# Benthic Macroinvertebrate Water Quality Metrics and Landscape Variables in the San Francisco Bay Area

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# ABSTRACT

Expanding geospatial databases offer new opportunities for correlating landscape characteristics (LCs) with water quality metrics, thereby improving stream management. CEDEN (California Environmental Data Exchange Network) is an online data portal with many public databases, including benthic macroinvertebrate (BMI) data. This study used four years of SWAMP BMI data (Surface Water Ambient Monitoring Program) alongside National Land Cover Data to examine correlations between BMI metrics and LCs within the San Francisco Bay Area. LCs were sampled in ArcGIS using 100m, 500m, and 1000m point buffers, and catchment areas around BMI sampling locations. Catchment-scale landscape data were more normally distributed, but 500m and 1000m buffers returned the strongest correlations. Linear regression revealed land cover type, % canopy cover, and slope as most predictive of BMI metrics, while road-based metrics, % impervious surface coverage, and population data were not correlated for this dataset, likely due to selection biases during BMI site selection.

# **KEYWORDS**

National Land Cover Dataset (NLCD), geospatial information systems (GIS), benthic macroinvertebrate, landscape characteristic, urbanization, ArcGIS, zonal statistics, Modifiable Areal Unit Problem (MAUP)

### **INTRODUCTION**

Urbanization induces change in habitat, biota and physiological stressors by physically modifying the natural and built environments (1). These changes can be extremely harmful to biotic assemblages, and alterations to urban landscape factors have been extensively linked to deteriorating stream ecological function (2 - 4). Urbanization inflicts "urban stream syndrome," wherein habitat quality is negatively impacted by elevated nutrient and contaminant levels, increased periods of water stress, and flash floods (3, 4). These changes are driven by a variety of stressors, including sedimentation, altered runoff patterns, altered geomorphology, and runoff borne contaminants (5). These stressors and landscape changes threaten biological communities essential for ecosystem functioning.

Bottom-dwelling aquatic insects, or benthic macroinvertebrates (BMI), are widely used as bioindicators of stream ecological functioning and water quality (6). BMI are ideal bioindicators, because they are well-studied, ubiquitous, and vary widely in size and ecological role (7). Variation in survivorship, size, and growth conveys information about potential stressors such as sedimentation, pollutants, or abnormal flow conditions (6). The orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) are three widely used bioindicators that effectively gauge water quality because of their sensitivity to pollutants (8). In the last several decades the use of benthic macroinvertebrates as indicators of stream quality in rapid bioassessment protocols has increased dramatically (6). Combining biotic data with landscape characteristics such as vegetation or population density can help estimate the impact of stressors on stream health (9, 10).

Many studies examine the effects of urbanization on ecosystem functioning (3), from studies testing for correlation between one landscape variable and multiple bioindicator effects (9) to studies testing multimetric urbanization indices urbanization against multiple bioindicator effects (11, 10, 12). These studies have found significant and meaningful correlations between bioindicator responses and landscape variables such as road and population density (12) and land cover type and use (13). However, there are relatively few published studies using existing public datasets regarding factors of urbanization to predict stream water quality (14, 13), and there are no published studies regarding this SWAMP dataset in the San Francisco Bay Area. Extant studies tend to use independently sample data rather than examining publically available data (15, 16, 17).

The primary objective of this study was to assess how well landscape characteristics can predict the relationship between stream health and ecological functioning in urban creeks in nine counties of the San Francisco, California Bay Area. I used landscape variables drawn from federal and state sources and BMI data from the CA State Water Resources Control Board (SWAMP). To that end I sampled mean values of landscape characteristic variables around each BMI sample location using buffer polygons and catchment basins. I then ran single and multiple linear regressions between these means and BMI health metrics.

# **METHODS**

My thesis project used preexisting federal data sources and benthic macroinvertebrate (BMI) data from the Surface Ambient Water Monitoring Program (SWAMP) to determine which urban landscape characteristic (LC) variables best predict benthic macroinvertebrate health in urban streams in the Bay Area. In ArcGIS I sampled data from LC layers surrounding each BMI sampling point using two different techniques, circular buffers and watershed areas. I used 5 BMI metrics (Table 1) and 8 landscape-level LC variables in this study (Table 2). The project had three overarching phases:

- I. **Data acquisition** consisted of locating, procuring, and processing LC data.
- II. **Data sampling** involved the extraction and averaging of data values from LC layers.
- III. **Data analysis** consisted of testing for normality and differences between treatment groups, and running linear and multiple linear regressions.

