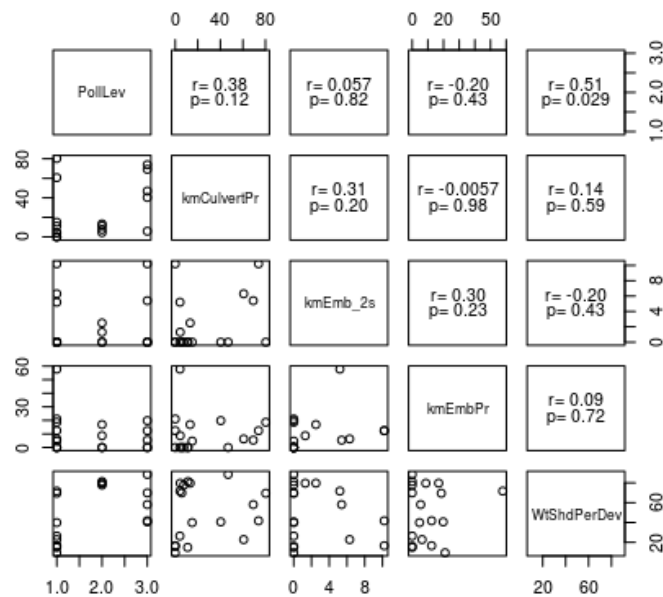


Figure A2.2: Paired matrix of Pearson correlation coefficients for pollution level and stream modifications to 1 km stretch upstream and the sites: Pollution level, percent culverted 1 km upstream, percent two sided embanked 1 km upstream, percent embanked 1 km upstream, percent of watershed developed.

