Maryland Biological Stressor Identification Process

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List of Abbreviations

AMD	Acid Mine Drainage
ANC	Acid Neutralizing Capacity
AR	Attributable Risk
BIBI	Benthic Index of Biotic Integrity
BSID	Biological Stressor Identification
COMAR	Code of Maryland Regulations
CWA	Clean Water Act
DO	Dissolved Oxygen
FIBI	Fish Index of Biologic Integrity
IBI	Index of Biotic Integrity
LULC	Land Use Land Cover
MBSS	Maryland Biological Stream Survey
MDDNR	Maryland Department of Natural Resources
MDE	Maryland Department of the Environment
mg/L	Milligrams per liter
MSSCS	Maryland Synoptic Stream Chemistry
µeq/L	Micro equivalent per liter
µS/cm	Micro Seimens per centimeter
P/G/E	Pool/Glide/Eddy
QA/QC	Quality Assurance/Quality Control
RESAC	Regional Earth Science Applications
SSA	Science Services Administration
TMDL	Total Maximum Daily Load
USEPA	United States Environmental Protection
WQA	Water Quality Analysis
WQLS	Water Quality Limited Segment

Executive Summary

Section 303(d) of the federal Clean Water Act (CWA) and the U.S. Environmental Protection Agency's (USEPA) implementing regulations direct each state to identify and list waters, known as water quality limited segments (WQLSs), in which current required controls of a specified substance are inadequate to achieve water quality standards. For each WQLS listed on the 303(d) List in the *Integrated Report of Surface Water Quality in Maryland* (Integrated Report), the State is to either establish a Total Maximum Daily Load (TMDL) of the specified substance that the waterbody can receive without violating water quality standards, or demonstrate via a Water Quality Analysis (WQA) that water quality standards are being met.

Current Integrated Report listing categories are:

- Category 2 ("meeting some water quality standards, but with insufficient data to assess completely"), if the potential or relevant stressors were found not to be present or to have a limited association with biological integrity in the subject segments.
- Category 3 ("insufficient data to determine if any water quality standard is being attained"), if the potential or relevant stressors were identified as having insufficient data to directly link them to degrading biological conditions in the subject segments.
- Category 4c ("waterbody impairment is not caused by a pollutant"), when the only remedy for degraded biological conditions in the subject segments is a technical correction.
- Category 5 ("does not meet water quality standards"), if the potential or relevant stressors were degrading biological conditions in the subject segments.

In 2002, the State began listing biological impairments on the Integrated Report. The current Maryland Department of Environment (MDE) biological assessment methodology assesses and lists at the Maryland 8-digit watershed scale, which maintains consistency with how other listings on the Integrated Report are made, how TMDLs are developed, and how implementation is targeted. The listing methodology assesses the condition of Maryland 8-digit watersheds with multiple impacted sites by measuring the percentage of stream miles that are degraded, and calculating whether they differ significantly from a reference condition watershed.

Maryland developed water quality standards to protect, maintain and improve the quality of Maryland surface waters. A water quality standard is the combination of a designated use for a particular body of water and the water quality criteria designed to protect that use. Designated uses include support of aquatic life, primary or secondary contact recreation, drinking water supply, and shellfish propagation and harvest (COMAR 2014a). Water quality criteria consist of narrative statements and numeric values designed to protect the designated uses. There are numerous 8-digit watersheds in Maryland that are not attaining their designated use because of biological impairments. As an indicator of designated use attainment, MDE uses Fish and Benthic Indices of Biotic Integrity (BIBI/FIBI) developed by the Maryland Department of Natural Resources Maryland Biological Stream Survey (MDDNR MBSS).

The current listings for biological impairments represent degraded biological conditions for which the stressors, or causes, are unknown. The MDE Science Services Administration (SSA) has developed a biological stressor identification (BSID) analysis that uses a case-control, risk-based approach to systematically and objectively determine the predominant cause and source of degraded biological conditions, which will enable the Department to most effectively direct corrective management action(s).

MDE SSA generated a principal dataset after a quality assurance/quality control (QA/QC) review and vetting process of the Maryland Department of Natural Resources Maryland Biological Stream Survey (MDDNR MBSS) round two and round three data. Parameters were selected from the principal dataset to represent either specific “stressors” or potential “sources” of stressors. Stressors were grouped into categories representing sedimentation, habitat conditions or water chemistry.

The BSID analysis is a risk-based approach, adapted from the field of epidemiology, which estimates the strength of association between various stressors and the biological community, and the potential improvement of biology if a given stressor were removed. The assessment compares the likelihood that biological condition is degraded, given that a stressor is beyond its threshold, by using the ratio of the prevalence within the case group as compared to the prevalence in the control group. The case group is defined as the sites within the assessment unit with degraded biological conditions, and the control group is defined as the sites with similar physiographic characteristics that have good biological conditions. In Maryland three physiographic eco-regions were identified from the MDDNR MBSS index of biotic integrity (IBI) metrics: Highland, Eastern Piedmont, and Coastal (Southerland et al. 2005b).

Results of the BSID analysis may identify one or several stressors (pollutants and habitat aberrations) as likely causes of the poor biological conditions within the Maryland 8-digit watershed. The results can be used together with a variety of analyses to update and/or support the probable causes and sources of biological impairment in the Integrated Report.

1. Introduction

Section 303(d) of the federal Clean Water Act (CWA) and the U.S. Environmental Protection Agency's (USEPA) implementing regulations direct each state to identify and list waters, known as water quality limited segments (WQLSs), in which current required controls of a specified substance are inadequate to achieve water quality standards. For each WQLS listed on the 303(d) List in the *Integrated Report of Surface Water Quality in Maryland* (Integrated Report), the State is to either establish a Total Maximum Daily Load (TMDL) of the specified substance that the waterbody can receive without violating water quality standards, or demonstrate via a Water Quality Analysis (WQA) that water quality standards are being met.

In 2002, the State began listing biological impairments on the Integrated Report. The current Maryland Department of Environment (MDE) biological assessment methodology assesses and lists at the Maryland 8-digit watershed scale (average watershed size approximately 90 mi^2), which maintains consistency with how other listings on the Integrated Report are made, how TMDLs are developed, and how implementation is targeted. The listing methodology assesses the condition of Maryland 8-digit watersheds with multiple impacted sites by measuring the percentage of stream miles that are degraded, and calculating whether they differ significantly from a reference condition watershed (i.e., healthy stream based on reference sites determined independent of biological condition).

The current listings for biological impairments represent degraded biological conditions for which the stressors, or causes, are unknown. The MDE Science Services Administration has developed a biological stressor identification (BSID) analysis that uses a case-control, risk-based approach to systematically and objectively determine the predominant cause(s) of reduced biological conditions, which will enable the Department to most effectively direct corrective management action(s). The risk-based approach, adapted from the field of epidemiology, estimates the strength of association between various stressors and the biological community, and the likely improvement of biology if a given stressor were removed.

2. Biological Impairments

MDE's Integrated Report listing methodology incorporates indices of biological integrity (IBI) to determine attainment of the designated use of aquatic life protection. IBIs are broad, comprehensive measures of biological condition that represent numerous individual metrics that are scored based on comparison to reference conditions. An IBI score compares existing with expected conditions at sample sites using region-specific baseline conditions that reflect little or no human impact. In Maryland three physiographic eco-regions were identified from the Maryland Department of Natural Resources Maryland Biological Stream Survey (MDDNR MBSS) IBI metrics: Highland, Eastern Piedmont, and Coastal Plain (Southerland et al. 2005a). The three eco-regions are identified in Figure 1 below.