# I. Data Acquisition

I used 444 BMI sample events from 2001 - 2004 of the total 517 sample events from 2001 - 2008 in the SWAMP dataset. The sample sites include a wide variety of microclimates, stream orders, and environments. Due to time and manpower constraints, local environmental conditions were not considered. I used five of 132 BMI metrics in the SWAMP dataset (Table 1).

Except land cover, all layers were continuous data layers from publicly available governmental sources (Table 2). I derived elevation and slope from 10 meter resolution Digital Elevation Models (DEMs) available through the United States Geographical Survey (USGS)'s

National Map Viewer. USGS' National Land Cover Dataset (NLCD) provided LC data (Table 2). I obtained population density data from the US Census, and created both a road density raster and a distance to nearest roads raster from US Census TIGER road shapefiles. Data layers not already in Universal Transverse Mercator Zone 10 North projection, North American Datum 1983 were converted into it to avoid area distortion errors.

# **II.** Data Sampling

I used two sampling methods to obtain mean and median LC data values for areas surrounding each of the 444 BMI sampling points. First, I created circular fixed-distance buffers at multiple distances (100m, 500m, 1000m) to allow analysis of the extent of the modifiable areal unit problem (MAUP), which can skew results (18). Second, I used ArcGIS hydrology tools on 10m resolution DEMs to generate catchment basins for each sample point. Both buffers and catchment basins were converted into individual rasters and used as inputs for the zonal statistics tool in ArcGIS, which used them as "cookie cutters" to extract data from underlying LC data layers. The resulting statistics were collated into tables for analysis.

# **III.** Data Analysis

I ran boxplots, histograms, and basic statistical analysis tests in the statistics program R. I used a Shapiro-Wilks test and a D'Augustino-Pearson test to check normality. To test whether MAUP bias created any significant difference between sampling groups, I ran Friedman tests and Kruskal-Wallace tests, which are appropriate for non-normal data. To determine if any relationships existed between the LC and the BMI variables, I ran a set of linear regressions comparing each LC variable to all BMI variables. I used an ANOVA to determine the strength of variation between land cover type and BMI scores. I then created multi-variable linear regression models to determine which LC variables explained the most variation.

#### RESULTS

### Transect-introduced distortion in the BMI data

The landscape data did not significantly differ on the basis of whether BMI samples at a site were taken at one transect or at three transects (Table 5, Table 6). However, I removed single-transect sample events due to the differences in the medians between the two groups (Table 6). Of the total 517 samples, 444 (86%) were from sites with three transects, and 73 (14%) were from sites sampled at one transect.

#### Normality

The bulk of the sampled LC data were non-normal. The Shapiro-Wilks test found seven treatments were potentially data (Table 3), but of these only two appeared normal from visual heuristics and statistical summaries. The ten treatments with a W statistic between 0.8 and 0.9 (Table 3) appeared non-normal and left-skewed. A D'Agostino-Pearson normality test found all data non-normal with p values < 0.000001.

Overall, catchment level treatment sampled data had the most normality, with %CC, RDI, EV, and SL all possessing W statistics above 0.9 (Table 3). The circular buffer treatments of RDI and SL also had elements of normality, as indicated by a W statistic above 0.9 (Table 3). In addition, %CC, RDE, EV, and SL all had circular buffer treatments with W > 0.8 (Table 3), which appears non-normal in histograms.

### **Sampling Zone Selection Effects**

I found significant differences in scatter, skewness, and distribution between sampled mean values in watershed and in circular buffer treatments for all variables (Table 8). However, these trends only appeared in %CC, RDI, EV, LCO, and SL. RDE, IM, and PD did not change dramatically between circular buffer and watershed treatments.

In LCO, urban land cover types decreased in frequency and forest types increased in frequency as sampling size increased (Table 8). %CC, EV, and SL all switched from a right-tailed distribution under circular buffer sampling to a normal distribution under watershed sampling. RDE, %IM, and PD all retained a right-tailed distribution for all sampling treatments.

The distribution of %IM also changed drastically as sampling size increased. With circular buffer sampling methods, around 30% of the data (133 of 444 sites) possessed greater than 10% impervious surface coverage; yet, with the watershed sampling method only 5% of the data (22 of 444 sites) had greater than 10% impervious surface coverage.

For each landscape variable, a Kruskal-Wallis chi-squared test (Table 10) and a a Friedman rank-sum chi-squared test showed that every sampling technique generated data that were significantly different from one another (Table 9, Table 10).