The only correlation was between percent watershed developed and pollution level

Site-specific Environmental Variables' Effect on Ecosystem Functioning

Pearson correlation coefficient matrixes

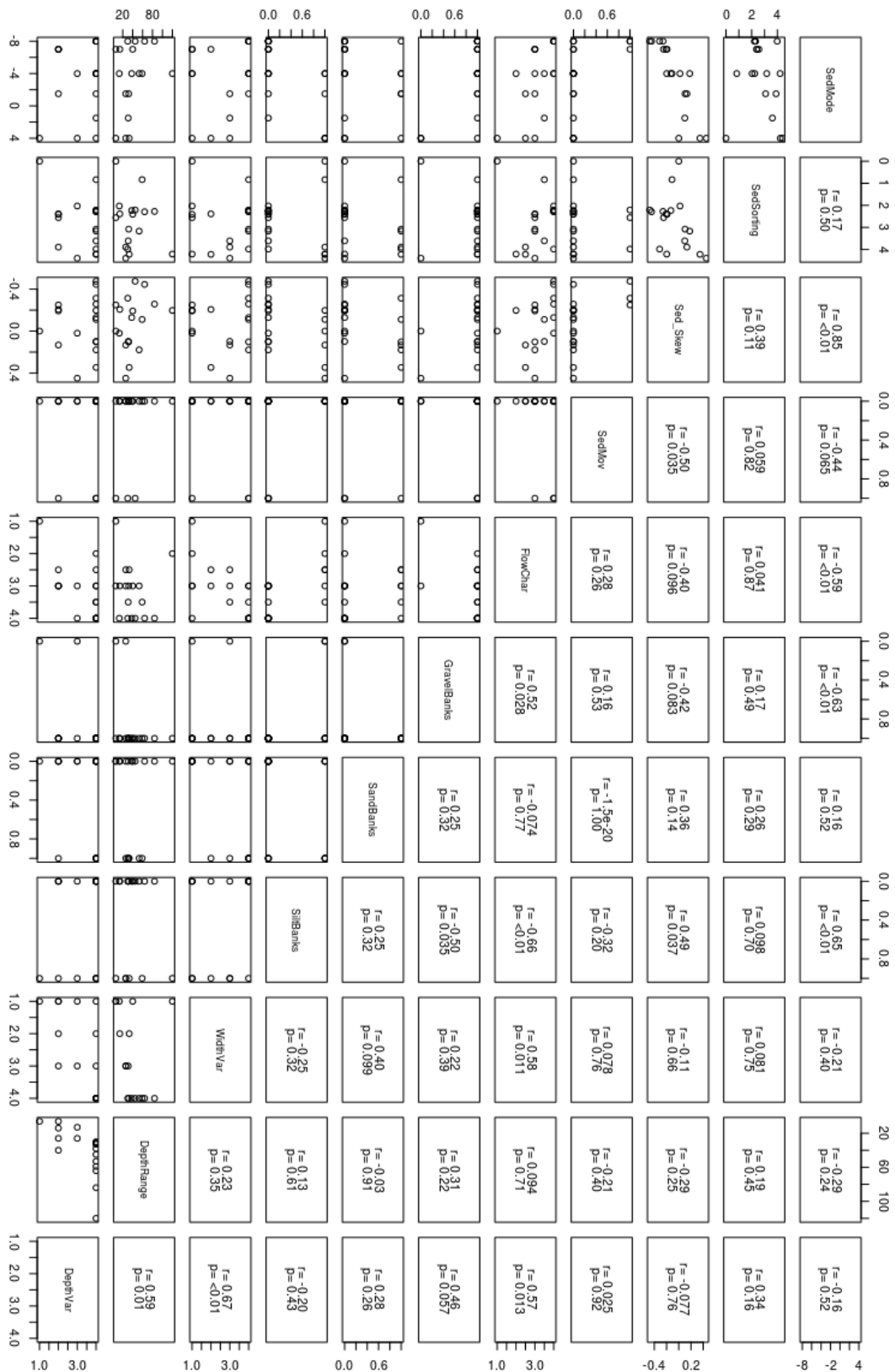


Figure A2.3: Pearson correlation coefficient matrix for stream dimensional and sediment variables: Sediment mode, sediment sorting, sediment skew, sediment movement, flow character, gravel banks, sand banks, silt banks, width variability, depth range, depth variability

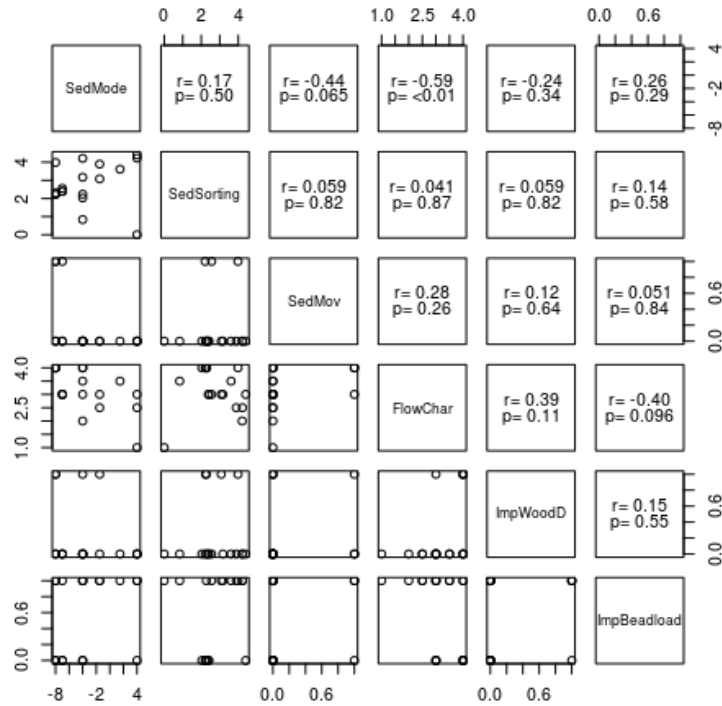


Figure A2.4: Person correlation coefficient matrix of sediment variables: Sediment mode, sediment sorting, sediment movement, flow character, important woody debris accumulation, important bedload accumulation.