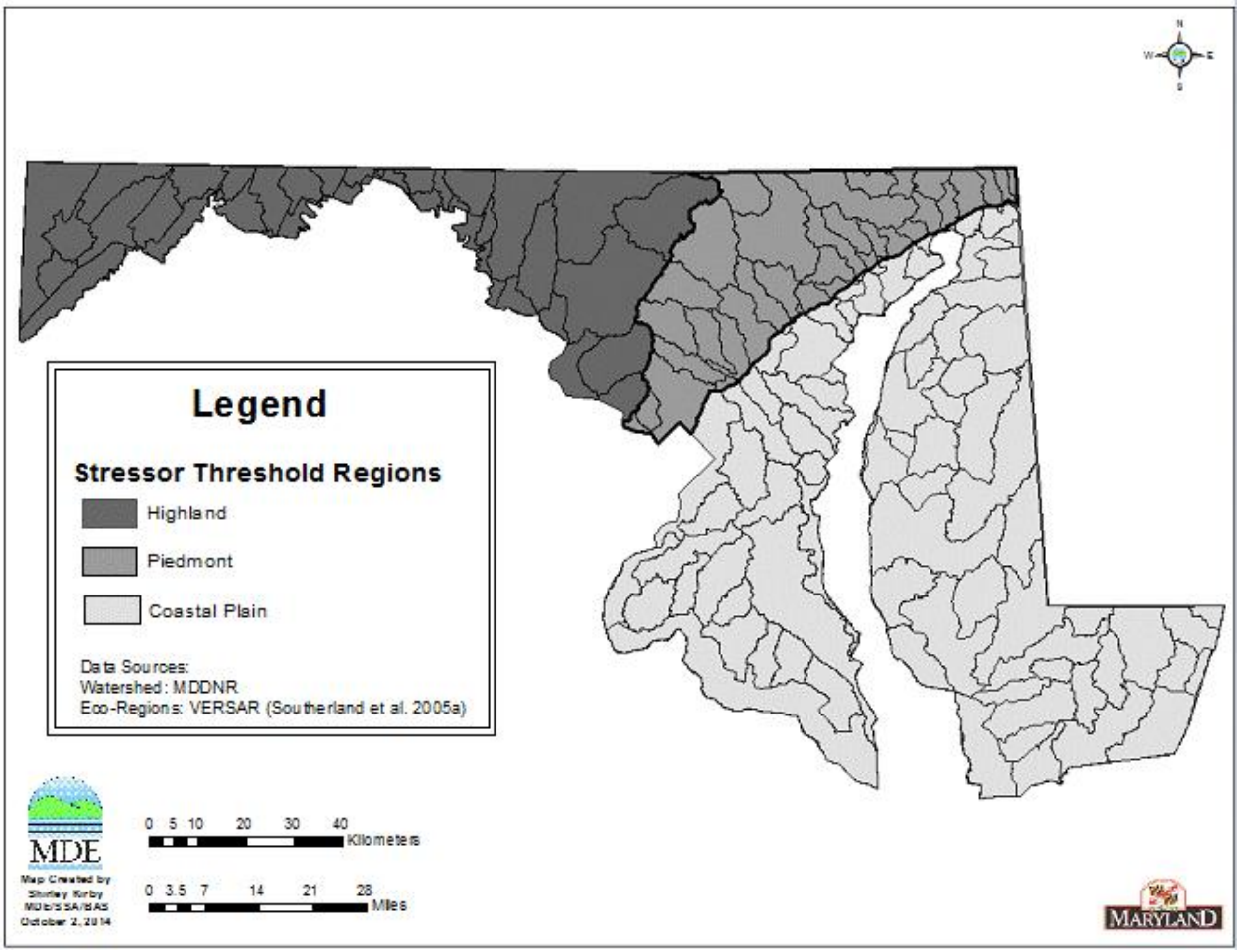

Figure 1. Eco-region map of Maryland.

Benthic and fish IBIs (BIBI and FIBI, respectively) are quantitative ratings of the integrity of benthic macroinvertebrate and fish assemblages found at each site. Scores below the threshold value of 3 indicate poor biological conditions. Table *1* contains a more detailed description of each of the IBI categories developed.

Table 1. IBI Metrics (Mercurio et al. 1999).

Narrative descriptions of stream biological integrity associated with each IBI category.		
Good	IBI score 4.0 - 5.0	Comparable to reference streams considered to be minimally impacted. Fall within the upper 50% of reference site conditions.
Fair	IBI score 3.0 - 3.9	Comparable to reference conditions, but some aspects of biological integrity may not resemble the qualities of these minimally impacted streams. Fall within the lower portion of the range of reference sites (10th to 50th percentile).
Poor	IBI score 2.0 - 2.9	Significant deviation from reference conditions, with many aspects of biological integrity not resembling the qualities of these minimally impacted streams, indicating some degradation.
Very Poor	IBI score 1.0 - 1.9	Strong deviation from reference conditions, with most aspects of biological integrity not resembling the qualities of these minimally impacted streams, indicating severe degradation.

Maryland's IBIs assess biological integrity by comparing the community structure of streams to that of high quality (or reference) streams. Biological integrity is influenced by five broad factors: biological interactions, flow regime, energy source, water chemistry, and physical habitat (Karr 1991). Biological impairments could result from the influence of one or any combination of factors. All stream parameters available to diagnose the cause of biological impairments were carefully reviewed to generate the best possible representation of each factor to ensure the most comprehensive stressor identification.

Biological interactions such as competition and predation are dynamic controls for species population sizes within any community. Anthropogenic influences such as the inadvertent or intentional introduction (e.g., fish stocking) of exotic species may amplify the divergence of community structure from reference conditions, thus indicating biological impairment.

The biota of aquatic systems is dependent on a recurring flow pattern including both high and low flow conditions to sustain functions such as feeding, reproduction, and dispersal. Altered flow regimes that either homogenize flow conditions (e.g., dams) or exaggerate extreme conditions (e.g., increased surface flow from impervious surface) may not provide adequate conditions to sustain populations (e.g., periodic flush of sediment from interstitial spaces, sustained current to support feeding strategy) or diversity.

Aquatic community structure reflects the mosaic of energy inputs into each stream system due to the association of organisms with unique feeding strategies. The proportion of allochthonous inputs (originating from outside the aquatic system) or autochthonous inputs (originating within the aquatic system) as well as the size of available organic materials (e.g., coarse or fine particulates) may determine which species proliferates in a community. Any modifications that could effect a change in the energy source of a system (e.g., increased nutrients, increased fine particulate organics, increased sunlight, increased temperature, decreased leaf litter or woody debris) could alter community composition, thus biological integrity.

Water chemistry is the most commonly considered factor controlling biological integrity because we have long recognized that biological organisms have specific tolerances and requirements. Exceedance of species tolerances (e.g., dissolved oxygen, pH) may reduce or eliminate populations thus altering biological integrity.