The majority LCO type changed between each treatment, moving from a dataset with a more diverse mix of land cover types at smaller sampling sizes to a dataset possessing three main categories with small numbers of additional LCO types (Table 11).

### Sampled Landscape Characteristic Data Summary

Given the lack of normality in the circular buffer results, this sub-section *refers exclusively* to watershed-level sampled landscape data.

### Population Density

429 of 444 BMI sampling sites (97%) of BMI sampling sites were located in areas with a mean sampled population density of less than 2000 people per square mile. The mean population density of all sampled sites was  $451.6 \pm 1271.8 (\pm SD)$ , while the median value was 64.9 people per square mile (Table 8). There were no significant correlations between population density and other LC or BMI variables (Table 12).

#### % Impervious Cover

The %IM data was concentrated at the lower bound, with 95% of the data (421 of 444 sites) below 10% impervious surface coverage. The data's mean and median were 3.8% (± 6.8% standard deviation) and 0.49% (Table 8, Figure 2a). %IM had a weak correlation with %CC, where higher %IM values occurred at lower %CC values (Figure 3b,  $R^2 = 0.1379$ , *p* < 0.000001).

There were also higher %IM values at lower SL values (Figure 3f).

# % Canopy Cover

%CC had peaks between 0% and 10% and between 30% and 40% coverage, while the median was at 35% (Table 8). RDE, RDI, PD, and %IM all showed no relationship. %CC and EV showed a semi-linear relationship (Figure 3a,  $R^2 = 0.2922$ , p < 0.000001), while %CC and SL showed a stronger correlation (Figure 3c,  $R^2 = 0.4967$ , p < 0.000001).

# Land Cover

Forests (67%, 297 of 444 sites) and Grasslands (27%, 119 of 444 sites) made up the bulk of the data, with the rest of the data evenly distributed throughout all four categories of urban sites (5%, 22 of 444 sites) (Table 8, Figure 2g).

# Road Density and Road Distance

RDE was distributed mostly at the left bound, with high right-tail skewedness (Figure 2e). The only LC variable that showed correlation with RDE was RDI, which showed an inverse relationship (Figure 3e,  $R^2 = 0.3892$ , p < 0.000001). RDI was almost normally distributed (Figure 2b), although left skewed (Table 8).

# Elevation and Slope

Elevation and slope both appear normally distributed (Figure 2D and 2F). They correlated well with each other (Figure 3d,  $R^2 = 0.5441$ , p < 0.000001), while EV correlated slightly with %CC (Figure 3a,  $R^2 = 0.2922$ , p < 0.000001) and SL correlated slightly with %CC (Figure 3c,  $R^2 = 0.4967$ , p < 0.000001).

# **Relationships between Landscape and BMI variables**

### Bias in BMI Sampling Site Locations

The BMI sampling sites were uniform with respect to their RDI and RDE values, due to the selection of sampling sites within walking distance of roads. Low %IM and PD values show that the sites selected are also predominantly far from highly developed urban areas (Table 8).

# Linear Regressions and ANOVAs

Single-variable linear regression returned highly significant results ( $p < 2.2 * 10^{-16}$ ), yet generated models that fit the data poorly ( $R^2 < 0.6$  in nearly all cases). LCO, %CC, and SL best predicted BMI health metrics, with  $R^2$ 's averaging over 0.50, 0.40, and 0.40 (Table 10). RDE and %IM averaged  $R^2$  values above 0.30, while RDI, PD, and EV averaged  $R^2$  values below 0.20 (Table 12, appendix A).

Across all treatments, EPT Index, PI, and Simpson's Index were less well explained by LC variables than EPT Taxa, and Shannon's Index. Additionally, LC variable sample from 500m and 1000m buffers were better at explaining variation in BMI metrics than the 100m buffer or the watershed buffer (Table 8).

LCO's 500m buffer performed the best, with EPT Taxa and Shannon's Index scoring above  $R^2 = 0.60$  and PI scoring above  $R^2 = 0.50$  (Table 12). After that, %CC's 1000m buffer treatment was well correlated with EPT Taxa (Figure 4a,  $R^2 = 0.53$ , p < 0.000001), PI (Figure 4b,  $R^2 = 0.52$ , p < 0.000001), and Shannon's Index (Figure 4d,  $R^2 = 0.52$ , p < 0.000001). Slope's 1000m buffer treatment was well correlated with EPT Taxa (Figure 4d,  $R^2 = 0.52$ , p < 0.000001). Slope's 1000m buffer treatment was well correlated with EPT Taxa (Figure 4d,  $R^2 = 0.54$ , p < 0.000001) and Shannon's Index (Figure 4e,  $R^2 = 0.49$ , p < 0.000001) and Shannon's Index (Figure 4f,  $R^2 = 0.54$ , p < 0.000001).