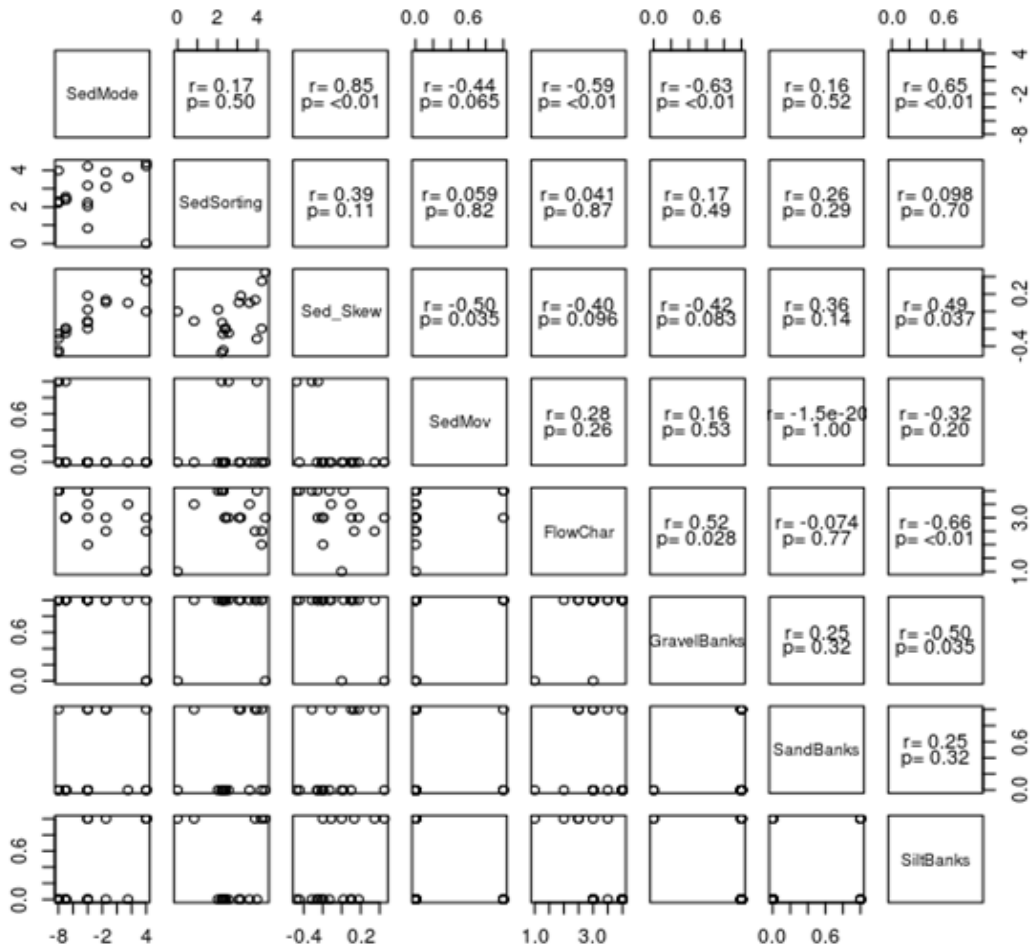


Figure A2.5: Person correlation coefficient matrix of sediment and accumulation variables: Sediment mode, sediment sorting, sediment skew, sediment movement, flow character, gravel banks, sand banks, silt banks.