Proliferation of aquatic organisms is dependent on adequate physical habitat, including substrate, current, and temperature. Diversity of physical habitat generally supports larger number of community members. If the diversity of physical habitat is reduced (e.g., channel widened, channel straightened, woody debris removed, etc.) fewer species may find suitable conditions for feeding and reproduction, thus altering community structure.

3. Data Used in Stressor Identification

The BSID analysis is based primarily on the MDDNR MBSS dataset, which provides a statewide broad spectrum of paired data variables (i.e., instream biological data are paired with chemical, physical, and land use variables). This principal dataset uses a statewide probability-based sampling design to assess the biological condition of first-, second-, third-, and fourth-order non-tidal streams (determination based on the solid blue line shown on U.S. Geological Survey 1:100,000-scale maps) within Maryland's 8-digit watersheds (Klauda et al. 1998, Roth et al. 2005). MDDNR MBSS sites are sampled within a 75-meter segment of stream length. The MDDNR MBSS conducted three rounds of sampling between 1995 and 2009. This BSID analysis is an update of the previous BSID analysis, which was run in 2009 using data from round two (2000-2004) (MDE 2009). The current analysis was constrained to rounds two and three (2000-2004 and 2007-2009) to base the stressor identifications on the most recent data. MBSS methodology is discussed in detail in Roth et al. (2005); quality assurance is addressed in MDNR (2009).

MDE conducted a thorough data quality review and vetting process of all MDDNR MBSS round two and three randomly sampled data to ensure that they meet the biological listing methodology criteria of the Integrated Report (MDE 2008). In all cases, State biologists may use professional judgment in evaluating biological results. However, to aid in the data review, a set of rules was used guide the data vetting process. These rules evaluate specific data parameters such as flow, catchment size, and buffer width to determine whether the IBIs are reliable indicators of current watershed conditions. As a specific example, if there was a temporary or significant natural stressor such as drought or flood, sample results were evaluated to determine whether IBI scores resulted from anthropogenic influences or natural conditions. The final master database contains all biological sites considered valid for use in the assessment process. MDE, with help from DNR, developed nine vetting rules for eliminating site results with IBIs that are not representative of stream condition. The nine vetting rules are:

1. Sampling locations with less than 300-acre catchment
2. Blackwater streams, due to their unique chemistry and lack of a defined blackwater reference conditions
3. Fewer than 60 organisms in a benthic sample
4. Heavy rain and other runoff events
5. Tidally influenced sampling sites
6. Streams affected by excessive drought or intermittent conditions (i.e., low flow)
7. Sampling sites dominated by wetland-like conditions
8. Streams impounded by beaver dams
9. Sampling sites where the results may be skewed due to sampling error

In addition to these circumstances, State biologists may use best professional judgment to evaluate any streams sampled under conditions that are not characterized by reference stations. The final master dataset contains all random round two and three biological sites considered valid for use as the principal dataset for the BSID analysis and the listing process. For a more detailed reference, please see section "DRAFT Biological Assessment Methodology for Non-

Tidal Wadeable Streams" of the Maryland 2014 Integrated Report of Surface Water Quality (MDE 2014).

The rounds two and three datasets contain counts from numerous taxonomic groups (e.g., fish, macroinvertebrates, reptiles, amphibians), have more than 190 abiotic parameters, and identify upstream drainage areas for calculation of spatial information (e.g., land use proportions). Each abiotic parameter represents a specific ecosystem component within the watershed (e.g., physical habitat, water chemistry, and land use sources).

The MDDNR MBSS dataset has three data types for abiotic parameters. First, continuous quantitative parameters (e.g. chemical data) have a wide range of numerical values. Next, ordinal, qualitative habitat parameters (e.g. pool/glide/eddy quality) are typically integer values with a logical numerical order (scale 0-20). Finally, binary variables (e.g., concrete/gabion present) have a logical present or absent (yes/no) value.

The State of Maryland is required to consider all readily available data for listing impairments in the Integrated Report; therefore, relevant data from federal, state, and county environmental programs, and from private organizations, will be reviewed for possible inclusion into the principal dataset. For inclusion in the principal dataset, all relevant data must incorporate all MDDNR MBSS round two and three parameters and be consistent with all MDDNR MBSS protocols.

4. Stressors and Sources

Parameters were selected from the principal dataset to represent either specific "stressors" or potential "sources" of stressors causing biological degradation. Parameters representing stressors are grouped into four categories: 1) sediment transport and deposition, 2) instream habitat condition, 3) riparian habitat condition, and 4) water chemistry. Parameters representing potential sources of stressors are grouped into two categories: land uses within a watershed and potential sources of acidity.

4.1. Stressor and Source Thresholds

To ultimately compare biological conditions to stressor conditions, stressor thresholds are needed to differentiate biologically harmful stressor levels from biologically tolerable stressor levels. Some parameters have existing threshold values as defined by MDDNR MBSS, the Code of Maryland Regulations, or literature; when available, these were compiled and used in the BSID. For the parameters without existing threshold values, however, they were established for this analysis to indicate levels above which degradation to biological communities is likely to occur.

Threshold values were determined by comparing stressor levels among different biological conditions. First, sites were pooled into good, fair, poor, and very poor benthic and fish IBI groups, further stratified by eco-region (Highland, Eastern Piedmont, and Coastal). Next, the sites' stressor measures within these groups were bootstrapped to 10,000 iterations (Canty et al. 2012) to better represent percentile distributions. For stressors in which high values are detrimental (e.g., sulfates), 90th percentiles were calculated. For stressors in which low values are detrimental (e.g., percentage of forest cover in watershed), 10th percentiles were calculated (Mowat et al. 2008). Graphs displaying the 80% confidence intervals of these grouped percentile distributions were generated for each stressor, providing an avenue to analyze variation of stressor levels among different IBI scores (Appendix A).

These percentile distributions were then tested for statistical significance. Since the goal was to delineate stressor levels beyond which biology is poor, the poor group ($2 \leq IBI < 3$) and the fair group ($3 \leq IBI < 4$) were compared first. Non-overlapping confidence intervals in the direction of interest indicate at least 90% confidence that the groups are statistically distinct. In this case, the default recommendation was the mean of the fair group. If the intervals did overlap, the very poor ($1 \leq IBI < 2$) and good ($4 \leq IBI \leq 5$) groups were similarly compared, and the default recommendation was the mean of the poor and fair groups. These default threshold recommendations were generated systematically (recorded on the right side of tables in Appendix A) and informed the final threshold decisions. Each stressor, region, and biological community comparison was then individually reviewed by MDE to ensure sound BSID threshold determinations.

4.2. Stressors

4.2.1. Indicators of Sediment Transport and Deposition

MDE selected several parameters from the principal dataset that evaluate the overall amount of sedimentation in the stream and provide information about the hydrologic regime of the watershed. The sedimentation parameters used in the BSID analysis are: bar formation, channel alteration, embeddedness, epifaunal substrate, and presence of erosion. Each of these parameters is measured once during summer index period.

Bar Formation

Bar Formation represents deposition of sand, gravel, and small stones in an area of the stream with a gentle slope and an elevation very close to the stream's water level. Bar formation typically reflects the overall sediment transport capacity of the stream with observed categories of moderate to extensive or extensive bar formation present. Moderate to extensive bar formation indicates channel instability related to frequent and intense high stream velocities that quickly dissipate and rapidly lose the capacity to transport excessive sediment loads downstream (Allan et al. 2007).