I found variability in R<sup>2</sup> values for certain treatment and LC variable combinations. For example, the R<sup>2</sup> values of RDE stayed constant across all circular buffer treatments, but dropped precipitously for the watershed treatment (Table 14 / Appendix A). In contrast, R<sup>2</sup>'s for LCO, %CC, and SL varied more for each treatment (Table 12).

Multi-Variable Linear Regressions (MLR)

I used stepwise multiple linear regression with all variables and treatments to generate optimal MLRs for each BMI metric (table 13). 100 meter circular buffer treatments were insignificant, except for %IM. Several LC variables were highly significant as long as other treatments of that variable were present, for example EV 1000, EV 500, and EV watershed, however once alone they were less significant. This also occurred for related variables such as RDI/RDE and EV/SL.

### DISCUSSION

In this study, I identified the primary trends in relationships between land characteristic variables and benthic macroinvertebrate health metrics in the San Francisco, California, Bay Area. I showed that the size of the sample area across which the land characteristic is sampled significantly affects results, with 100m buffers and watershed buffers performing worse than 500m and 1000m buffers at all spatial scales. Land cover type, percent canopy cover, and slope were the most important LC variables for predicting BMI water quality metrics.

#### Landscape Characteristic Variables & Models

The most explanatory LC variables were LCO, %CC, and SL (Table 12). These variables not only contain information reflecting stressors that affect stream water quality (9, 10, 14, 19), but are also the most efficient for this dataset at reflecting the extent of human development, with the capacity to detect human impact even at low levels of human activity (9).

Slope not only illustrates areas that are difficult to build on and thus less settled, but can discriminate between intensely and less-intensely developed areas if the data resolution is high enough. Further, slope also can be used locally as a proxy to measure erosion, channelization, and other stressors that directly affect BMI bioindicators (6).

Land cover also distinguishes between areas with no human settlement impact and areas with human settlement impact. However, this measure was most effective because it includes breaks urban and non-urban down into further land types, including forests, grasslands, cropland, shrub, and pasture (20).

%CC also reflects both human activity and natural processes that affect water quality in streams. Roads, buildings, and power lines require a footprint of cleared land, and this represents itself in the canopy coverage pattern. Further, %CC not only measures this aspect of human settlement, but also contains information regarding BMI environmental conditions: many species of BMI rely on terrestrial inputs such as fallen tree leaves for nourishment (21).

While variables such as %IM can highlight large human settlements, they fail at discriminating between rural areas and smaller residential enclaves that have a smaller impact. Since nearly all of the sampled areas were remote from urban or suburban centers with large populations or paved surfaces, %IM and population density were all around the same level and inefficient at predicting BMI water quality metrics (Table 14,). Arnolds and Gibbons found that stream degradation from %IM related stressors occurs at 10% or higher impervious surface coverage (9), yet my results showed 95% (421 of 444 sites) of the watershed-sampled data were below 10% IM coverage, while 70% (310 of 444 sites) of the buffer-sampled data were below that threshold. Even taking more sensitive theories about the impact of impervious surface coverage into account (22), 79% (350 of 444 sites) of the watershed-sampled IM data and 62% (275 of 444 sites) of the IM data is below that threshold. Therefore, the sampled region was not enough of an urban-rural gradient for %IM to correlate well with BMI health.

EV was ineffective because while correlated with some measures that affect water quality, it does not actually affect water quality, and the sample sites were largely at a similar band of elevations (Table 14).

### **Benthic Macroinvertebrates**

EPT Taxa and Shannon's Index were the most responsive measures to variation in LC variables (Table 10), while EPT Index, PI, and Simpson's Index were less responsive to LC variables (Table 14). EPT Taxa and Shannon's Index likely performed better due to their absolute nature, given the variety of site environments and the variety of benthic macroinvertebrate assemblages at each site. For a selection of sites with similar environments and assemblages, EPT Index, PI, and Simpson's Index would be more useful measures.