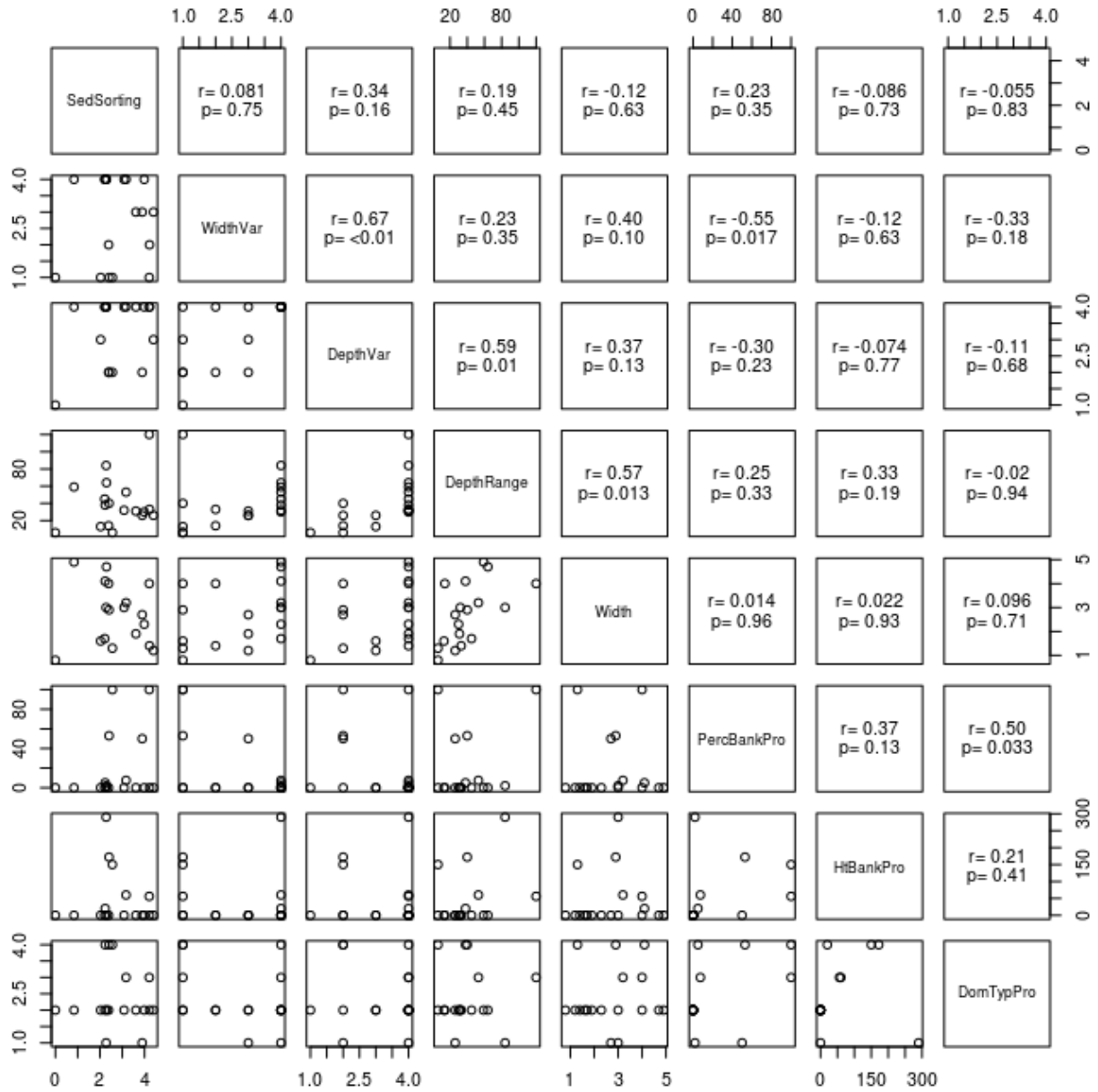


Figure A2.6: Persons correlation coefficient matrix of habitat variability and bank protection: Sediment sorting, width variability, depth variability, depth range, width, percent bank protection, height bank protection, dominant type of bank protection.