Sediment loads may originate from terrestrial (surface) erosion or from instream channel/bank erosion. Excessive sediment loading is expected to reduce and homogenize available feeding and reproductive habitat, degrading biological conditions (Allan 2004). Distinguishing between terrestrial or aquatic sources of sediment is not possible from this measure. Since many pollutants readily attach to sediment particles, it is possible that this parameter may also represent the presence of pollutants other than sediment. For example, sediment loads from terrestrial erosion may also introduce phosphorus into the stream segment. Conditions indicating biological degradation are *moderate bar formation present* and *extensive bar formation present*.

Channel Alteration

Channel Alteration is a rating of large-scale changes in the shape of a stream channel. This rating addresses deliberate stream manipulations within a 75-meter sample station (e.g., concrete channels, artificial embankments, obvious straightening of the natural channel, rip-rap, or other structures), as well as stream alterations resulting from large changes in hydrologic energy (e.g., recent bar development; Mercurio et al. 1999). Deliberate alterations typically result in higher velocities by smoothing channel surfaces, straightening channels, or raising/steepening banks. Thus, the presence of alterations assessed in this rating is considered to demonstrate increased probability that the stream is prone to frequent high velocities. The corresponding occurrence of more frequent low discharges is also expected, due to reduced base flow resulting from rapid exit of water from a watershed. Many channel alterations may also directly reduce habitat heterogeneity (Allan 2004).

Channel alteration is described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels. The first level, poor channel alteration, is defined as heavy deposits of fine material and/or extensive bar development, or recent channelization, or evidence of dredging, or greater than 80% of the banks artificially armored. The second level, marginal channel alteration, is defined as recent but moderate deposition of gravel and sand on bars and/or embankments; and/or 40% to 80% of banks artificially armored or channel lined in concrete (Mercurio et al. 1999). Conditions indicating biological degradation for the BSID analysis are *channel alteration marginal to poor* and *channel alteration poor*.

Embeddedness

Embeddedness is determined by the percentage of fine sediment surrounding gravel, cobble, and boulder particles in the streambed. Embeddedness is categorized as a percentage from 0% to 100% with low values as optimal and high values as poor. High embeddedness is a result of excessive sediment deposition (Mercurio et al. 1999).

High embeddedness suggests that sediment may interfere with feeding or reproductive processes and result in biological impairment. Although embeddedness is confounded by natural variability (e.g., Coastal Plain streams will naturally have more embeddedness than Highlands streams; Roth et al. 2005), embeddedness values higher than reference streams are indicative of anthropogenic sediment inputs from overland flow or stream channel erosion.

Embeddedness threshold values were determined by comparing the 90th percentile values among very poor, poor, fair, and good biological conditions (fish and benthic separately). Threshold values based on statistically significant relationships between biology and embeddedness were identified in Highland (50%) and Eastern Piedmont (60%) eco-regions for both benthic and fish (see Appendix A: Table A-1). Because the Coastal Plain is naturally embedded, all IBI levels' 90th percentiles were equal and relationships were insignificant. A threshold of 100% was applied reflecting the uniformity of the region. Applying these threshold values to individual sites allows the determination of the *high embeddedness* condition considered for the BSID.

Epifaunal Substrate Condition

Epifaunal Substrate is a visual observation of the abundance, variety, and stability of substrates that offer the potential for full colonization by benthic macroinvertebrates. Varied habitat types such as cobble, woody debris, aquatic vegetation, undercut banks, and other commonly productive surfaces provide valuable habitat for benthic macroinvertebrates (Mercurio et al. 1999). Like embeddedness, epifaunal substrate is confounded by natural variability (i.e., streams will naturally have more or less available productive substrate). Greater availability of productive substrate increases the potential for full colonization; conversely, less availability of productive substrate decreases or inhibits colonization by benthic macroinvertebrates (Covich et al. 1999).

Epifaunal substrate conditions are described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels: 1) poor, where stable substrate is lacking, or particles are over 75% surrounded by fine sediment and/or flocculent material; and 2) marginal, where large boulders and/or bedrock are prevalent and cobble, woody debris, or other preferred surfaces are uncommon (Mercurio et al. 1999). Conditions considered for the BSID analysis are *epifaunal substrate marginal to poor* and *epifaunal substrate poor*.

Erosion Severity

Erosion Severity represents a visual observation that the stream discharge is frequently exceeding the ability of the channel and/or floodplain to attenuate flow energy, resulting in channel instability, which in turn affects bank stability. Where such conditions are observed, flow energy is considered to have increased in frequency or intensity, accelerating channel and bank erosion (Allan et al. 2007). Increased flow energy suggested by this measure is also expected to negatively influence stream biology.

Erosion severity is described categorically as minimal, moderate, or severe. Conditions indicating biological degradation are set at moderate and severe. A level of severe indicates that a substantial amount of stream banks show severe erosion, and the stream segment exhibits high levels of instability due to erosion. A level of moderate indicates that a marginal amount of stream banks show erosion and the stream segment shows elevated levels of instability due to erosion. Conditions considered for the BSID analysis are *moderate to severe erosion present* and *severe erosion present.*

4.2.2. Indicators of Instream Habitat Conditions

MDE selected several qualitative parameters from the principal dataset that evaluate the overall physical instream habitat conditions of the watershed. The habitat parameters used in the BSID analysis are: presence of channelization, instream habitat, pool/glide/eddy quality, riffle/run quality, velocity/depth diversity, presence of concrete/gabion, and presence of beaver ponds. Each of these parameters is measured during spring and/or summer index period.

Channelization

Channelization describes a condition determined by visual observation of the presence or absence of the channelization of the stream segment and the extent of the channelization. Channelization is the human alteration of the natural stream morphology by altering the stream banks, (i.e., concrete, rip rap, and ditching). Streams are channelized to increase the efficiency of the downstream flow of water. Channelization likely inhibits heterogeneity of stream morphology needed for colonization, abundance, and diversity of fish and benthic communities (Petersen et al. 1987). The condition considered for the BSID analysis is *channelization present.*

Instream Habitat Condition

Instream Habitat is a visual rating based on the perceived value of habitat within the stream channel to the fish community. Multiple habitat types, varied particle sizes, and uneven stream bottoms provide valuable habitat for fish. High instream habitat scores are evidence of the lack of sediment deposition. Like embeddedness, instream habitat is confounded by natural variability (i.e., some streams will naturally have more or less instream habitat). Low instream habitat values can be caused by high flows that collapse undercut banks and by sediment inputs that fill pools and other fish habitats (Allan et al. 2007).

Instream habitat conditions are described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels: 1) poor, which is defined as less than 10% stable habit where lack of habitat is obvious; and 2) marginal, where there is a 10-30% mix of stable habitat but habitat availability is less than desirable (Mercurio et al. 1999). *Marginal* and/or *poor* ratings of this measure indicate excessive erosion and/or sedimentation. Conditions considered for the BSID analysis are and *instream habitat structure marginal to poor* and *instream habitat structure poor.*

Pool/Glide/Eddy Quality

Pool/Glide/Eddy (P/G/E) Quality is a visual observation and quantitative measurement of the variety and spatial complexity of slow or still water habitat and cover within a stream segment (Roth et al. 2005). Stream morphology complexity directly increases the diversity and abundance of fish species found within the stream segment. The increase in heterogeneous habitat such as a variety in depths of pools, slow moving water, and complex covers likely provide valuable habitat for fish species; conversely, a lack of heterogeneity within the P/G/E habitat decreases valuable habitat for fish species.