#### **Spatial Sampling Issues (Buffer & Watershed)**

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The 500 and 1000m buffer sampling zones were the best predictors, with the 100m buffer and watershed sampling zones proving the least useful sampling size for this dataset (Table 12). The 1000m buffer was slightly more accurate in all cases except for with LCO. While Sliva and Williams found that catchment-scale analysis was slightly more accurate than 100m buffers, they did not test any buffers larger than 100m (14). However, their finding that catchment-level analysis was slightly more accurate than 100m buffers was replicated in this study (Table 10).

The watershed analysis would likely be most useful for study areas closer to settled areas in the urban – rural gradient, while the 500m and 1000m buffers seem useful for site-specific restoration planning. Another potential alternative that could merge the strengths of both these approaches is a distance-based buffer around the stream itself (14).

### **Limitations and Future Directions**

The main limitation of this study was that its study design was intentionally broad so as to take advantage of the entire BMI dataset. This limited the power, accuracy, and applicability of its findings, as site conditions such as stream order, elevation, or adjacent terrestrial environment were ignored. The breadth of the study also reduced opportunities for detailed analysis of landscape characteristics interrelationships and the use of more specific BMI metrics or site condition measures. Finally, it introduced a reliance on the BMI data and its sampling procedure, which selected sites which were predisposed to a certain proximity to road networks and distance from urban areas, which limited the range of landscape characteristics.

There are some limitations related to the statistical distribution of the LC data, as they were largely non-normal, highly skewed, and leptokurtic. This reduced the number of options for statistical tests, and the number of treatments that could be used for linear regression. Large numbers of zero values or extremely low values also biased the linear regression and other values, but removing these values was not an option within the scope of the study. While some of these statistical limitations may be due to the resolution of the source datasets, it is unlikely that all of this variation originates from the low resolution of the source data. Given the low %IM and RDE values (Table 8), rural areas were more predominantly sampled than urban or suburban areas, creating a bias in sampling site selection and thus in LC values.

This study also had several methodological limitations with regards to the GIS component. The study did not calculate error accumulation, either inherent in NLCD or SWAMP datasets, or propagated through ArcGIS analysis.

In addition, land cover type was underused relative to the other continuous data layers. The Anderson land cover classification proposed in 1976 (23) has long become the standard land cover classification system, but such one-dimensional systems have accumulated their share of criticism (24 - 27).

The results in this study also point to significant potential future research. Splitting the dataset into subsets based upon the BMI water quality data would allow more precise analysis of specific relationships between BMI and LC values. Land cover could be better integrated into the study by using patch dynamics to break land cover down into quantitative metrics such as patch density, patch size, and shape indices (28), or by reclassifying continuous LC variables into discrete, categorical variables as in suitability analysis (29, 30). Alternatively, a more focused approach could take a single LC variable and examine its relationships to functional feeding groups and other site characteristics such as water chemistry or the physical form of the site.

# CONCLUSION

This study examined new BMI data available for the San Francisco, California, Bay Area. Sites with high and low BMI scores often shared the same range of landscape characteristic values. This reinforces that multiple variables need to be considered in any analysis of BMI scores (19). Sites may possess landscape characteristics that would create a good environment for benthic macroinvertebrates, but which are counterbalanced by the shortcomings of another LC variable (31).

Higher scores of %CC, slope, and non-urbanized values of LCO were the main factors associated with high BMI scores, particularly Shannon's Index and EPT Taxa. This suggests expanding planting of tree cover over and near non-vegetated creeks could improve stream water quality and assemblage health (31, 32) and would also improve property values (33). Given the presence of previous restoration of culverted and underground creeks in the Berkeley area (34), and the presence of creek stewardship groups like The Urban Creeks Council, Friends of Five

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Creeks, The Watershed Project, and the East Bay Chapter, these findings or findings from future research may prove useful to these groups for decision making.

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Metric	Description	Use
EPT Taxa	Total number of Ephemeroptera,	Indicate general level of water quality
(abundance)	Plecoptera, and Trichoptera (EPT) found	at site.
EPT Index	Number of different taxa of	Indicate general level of water quality
(richness)	Ephemeroptera, Plecoptera, and	at site.
	Trichoptera (EPT) given as % of total	
BMI richness (PI)	Number of different taxa of <b>B</b> enthic	In combination with EPT data, helps
	Macroinvertebrates found given as % of	identify whether EPT data results are
	total	due to water pollution conditions or
		environmental conditions
Shannon-Weaver	The proportion of a single species relative	Diversity indices account for
<b>Diversity Index</b>	to the total number of species, multiplied	abundance (how many individuals
·	by natural logarithm of that proportion.	there are), richness (how many
Simpson's Diversity	The reciprocal of the summed squared	species there are), and evenness (how
Index	proportions of a single species relative to	a species is distributed) of a single
	the total number of species.	species in relation to the community.