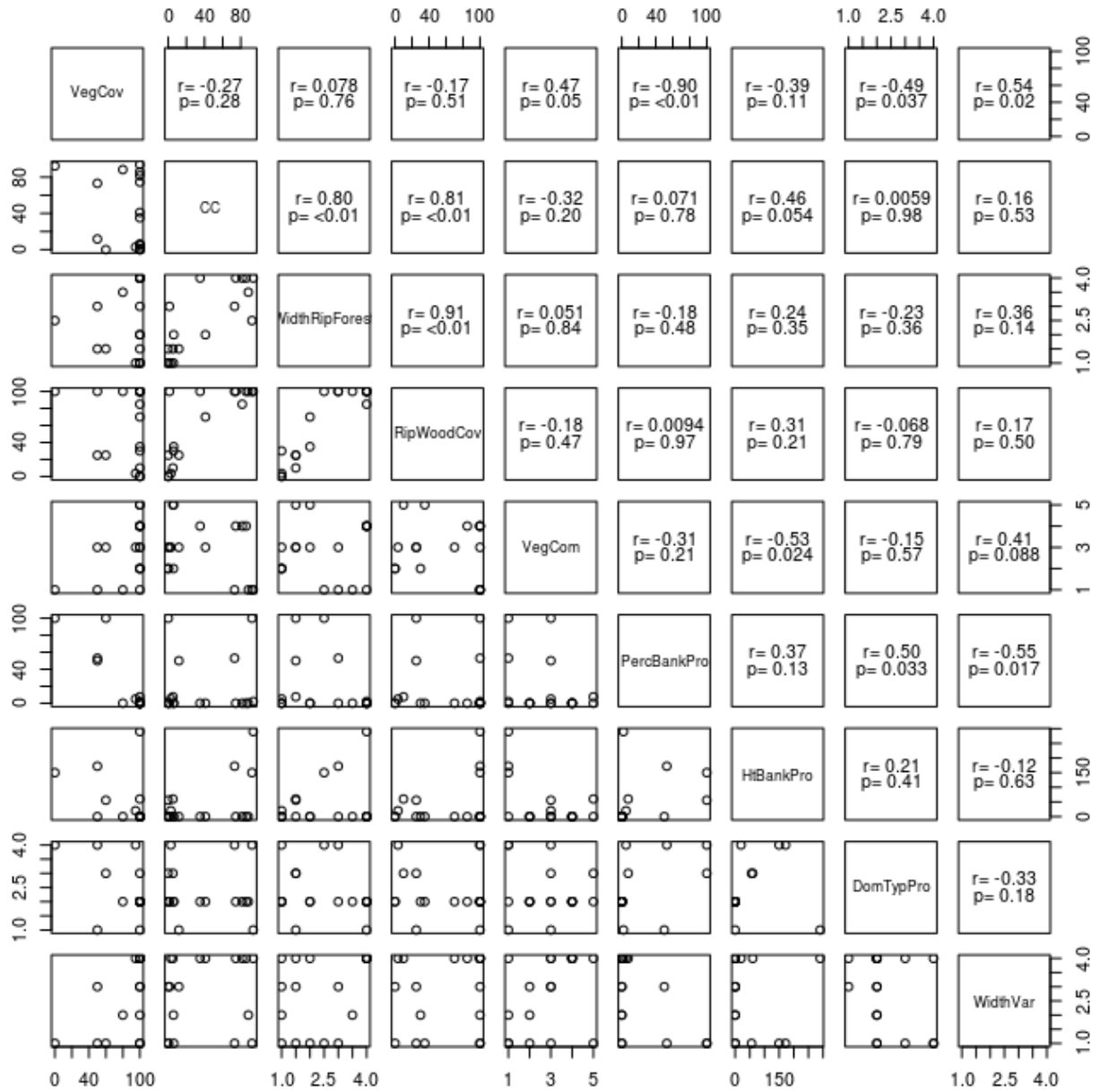


Figure A2.7: Persons correlation coefficient matrix of vegetation and bank alteration variables: Vegetation cover, canopy cover, width of riparian forest, riparian forest bank cover, vegetation community, percent bank protection, height bank protection, dominant type of bank protection, width variability



Figure A2.8: Persons correlation matrix of final variable set for modelling ecosystem functions: Pollution level, percent culverted 1 km upstream, percent embanked 1 km upstream, percent two sided embanked 1 km upstream, vegetation community, width of riparian forest, percent bank protection, height of bank protection, habitat variability (average of width variability and depth variability), sediment sorting, sediment mode, stream width.