P/G/E quality conditions are described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels 1) poor, defined as minimal heterogeneous habitat with a max depth of 0.2 meters or absent completely; and 2) marginal, defined as <10% heterogeneous habitat with shallow areas (<0.2 m) prevalent and slow moving water areas with little cover (Mercurio et al. 1999). Conditions considered for the BSID analysis are *pool/glide/eddy quality marginal to poor* and *pool/glide/eddy quality poor*.

Riffle/Run Quality

Riffle/Run Quality is a visual observation and quantitative measurement based on the depth, complexity, and functional importance of riffle/run habitat within the stream segment (Roth et al. 2005). Like P/G/E quality, an increase of heterogeneity of riffle/run habitat within the stream segment likely increases the abundance and diversity of fish species, while a decrease in heterogeneity likely decreases abundance and diversity.

Riffle/run quality conditions are described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels: 1) poor, defined as riffle/run depths < 1 cm or riffle/run substrates concreted; and 2) marginal, defined as riffle/run depths generally 1 – 5 cm with a primarily single current velocity (Mercurio et al. 1999). Conditions considered for the BSID analysis are *riffle/run quality marginal to poor* and *riffle/run quality poor.*

Velocity Depth Diversity

Velocity/Depth Diversity is a visual observation and quantitative measurement based on the variety of velocity/depth regimes present at a site (i.e., slow-shallow, slow-deep, fast-shallow, and fast-deep; Roth et al. 2005). Like riffle/run quality, an increase in the number of different velocity/depth regimes likely increases the abundance and diversity of fish species within the stream segment and vice versa. The *marginal* or *poor* diversity categories could identify the absence of available habitat to sustain a diverse aquatic community. This measure may reflect natural conditions (e.g., bedrock), anthropogenic conditions (e.g., widened channels, dams, channel dredging, etc.), or excessive erosional conditions (e.g., bar formation, entrenchment, etc.).

Velocity/depth diversity conditions are described categorically as optimal, sub-optimal, marginal, or poor. Conditions indicating biological degradation are set at two levels: 1) poor, defined as the stream segment being dominated by one velocity/depth regime, usually pools; and 2) marginal, defined as having only two out of the four velocity/depth diversity regimes present with in the stream segment (Mercurio et al. 1999). Conditions considered for the BSID analysis are *velocity/depth diversity marginal to poor* and *velocity/depth diversity poor.*

Concrete/Gabion

The presence or absence of concrete is determined by a visual observation within the stream segment, resulting from the field description of the types of channelization. Like the parameter channelization, concrete inhibits the heterogeneity of stream morphology needed for colonization, abundance, and diversity of fish and benthic communities. Concrete channelization increases flow and provides a homogeneous substrate, conditions which are detrimental to diverse and abundant colonization (Petersen et al. 1987). The condition considered for the BSID analysis is *concrete/gabion present.*

Beaver Dam

The presence or absence of a beaver pond within the stream segment is determined from a visual observation. Beaver dams often create stream impoundments causing numerous physical and chemical changes of a free-flowing stream resulting in a more lentic environment. These impoundments create a physical barrier within the stream preventing fish migration. Natural biological response to beaver activity may appear to suggest that a stream's biological community is 'impaired' because the biotic composition differs from regional reference stations. The presence of beaver pond at a station will demonstrate the potential for natural community alteration to explain low IBI scoring. Beaver pond is categorized as a presence/absence binary data result. The condition considered for the BSID analysis is *beaver pond present.*

4.2.3. Indicators of Riparian Habitat Condition

MDE selected two parameters from the principal dataset that evaluate the overall riparian habitat conditions of the watershed. The riparian habitat parameters used in the BSID analysis are riparian buffer width and shading. Each of these parameters is measured once during summer index period.

Riparian Buffer Width

Riparian Buffer Width represents the minimum width of vegetated buffer in meters, considering both sides of the stream. Riparian buffer width is measured from 0 m to 50 m, with 0 m having no buffer and 50 m having a full buffer (Mercurio et al. 1999). Riparian buffers serve a number of critical ecological functions. They control erosion and sedimentation, modulate stream temperature, provide organic matter, and maintain benthic macroinvertebrate communities and fish assemblages (Lee et al. 2004).

Riparian buffer threshold values were evaluated by comparing the 10th percentile widths among very poor, poor, fair, and good biological conditions (fish and benthic separately). A threshold value based on statistically significant relationships between biology and shading was identified in the Coastal eco-region (5 m). Due to the abundance of sites lacking a riparian buffer in Highland and Eastern Piedmont eco-regions, the 10th percentiles calculated across all IBI levels were zero feet (see Appendix A: Table A-2). It was decided that a stream segment having no riparian buffer would indicate potential biological degradation, so a degradation threshold of 1 m was set for these eco-regions. The condition considered for the BSID analysis is *no riparian buffer.*

Shading

Shading is a metric indicating the percentage of the stream segment that is shaded, taking duration into account. Because solar radiation increases the temperature of the stream segment, causing thermal stress on fish and invertebrates, shading is important in protecting the stream from this impact. Other impacts from increased water temperature are decreased dissolved oxygen, and increased bacterial and algal growth.

Shading threshold values were determined by comparing the 10th percentile values among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between benthic biology and shading were identified in Highland (40%) and Coastal (20%) eco-regions (see Appendix A: Table A-3). Eastern Piedmont trends were insignificant, so a lower regional confidence limit was used (35%). All fish relationships were insignificant for shading. Applying these thresholds to individual sites allow the determination of the *low shading* condition considered for the BSID.

4.2.4. Indicators of Water Chemistry Conditions

MDE selected several quantitative parameters from the principal dataset that evaluate the overall water quality of a stream and provide information about nutrient and inorganic loading. The water quality parameters used in the BSID and measured once during the spring index period are total phosphorus, orthophosphate, total nitrogen, total ammonia nitrogen, pH (lab), ANC, chlorides, conductivity (lab), and sulfates. In addition, in-situ measurements used in the BSID and taken once during the summer index period are dissolved oxygen, pH (field), and conductivity (field).

Total Phosphorus

Total phosphorus (TP) is a measure of the amount of TP in the water column. Phosphorus forms the basis of a very large number of compounds, the most important class of which is the phosphates. For every form of life, phosphates play an essential role in all energy-transfer processes such as metabolism and photosynthesis. About three-quarters of the TP (in all of its chemical forms) used in the United States goes into fertilizers. Other important uses are as builders for detergents and nutrient supplements for animal feeds. Phosphorus plays a crucial role in primary production. Elevated levels of phosphorus can lead to excessive growth of filamentous algae and aquatic plants. Excessive phosphorus input can also lead to increased primary production, which potentially results in species tolerance exceedances of dissolved oxygen and pH levels. TP input to surface waters typically increases in watersheds where urban and agricultural developments are predominant (Johnes 1996).