 Table 1. BMI metrics used in this study. These metrics are standard metrics commonly used in benthic macroinvertebrate studies that are easy to calculate and use, while communicating different information.

 Table 2: LC variables used in this study. These variables were freely available as part of the NLCD on the National Map Viewer (<a href="http://nationalmap.gov/viewer.html">http://nationalmap.gov/viewer.html</a> ). \* SRTM = Shuttle Radar Topography Mission, USGS = US Geological Survey, DEM = Digital Elevation Model.

 Landscape

Landscape			
Variable	Abbreviation	Units of Variable	Associated Factors
Percent Canopy	%CC	Area with canopy cover /	Terrestrial inputs, temperature
Cover		total area of cell	effects through shade
Road Density	RDE	km of road / km <sup>2</sup> of area	Pollutants, run-off, vibrational
			disturbance
<b>Distance to Nearest</b>	RDI	Distance to nearest road from	Pollutants, run-off, vibrational
Road		cell center point in meters	disturbance
Elevation	EV	Distance from SRTM*	Stream order, stream
		USGS DEM designated zero	geomorphology
		elevation (sea level) in	
		meters.	
Percent Impervious	%IM	Area with impervious surface	Urbanization impacts, run-off
Surface Coverage		cover / total area of cell	
<b>Population Density</b>	PD	People per mile <sup>2</sup>	Urbanization impacts
Slope	SL	Degrees rise per cell	Stream order, stream
			geomorphology, erosion,
			channelization

Table 3: Median mean values,  $\chi^2$ , and p-values of landscape variables per transect and Kruskal-Wallis Test Result (df = 3, crit value = 7.815). This test shows that while there are differences in the landscape variables between transect sampling methods, these differences are not significant. (\* = significant results; <sup>x</sup> = groups were similar, <sup>†</sup> = groups were different).

	Three Transect	X (Single			
Medians	Samples	Transect)	<b>Result</b> $(\chi^2)$	<i>p</i> -value	Result
%CC	27.2	24.0	0.2362	.9722	†
RDE	2.90	4.06	37.50	< .00001	X *
RDI	123	60.9	17.65	.0005	X *
EV	135	131	0.9657	.8095	x *
%IM	1.31	4.43	28.48	< .00001	x *
PD	93.8	60.0	3.221	.3588	†
SL	12.2	13.0	4.52	.2100	†

Table 4: Normality of data per landscape variable and treatment type. All data were explicitly non-normal, although watershed treatments were more normal than circular buffer treatments. All *p*-values are less than 0.00001 unless otherwise stated. W = Shapiro-Wilks statistic denoting degree of normality with 0 = non-normal and 1 = perfectly normally distributed. (<sup>†</sup> = potentially normal, <sup>† †</sup> = relatively normal).

	Circular Buffer Treatment							
Distance of Buffer	100m	500m	1000m	Watershed Treatment				
Population Density	W = 0.389	W = 0.5377	W = 0.5982	W = 0.3487				
% Impervious Cover	W = 0.6977	W = 0.6896	W = 0.7059	W = 0.3754				
% Canopy Cover	W = 0.8552 <sup>+</sup>	W = 0.8603 <sup>+</sup>	W = 0.8841 <sup>†</sup>	W = 0.9598 <sup>†</sup> <sup>†</sup>				
Road Density	W = 0.8452 <sup>†</sup>	$W = 0.8456^{+}$	W = 0.845 <sup>†</sup>	W = 0.5974				
Road Distance	W = 0.5375	W = 0.8281 <sup>†</sup>	W = 0.9353 <sup>+</sup> <sup>+</sup>	W = 0.9252 <sup>†</sup> <sup>†</sup>				
Elevation	W = 0.8345 <sup>†</sup>	W = 0.8699 <sup>+</sup>	W = 0.8974 <sup>+</sup>	W = 0.9885 <sup>+</sup> <sup>+</sup> p-value = 0.001434				
Slope	$W = 0.8961^{+}$	$W = 0.9396^{++}$	W = 0.9384 <sup>†</sup> <sup>†</sup>	$W = 0.92^{++}$				

**Table 5:** Numerical summaries of LC variable data. STD = standard deviation, cv = coefficient of variation (standard deviation divided by mean), IQR = interquartile range. Skew measures the distortion of a probability distribution's spread from the mean; kurtosis measures how distorted a probability distribution's magnitude is.