Graphs of vegetation communities vs restoration and pollution levels

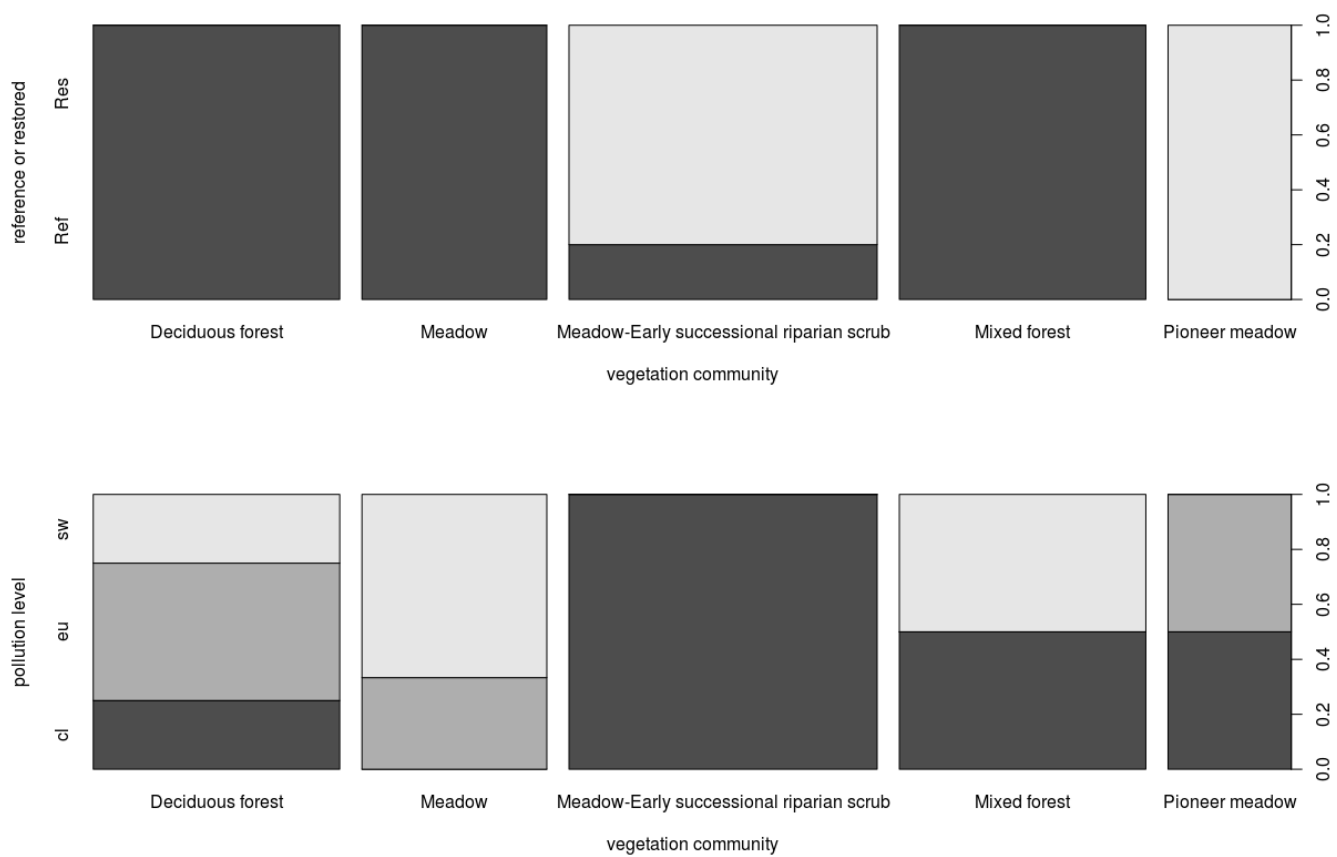


Figure A2.9: Vegetation communities vs reference/restored and pollution levels: X-axis shows the relative abundance of each vegetation community, y-axis shows the percentage of the respective pollution level or restoration status within each vegetation community.