TP threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between biology and TP were identified in all three regions for benthics, and additionally in Highland for fish (see Appendix A: Table A-4). Based on the results, thresholds were set at 0.03 mg/L for Highland, 0.05 mg/L for Eastern Piedmont, and 0.10 mg/L for Coastal. Applying these thresholds to individual sites allows the determination of the *high total phosphorus* condition considered for the BSID.

Orthophosphate

Orthophosphate (OP) is a measure of the amount of OP in the water column. OP is the only form of phosphorus that algae, bacteria, and plants can assimilate (Correll 1998). OP threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between benthic biology and OP were identified in Highland (0.01 mg/L) and Eastern Piedmont (0.025 mg/L) eco-regions (see Appendix A: Table A-5). Coastal trends were insignificant, so a regional upper confidence limit (0.035 mg/L) was used. All fish relationships were insignificant for OP. Applying the thresholds to individual sites will allow the determination of the *high orthophosphate* condition considered for the BSID.

Total Nitrogen

Total nitrogen (TN) is a measure of the amount of TN in the water column. TN is comprised of organic nitrogen, ammonia nitrogen, nitrite and nitrate. Nitrogen plays a crucial role in primary production. Elevated levels of nitrogen can lead to excessive growth of filamentous algae and aquatic plants. Excessive nitrogen input also can lead to increased primary production, which potentially results in species tolerance exceedances of dissolved oxygen and pH levels. Runoff and leaching from agricultural land can generate high instream levels of nitrogen (Johnes 1996).

TN threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between biology and shading were identified in the Highland eco-region (3.0 mg/L) for benthic and fish (see Appendix A: Table A-6). Eastern Piedmont (5.5 mg/L) trends were insignificant, and Coastal (7.0 mg/L) trends further appeared opposite, suggesting that TN is not a good indicator of IBI scores in this analysis. Regional upper confidence limits were therefore used for each. Applying the thresholds to individual sites will allow the determination of the *high total nitrogen* condition considered for the BSID.

Nitrite

Nitrite (NO_2^-) is a measure of the amount of NO_2^- in the water column. NO_2^- is an inorganic ion formed as an intermediate from ammonium (NH_4^+) to nitrate (NO_3^-) by bacteria in soil, sewage, and water. It can lead to eutrophication; can bioaccumulate in organisms, and causes biological harm to benthics and fish mainly through anoxia. Human sources that increase NO_2^- concentrations include agriculture, sewage, and some industrial processes (Lewis and Morris 1986, Doull et al. 1980).

Nitrite threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between biology and nitrite were identified in all three regions (see Appendix A: Table A-7). Based on the results, thresholds were set at 0.01 mg/L for Highland, 0.02 mg/L for Eastern Piedmont, and 0.03 mg/L for Coastal. Applying these thresholds to individual sites allows the determination of the *high nitrites* condition considered for the BSID.

Nitrate

Nitrate (NO_3^-) is a measure of the amount of NO_3^- in the water column. Nitrifying bacteria oxidize ammonium (NH_4^+) to nitrite (NO_2^-) to nitrate (NO_3^-), three inorganic forms of nitrogen. NO_3^- is highly soluble and tends to exist in greater concentrations than other inorganic forms do, even in the presence of relatively low dissolved oxygen. In addition to agriculture, sewage, and industrial sources, atmospheric deposition can be a source of NO_3^-. Like NO_2^-, it causes biological harm via anoxia. Unlike NH_4^+ and NO_2^-, however, biological uptake of NO_3^- is limited, making it less toxic (Carmago et al. 2005, Doull et al. 1980).

Nitrate threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Threshold values based on statistically significant relationships between biology and nitrate were identified in the Highland eco-region (3.0 mg/L; see Appendix A: Table A-8). However, Eastern Piedmont (6.0 mg/L) and Coastal (6.0 mg/L) trends were insignificant and opposite, suggesting that nitrate is not a good indicator of IBI scores in this analysis. Regional upper confidence limits were therefore used for each. Applying these thresholds to individual sites allows the determination of the *high nitrates* condition considered for the BSID.

Dissolved Oxygen

Dissolved Oxygen (DO) is a measure of the amount of oxygen dissolved in the water as a function of variables such as water temperature, atmospheric pressure, physical aeration, and chemical/biological oxygen demand. DO is generally reported as a concentration (mg/L). MDDNR MBSS measures DO in situ once during the summer. Low DO concentrations may indicate organic pollution due to heterotrophic oxygen consumption and may stress aquatic organisms. Low DO concentrations are considered to demonstrate excessive oxygen demand, primarily from decomposition of organic material (Allan et al. 2007). Sources are agricultural, forested, and urban land uses.

The COMAR criterion for Use I waters is that the DO concentration may not be less than 5.0 mg/L at any time. The criterion for Use III waters (Nontidal Cold Water) is that the DO concentration may not be less than 5.0 mg/L at any time, with a minimum daily average of not less than 6.0 mg/L (COMAR 2014c). Applying both thresholds of 5.0 and 6.0 mg/L to individual sites will allow the determination of the *low dissolved oxygen* condition considered for the BSID.

Dissolved Oxygen Saturation

DO saturation accounts for physical solubility limitations of oxygen in water and provides a more targeted assessment of oxygen dynamics than concentration alone. Percent saturation is relative to the amount of oxygen that water can hold, as determined by temperature and atmospheric pressure. MDDNR MBSS only measures DO concentrations expressed in mg/L; therefore, MDE calculated DO saturation percentages. Percent saturation is the ratio of observed DO to DO saturation value, expressed as a percent (Chapra 1997).

$$\mathrm{T_a} = \mathrm{temp\_fld} + 273.15$$

where temp_fld is the MDDNR MBSS recorded water temperature (^{o}C) at a specified station and T_a is absolute temperature (K).

$$\ln \mathrm{O_{sf}} = -139.34411 + \frac{1.575701 * 10^{5}}{\mathrm{T_a}} - \frac{6.642308 * 10^{7}}{T_a^{2}} + \frac{1.243800 * 10^{10}}{T_a^{3}} - \frac{8.621949 * 10^{11}}{T_a^{4}}$$

$$\mathrm{O_{sf}} = \mathrm{e}^{\ln \mathrm{O_{sf}}}$$

where O_{sf} is the saturation concentration of dissolved oxygen in fresh water at 1 atm (mg/L) and e is the irrational constant 2.7182818.

$$\mathrm{O_{sp}} = \mathrm{O_{sf}} \times (1 - .000035 \times \mathrm{altitude\_f})$$

where O_{sp} is the saturation concentration of dissolved oxygen at a specified elevation and altitude_f is the altitude, in feet, of a specified MDDNR MBSS station.

$$\mathrm{dosat\_fld} = \frac{\mathrm{do\_fld}}{\mathrm{O_{sp}}}$$

where dosat_fld is the percent DO saturation and do_fld is the MDDNR MBSS recorded DO in situ at a specified station.

Natural diurnal fluctuations can become exaggerated in streams with excessive primary production, enabling stressor risk analyses. Low DO saturation levels are considered to demonstrate high respiration associated with excessive decomposition of organic material. Additionally, high DO saturation is considered to demonstrate oxygen production associated with high levels of photosynthesis. Sources are agricultural, forested and urban land uses.