Landscape	Sampling							
Variable	Buffer Size	mean	STD	median	IQR	cv	skew	kurtosis
% Canopy	0100m	32.6	30.1	22.1	60.5	0.920	0.368	-1.5
Cover	0500m	29.6	28.3	20.0	47.2	0.955	0.539	-1.2
	1000m	28.8	26.7	20.5	43.4	0.928	0.592	-0.95
	watershed	35.3	22.5	34.8	33.0	0.638	0.185	-0.89
<b>Road Density</b>	0100m	5.44	4.75	3.66	5.79	0.873	1.20	0.518
	0500m	5.38	4.68	3.49	5.68	0.868	1.19	0.468
	1000m	5.26	4.55	3.44	5.50	0.865	1.16	0.338
	watershed	3.01	2.74	2.26	1.98	0.909	3.89	18.4
Distance to	0100m	64.3	87.1	36.0	26.9	1.35	3.68	16.0
Nearest Road	0500m	115.	88.2	109	97.1	0.765	2.06	6.57
	1000m	159.	109.	145	141.	0.686	0.831	0.441
	watershed	264.	124.	263	150.	0.469	1.11	3.28
Elevation	0100m	97.5	90.1	73.3	111.	0.925	1.87	5.23
	0500m	120.	102.	106	119.	0.844	1.60	3.87
	1000m	139.	109.	135	135.	0.782	1.35	2.91
	watershed	292.	126.	290	163.	0.433	0.0907	0.0162
% Impervious	0100m	10.9	16.5	2.79	12.5	1.52	1.73	1.98
Cover	0500m	12.2	18.6	1.58	16.2	1.52	1.52	0.965
	1000m	12.3	18.4	1.26	19.2	1.49	1.52	1.10
	watershed	3.76	9.58	0.490	4.37	2.55	5.36	31.2
Population	0100m	1205.	3398.	69.9	743.	2.82	5.18	31.3
Density	0500m	1120.	2541.	93.9	849.	2.12	3.26	12.8
	1000m	1246.	2344.	111	1512.	1.88	2.72	8.49
	watershed	397.	1096.	64.9	355.	2.76	6.11	43.5
Slope	0100m	8.07	7.14	6.26	10.5	0.884	0.942	0.257
-	0500m	10.4	7.36	10.9	12.6	0.709	0.277	-0.947
	1000m	10.9	6.89	12.3	11.9	0.630	-0.0545	-1.18
	watershed	15.4	5.02	16.7	5.98	0.326	-1.01	0.743

 Table 6: Breakdown of land cover types per treatment. Buffer-sampled data were consistent between the three distances, excepting Urban – Open, while watershed-sampled data had less data spread across multiple categories.

 Treatments

	Treatments						
Land Cover Types	100m	500m	1000m	Watershed			
Cropland	7%	4%	3%	1%			
Forest - Evergreen	22%	26%	26%	37%			
Forest - Mixed	11%	11%	14%	30%			
Grassland	15%	28%	28%	27%			
Pasture	-	-	1%	-			
Shrub	1%	2%	2%	1%			
Urban - High	-	1%	-	1%			
Urban - Medium	10%	14%	14%	1%			
Urban - Low	7%	6%	5%	1%			
Urban – Open	25%	9%	7%	2%			
Woody Wetlands	1%	-	-	-			

**Figure 1: Distribution of data for LC Variables.** Histograms displaying data distribution for variables: (a) %IM, (b) Distance to Nearest Road, (c) % Canopy Cover, (d) Elevation, (e) Road Density, and (f) Slope.



**Figure 2: Correlations between LC variables.** Scatterplots of the watershed-sampled LC variables against one another, with a) %CC against EV, (b) %IM against %CC, (c) %CC against SL, (d) EV against SL, (e) RDE against RDI, and (f) SL against %IM.



**Table 7:**  $\mathbb{R}^2$  values of linear regression for LCO, %CC, and SL. Light blue cells have an  $\mathbb{R}^2$  above 0.50, light green cells have an  $\mathbb{R}^2$  above 0.40, light orange cells have an  $\mathbb{R}^2$  above 0.30, and dark red shaded squares have an  $\mathbb{R}^2$  below 0.20.