Model parameters

Pollution level

pollution level ~ % watershed developed (32% developed is clean, 80% developed is eutrophic, 60% developed is raw sewage) Adj R² = 0.47

Algae

Algae biomass accrual + 1.1*minimum value ~ e^{^(6.12 - 0.04*% canopy cover)} R² = 0.70

Algae primary production + 1.1*minimum value ~ e^{^(6.11 - 0.03*% canopy cover)} R² = 0.60

Algae grazing intensity + 1.1*minimum value ~ e^{^(0.78 + (1.07 if cl, -0.57 if eu, -0.08 if sw)*stream width (m) + 5.05 if eu +2.86 if sw)} Adj R² = 0.67

Wetex sponge cloth

k exponential decay factor for total decomposition + 1.1*minimum value ~ e^{^(-5.44 + 0.43*stream width (m) + 0.34*sediment mode (φ) + pollution level (0 if cl, 1.97 if eu, 2.25 if sw) + vegetation community (0 if deciduous forest, -1.12 if pioneer meadow, -2.72 if meadow riparian scrub, -3.33 if mixed forest, -4.57 if meadow))} Adj R² = 0.94

k exponential decay factor for microbial decomposition + 1.1*minimum value ~ e^{^(-6.45 + 0.13*sediment mode (φ) + pollution level (0 if cl, 2.03 if eu, 2.85 if sw) + vegetation community (0 if deciduous forest, -0.22 if pioneer meadow, -1.42 if meadow riparian scrub, -2.61 if mixed forest, -3.18 if meadow))} Adj R² = 0.90

k exponential decay factor for invertebrate decomposition + 1.1*minimum value ~ e^{^(-7.54 + (-0.35 if cl, 0.29 if eu, 0.26 if sw)*habitat variability (4 pt. scale)} Adj R² = 0.73

Leaf litter

k exponential decay factor for total decomposition + 1.1*minimum value ~ e^{^(-4.87 - 0.01*% culverted 1 km upstream + vegetation community (0.21 if mixed forest, 0 if deciduous forest, -0.69 if pioneer meadow, -0.76 if meadow, -0.85 if meadow riparian scrub))} Adj R² = 0.62

k exponential decay factor for microbial decomposition + 1.1*minimum value ~ e^{^(-5.83 - 0.01*% culverted 1 km upstream + vegetation community (0.29 if mixed forest, 0 if deciduous forest, -0.20 if meadow, -0.33 if pioneer meadow, -0.48 if meadow riparian scrub))} Adj R² = 0.68

k exponential decay factor for invertebrate decomposition + 1.1*minimum value ~ e^{^(-6.50 + pollution level (1.10 if eu, 0 if cl, -1.64 if sw) + vegetation community (0.68 if mixed forest, 0 if deciduous forest, -0.94 if meadow riparian scrub, -0.96 if pioneer meadow, -3.16 if meadow))} Adj R² = 0.58