DO saturation threshold values were determined by comparing the 10th percentiles for low saturation and 90th percentiles for high saturation among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, statistically significant relationships between biology and low DO saturation were identified in all regions, for benthics and fish (see Appendix A: Table A-9). Based on the results, threshold values were set at 70% for Highland, 80% for Eastern Piedmont, and 40% for Coastal; saturation below these levels indicates biological degradation.

Second, a threshold value based on a statistically significant relationship between benthic biology and high DO saturation was identified in the Coastal eco-region (100%; see Appendix A: Table A-10). Highland and Eastern Piedmont trends were insignificant for benthic and fish, so regional upper confidence limits of 115% were used for each. Applying these thresholds to individual sites allows the determination of the *low dissolved oxygen saturation* and *high dissolved oxygen saturation* conditions considered for the BSID.

Total Ammonia Nitrogen

Total Ammonia Nitrogen (TAN) is a measure of the amount of nitrogen in ammonia (un-ionized, NH_3) and ammonium (ionized, NH_4^+) in mg/L in the water column. In freshwater, NH_3 exists in equilibrium with NH_4^+ and hydroxide (OH^-) ions. The ratio of NH_3 to NH_4^+ decreases as pH and temperature decrease (Emerson 1975). In excess, NH_3 especially has potential toxic effects on aquatic life. NH_3 toxicity is associated with increased primary production, pH, sunlight exposure, and water temperature. Sources of TAN are fish and animal excretion, breakdown of organic waste matter, some industrial and commercial processes, and increased nutrient loads from urban and agricultural development (Appl 1999, USEPA 2013).

TAN toxicity is reported in COMAR and USEPA criterion (see below list, USEPA 1999, COMAR 2014b) in four categories: TAN acute with salmonid present, TAN acute with salmonid absent, TAN chronic where fish early life stages may be present, and TAN chronic where fish early life stages are absent. Acute toxicity refers to potential exceedences of species tolerance caused by a one-time, sudden, high exposure of TAN. Chronic toxicity refers to potential exceedences of species tolerance caused by repeated exposure over a long period of time. Acute criteria are pH-dependent; chronic criteria are pH- and temperature- dependent. All four criteria are checked at each site in the BSID analysis. Concentrations above a threshold value may indicate biological degradation.

MDDNR MBSS collects water chemistry samples for TAN and pH during the spring index period. Because water temperature is not collected in the MBSS spring index period (i.e. with ammonia samples), an average water temperature was estimated for each physiographic region using data from a representative Maryland CORE/Trend station. The water temperatures applied in the three regions were 7.0º C (Highland), 7.0º C (Eastern Piedmont), and 10.0º C (Coastal Plain).

The freshwater ammonia criteria in COMAR (2014b) are calculated as follows:

1. Acute water quality criteria with salmonids present:

$$\frac{0.275}{1+10^{7.204-pH}}+\frac{39.0}{1+10^{pH-7.204}}$$

2. Acute water quality criteria with salmonids absent:

$$\frac{0.411}{1+10^{7.204-pH}}+\frac{58.4}{1+10^{pH-7.204}}$$

3. Chronic water quality criteria with early life stages present:

$$\left(\frac{0.0577}{1+10^{7.688-pH}}+\frac{2.487}{1+10^{pH-7.688}}\right)\times MIN(2.85,1.45\times10^{0.028\times(25-T)})$$

4. Chronic water quality criteria with early life stages absent:

$$\left(\frac{0.0577}{1+10^{7.688-pH}}+\frac{2.487}{1+10^{pH-7.688}}\right)\times1.45\times10^{0.028\times(25-MAX(T,7))}$$

Applying the criteria to individual sites allows the determination of the *ammonia chronic with early life stages present*, *ammonia chronic with early life stages absent*, *ammonia acute with salmonid present*, and *ammonia acute with salmonid absent* conditions considered for the BSID.

pH

pH is a measure of the acid balance of a stream and uses a logarithmic scale range from 0 to 14, with 7 being neutral. MDDNR MBSS collects pH samples once during the spring, which are analyzed in the laboratory (*pH lab*), and measured once in situ during the summer (*pH field*). Most stream organisms prefer a pH range of 6.5 to 8.5. Values of less than 6.5 for pH are considered to demonstrate acidity, which can be damaging to aquatic life. Intermittent high pH (greater than 8.5) is often associated with eutrophication related to increased algal blooms (Smith et al. 1999). Exceedances of pH may allow concentrations of toxic elements (such as ammonia, nitrite, and aluminum) and high amounts of dissolved heavy metals (such as copper and zinc) to be mobilized for uptake by aquatic plants and animals (Playle 1989).

The pH threshold values, at which levels below 6.5 and above 8.5 may indicate biological degradation, are established from state regulations (COMAR 2014c). Low stream pH results from agricultural land use, acid mine drainage, atmospheric deposition and organic sources. High stream pH results from agricultural and urban land uses. Applying the low and high thresholds to individual sites will allow the determination of the *low lab pH, high lab pH, low field pH,* and *high field pH* conditions considered for the BSID.

Acid Neutralizing Capacity

Acid Neutralizing Capacity (ANC) is a measure of the capacity of dissolved constituents in the water to react with and neutralize acids. MDDNR MBSS measures ANC in the spring and reports it in µeq/L. ANC can be used as an index of the sensitivity of surface waters to acidification. The higher the ANC, the more acid a system can assimilate before experiencing a decrease in pH. Repeated additions of acidic materials may cause ANC to decrease. ANC values less than 50 µeq/L are considered to demonstrate chronic (highly sensitive to acidification) exposures for aquatic organisms (Southerland et al. 2007).

Levels below the chronic ANC threshold value of 50 µeq/L may indicate biological degradation, and is established from peer-reviewed literature (Kazyak et al. 2005, Southerland et al. 2007). Low ANC results from agricultural land use, acid mine drainage, atmospheric deposition and organic sources. Applying the thresholds to individual sites will allow the determination of the *acid neutralizing capacity below chronic level* condition considered for the BSID.

Chlorides

Chloride is a measure of the amount of dissolved chloride (Cl^-) in the water column. MDDNR MBSS measures chlorides during the spring index period and reports it as mg/L. Chlorides can play a critical role in the elevation of conductivity (an indicator of the presence of dissolved substances). Most fish and benthic communities cannot survive in waters with high levels of chlorides. Excessive chloride concentrations indicate potential damage to stream biology.

High concentrations of chlorides can be due to several types of pollution, including industrial discharges, leaking wastewater infrastructure, metals contamination, and application of road salts in urban landscapes. Although chloride can originate from natural sources, most of the chloride that enters the environment is associated with the storage and application of road salt (Sherwood 1989). Road salt accumulation and persistence in watersheds poses risks to aquatic ecosystems and to water quality. Approximately 55% of road-salt chlorides are transported in surface runoff, with the remaining 45% infiltrating through soils and into groundwater aquifers (Church and Friesz, 1993).

Chloride values greater than the threshold value of 50 mg/L may indicate biological degradation, and is established from peer-reviewed literature (Morgan et al. 2007). Applying this threshold to individual sites allow the determination of the *high chlorides* condition considered for the BSID.