		Adjusted R <sup>2</sup>					
	Sampling Zone Sizes	EPT Taxa	EPT Index	Shannon's Index	Simpson's Index	PI	Average R <sup>2</sup>
	100.00	0.43	0.35	0.46	0.40	0.38	0.40
•	500.00	0.61	0.49	0.60	0.47	0.58	0.57
TCC	1000.00	0.63	0.45	0.58	0.43	0.55	0.55
	wtr	0.53	0.27	0.44	0.32	0.41	0.42
	Average R <sup>2</sup>	0.55	0.39	0.52	0.40	0.48	
	100.00	0.38	0.41	0.44	0.32	0.40	0.39
	500.00	0.50	0.45	0.52	0.36	0.52	0.48
сc	1000.00	0.53	0.45	0.52	0.36	0.53	0.49
	wtr	0.49	0.33	0.43	0.33	0.39	0.41
	Average R <sup>2</sup>	0.48	0.41	0.48	0.34	0.46	
	100.00	0.27	0.32	0.34	0.24	0.28	0.29
	500.00	0.44	0.42	0.51	0.39	0.43	0.44
SL	1000.00	0.49	0.45	0.54	0.42	0.47	0.48
	wtr	0.33	0.28	0.34	0.29	0.27	0.31
	Average R <sup>2</sup>	0.38	0.37	0.43	0.34	0.36	
	100.00	0.43	0.30	0.41	0.37	0.30	0.37
	500.00	0.43	0.31	0.41	0.37	0.31	0.37
DE	1000.00	0.44	0.31	0.42	0.38	0.31	0.38
	wtr	0.16	0.12	0.18	0.20	0.12	0.15
	Average R <sup>2</sup>	0.37	0.26	0.36	0.33	0.26	
	100.00	0.34	0.34	0.39	0.42	0.24	0.34
ľ	500.00	0.39	0.34	0.40	0.38	0.28	0.36
%II	1000.00	0.41	0.35	0.41	0.39	0.29	0.37
Ŭ	wtr	0.14	0.12	0.17	0.19	0.10	0.14
	Average R <sup>2</sup>	0.32	0.29	0.34	0.34	0.23	
	100.00	0.06	0.04	0.05	0.03	0.04	0.05
	500.00	0.26	0.15	0.23	0.17	0.18	0.21
IQ	1000.00	0.36	0.19	0.32	0.25	0.25	0.28
	wtr	0.09	0.06	0.08	0.08	0.07	0.08
	Average R <sup>2</sup>	0.19	0.11	0.17	0.13	0.13	
	100.00	0.08	0.15	0.11	0.07	0.11	0.10
	500.00	0.13	0.20	0.17	0.12	0.17	0.16
EV	1000.00	0.19	0.25	0.24	0.16	0.22	0.22
	wtr	0.21	0.21	0.19	0.12	0.22	0.20
	Average R <sup>2</sup>	0.15	0.20	0.18	0.12	0.18	

PD	100.00	0.09	0.06	0.06	0.04	0.06	0.07
	500.00	0.19	0.13	0.16	0.13	0.13	0.15
	1000.00	0.26	0.18	0.22	0.18	0.18	0.21
	wtr	0.11	0.09	0.13	0.14	0.08	0.11
	Average R <sup>2</sup>	0.16	0.12	0.15	0.12	0.11	

**Figure 3: BMI variables plotted and regressed against LC variables.** Scatterplots with box plots showing data distribution for: (a) EPT Taxa against %CC, (b) PI against %CC, (c) EPT Index against %CC, (d)Shannon's Index against %CC, (e) EPT Taxa against SL, and (f) Shannon's Index against SL.



**Table 8: Stepwise minimal multiple linear regression.** The stepwise regression was calculated manually, with non-significant factors with largest *p*-values removed first, then with least significant factors duplicating a single LC variable removed until a single model with only one treatment for each LC variable was left. All models have *p*-value <.000001.

BMI	LC Variables use	Adjusted	
Metric	<b>Circular Buffer</b>	Watershed	$\mathbf{R}^2$
EPT Taxa	CC 500	EV watershed	0.6428
	RDI 1000		
	RDE 1000		
EPT Index	IM 0100	EV watershed	0.5476
	CC 500		
	RDI 500		
	RDE 1000		
Shannon's	CC 500	RDE watershed	0.6452
Index	EV 500		
	RDI 1000		
	SL 1000		
Simpson's	IM 100	SL watershed	0.5510
Index	CC 500		
PI	CC 500	RDE watershed	0.5895
	SL 1000	IM watershed	