Appendix 3: Missing Temperature Calculations

Table A3.1: Calculations for missing water temperature data

Degree Days for Algae Growth Experiment				Degree Days for Leaf Litter Decay Experiment					
Site	Degree Days measured	n days measured	avg temp per day measured	calculated avg diff corection	t-tests for significance of correction factor and models	avg temp days missing	n days missing	Estimated degree days missing	Total Degree Days
ALN1	178.08	17	10.475	CotHOF3*0.8052 = 1.170	SlopeALN1 = SlopeHOF3*0.8052, StError = 0.1029, t = 7.823, df = 15, p-value = 1.13e-06 ***	9.306	12	111.67 (1.94378+(FRO3DD/29)*0.8269)*29	289.75 = 288.35
ALN3		0			avgALN3 = 1.94378 + avgFRO3*0.82693, R ² =0.7958		29		
ALN4	235.15	19	12.376	1.961	t = 6.615, df = 39, p-value = 7.28e-08 ***	10.415	10	104.15	339.30
ALNresGAN	294.36	22	13.380	1.160	t = 3.627, df = 42, p-value = 0.000771 ***	12.221	7	85.54	379.90
ALNresHOL	308.94	22	14.043	2.136	t = 6.39, df = 42, 1.09e-07 ***	11.906	7	83.34	392.28
FRO1	231.59	17	13.623	0.655	t = 1.745, df = 39, p-value = 0.0889 .	12.968	12	155.62	387.21
FRO3	168.81	17	9.930	0.619	t = 4.046, df = 39, p-value = 0.000239 ***	9.311	12	111.73	280.54
FROresBES	215.82	19	11.359	1.367	t = 6.991, df = 39, p-value = 2.2e-08 ***	9.992	10	99.92	315.74
HOF1	173.48	17	10.205	1.127	t = 5.144, df = 39, p-value = 7.93e-06 ***	9.078	12	108.93	282.41
HOF2	158.88	17	9.346	1.673	t = 9.808, df = 39, p-value = 4.42e-12 ***	7.673	12	92.08	250.96
HOF3	186.48	17	10.969	1.453	t = 6.908, df = 39, p-value = 2.86e-08 ***	9.517	12	114.20	300.68
HOV1	142.61	16	8.913				0	0.00	142.61
HOV3	235.17	17	13.834	1.297	t = 3.366, df = 39, p-value = 0.00173 **	12.537	12	150.44	385.61
HOVresBJE	211.30	22	9.605	0.215	t = 1.214, df = 42, p-value = 0.231	9.390	7	65.73	277.03
HOVresENS	269.60	18	14.978				0	0.00	269.60
HOVresTEG	257.61	22	11.710	0.553	t = 1.80 df = 42, p-value = 0.0791 .	11.157	7	78.10	335.71
MARI	186.93	17	10.996	1.260	t = 6.584 df = 39, p-value = 8.03e-08 ***	9.736	12	116.83	303.76
OST1	211.74	19	11.144	0.788	t = 4.254 df = 39, p-value = 0.000127 ***	10.357	10	103.57	315.31
= DD measured / n days measured = difference between avg temp of second half of experiment and the avg temp measured * avg temp missing									
temp an equal interval right after the experiment - correction									
Site	Degree Days	Models							
ALN1	127.16								
ALN3	162.98	avgALN3 = 0.81892*avgALN1+2.10191, R ² =0.906							
ALN4	209.21								
ALNresGAN	61.80								
ALNresHOL	55.84								
FRO1	48.23								
FRO3	235.51								
FROresBES	90.89								
HOF1	66.97	avgHOF1 = 0.98193*avgFRO1+0.70054, R ² =0.9579							
HOF2	47.72								
HOF3	99.71								
HOV1	124.05								
HOV3	150.93								
HOVresBJE	133.36								
HOVresENS	85.65								
HOVresTEG	127.42								
MARI	80.68	avgMARI = 1.0466*avgHOF3-0.8455, R ² =0.9473							
OST1	151.61								



Figure A3.1: Pearson correlation matrix of water temperature between sites for second half of algae experiment



Figure A3.2: Pearson correlation matrix of water temperature between sites for 14 days after the algae experiment



Figure A3.3: Pearson correlation matrix of water temperature between reference sites for the 2017 litter decay experiment.

Appendix 4: Study Site Pictures

ALNI



ALN3



ALN4



FRO1



FRO3



HOF1



HOF2



HOF3



HOVI



HOV3



MARI



OSTI







FRoresBES





HOVresENS



HOVresTEG