Conductivity

Conductivity is a measure of water's ability to conduct electrical current and is directly related to the total dissolved salt content of the water. MDDNR MBSS collects conductivity samples once during the spring, which is analyzed in the laboratory *(conductivity lab).*

Most of the total dissolved salts of surface waters are comprised of inorganic compounds or ions such as chloride, sulfate, carbonate, sodium, and phosphate. Stream conductivity is determined primarily by the geology of the area through which the stream flows. Streams supporting fish assemblages usually have a range between 150 and 500 µS/cm; conductivity outside this range may indicate that the water is unsuitable for certain species of fish and/or macroinvertebrates resulting a shift to more salinity-tolerant species (USEPA 2013).

Conductivity threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. A statewide threshold value based on statistically significant relationships between benthic biology and conductivity were identified in all regions, for benthic and fish (see Appendix A: Table A-11). All regions' trends supported a threshold of 300 µS/cm; concentrations above this level indicate biological degradation. Applying these thresholds to individual sites allow the determination of the *high conductivity* condition considered for the BSID.

Sulfates

Sulfate is the amount of dissolved sulfate ($SO_4^{2-)}$ in the water column. MDDNR MBSS measures sulfate once in the spring and reports it as mg/L. Sulfur is an essential plant nutrient. Sulfates can play a critical role in the elevation of conductivity. Other detrimental impacts of elevated sulfates are their ability to form strong acids, which can lead to changes of pH levels in surface waters.

Sulfate loads to surface waters can be naturally occurring or originate from urban runoff, agricultural runoff, acid mine drainage, atmospheric deposition, and wastewater dischargers. When naturally occurring, they are often the result of the breakdown of leaves that fall into a stream, of water passing through rock or soil containing gypsum and other common minerals.

Sulfate threshold values were determined by comparing the 90th percentile concentrations among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. Statistically significant relationships between biology and sulfate were identified in all regions, for benthics and fish (see Appendix A: Table A-12). Based on the results, threshold values were set at 25 mg/L for Highland and Coastal, and at 15 mg/L for Eastern Piedmont. Applying these thresholds to individual sites allow the determination of the *high sulfate* condition considered for the BSID.

4.3. Sources

In addition to the above measures and assessments of stream condition, MDE analyzed biological integrity as it relates to potential "sources" of stressors present in watersheds. Parameters representing sources of stressors are grouped into two categories: land uses within a watershed and potential sources of acidity.

Landscape data evaluated in the BSID analysis is statewide land use land cover (LULC) data from the Chesapeake Bay Program, all data is updated to 2002 and allows for a simulation period from 1984 to 2005 (USEPA 2014). Using the CBP LULC data in ArcGIS and Spatial Analyst (ESRI, 1999), MDE calculated LULC proportions for each MDDNR MBSS site at two scales: for the whole watershed area upstream of the site, and for the 60-meter riparian areas upstream of the site. Land use parameters used in the BSID analysis were grouped into four categories: agricultural, anthropogenic, impervious, and urban.

As anthropogenic disturbance increases, biological condition in our rivers and streams generally deteriorates. However, land use is broadly associated with the biological condition of aquatic systems and does not provide the specificity to isolate and identify instream stressors responsible for observed biological conditions. While not independently useful in identifying biological stressors, land use data does enhance understanding of the influence of instream chemical and physical stressors. Land uses are considered sources of many biological stressors, for example pH, ammonia, and chlorides. However, causal sources are given far less weight than instream stressors in the final interpretation of causation in the risk analyses results.

MDE also selected numerous parameters within the principal dataset that represent sources of acidity to be included as causal sources. Increased acidity within a stream, resulting in levels that exceed species tolerance, may indicate biological degradation to biological communities. Sources of acidity represent acidic conditions due to loads from land use and chemical sources, categorized as acid mine drainage, organic sources, and agricultural influences. MDDNR MBSS derived the possible sources of acidification from analyzing water chemistry data collected by the Maryland Synoptic Stream Chemistry Survey (MSSCS) and other regional data (Southerland et al. 2005a).

4.3.1. Impervious Land Use

Impervious Surface in Watershed

Impervious surface is any land area that does not permit precipitation to percolate into the ground, including natural and anthropogenic surfaces. Human development typically increases the amount of impervious surface in a watershed by replacing natural vegetation and soils with buildings and pavement. A high proportion of impervious surface will result in increased surface flow and more rapid transport of precipitation out of a watershed. Increased surface flows to streams can result in more pollutant transport that may exceed species tolerances. The increased speed of runoff also can overpower natural stream morphology formed to attenuate flow energy, such as meanders and floodplains (Allan et al. 2007). Streams destabilize as they adjust to changes in flow energy, subjecting them to rapid changes in morphology that could episodically displace aquatic organisms as habitats are gained and lost. Aquatic organisms may also be repeatedly scoured from stream channels where high flows are experienced more frequently than in watersheds with low amounts of impervious surface (Allan 2004).

Impervious surface information is obtained from the RESAC 2001 CBW Impervious Product Version 1.3.1 (Mid-Atlantic RESAC 2003). Impervious surface area was measured as a proportion at two scales: within the entire drainage basin for each sample station (watershed scale) and within 60 meters of streams upstream from sample stations (60m buffer scale).

Impervious surface land use threshold values were determined at both scales by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, statistically significant relationships between biology and impervious surface were identified in all three regions (see Appendix A: Table A-13). Based on the results, thresholds were set at 2% for Highland, 5% for Eastern Piedmont, and 15% for Coastal eco-regions. Applying these threshold values to individual sites allows the determination of the *high % of impervious surface in watershed* condition considered for the BSID.

Second, the same relationships were significant at the 60m buffer scale (see Appendix A: Table A-14). Magnitudes of tolerance for imperviousness were lower closer to streams. Based on the results, thresholds were set at 1% for Highland, 2% for Eastern Piedmont, and 6% for Coastal eco-regions. Applying these threshold values to individual sites allows the determination of the *high % of impervious surface in 60m buffer* condition considered for the BSID.

Roads in Watershed

This land use classification of roads in watersheds was created in ArcGIS software using Maryland's and surrounding states' roads data. It generally conveys the potential for increased surface runoff and transport of pollutants due to the impervious nature of roadways. Reduced flow attenuation properties of floodplains as well as rapid delivery of surface flow and pollutants are potential effects associated with high proportions of roads.

Road land use threshold values were determined by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, threshold values based on statistically significant relationships between biology and roads were identified as 2% for Highland, 4% for Eastern Piedmont, and 10% for Coastal eco-regions (see Appendix A: Table A-15). Applying these threshold values to individual sites allows the determination of the *high % of roads in watershed* condition considered for the BSID.

Second, at the 60m buffer scale, threshold values based on statistically significant relationships between biology and roads were identified as 3% for Highland and Coastal, and 5% for Eastern Piedmont eco-regions (see Appendix A: Table A-16). Applying these threshold values to individual sites allows the determination of the *high % of roads in 60m stream buffer* condition considered for the BSID.

4.3.2. Urban Land Use

High Intensity Developed Land Use

High intensity developed sources represent the proportion of highly developed land including road area at two different scales: within the entire drainage basin for each sample station (watershed scale), and within 60 meters of streams upstream from sample stations (60m buffer scale). The watershed scale conveys the total system flow energy potential and developed proportions. The 60m buffer scale demonstrates the increased potential for pollutants to enter streams due to proximity and the corresponding lack of natural buffers to filter pollutants. High proportions also demonstrate the increased potential for encroachment of urban development on floodplains, which could reduce flow attenuation properties, thereby increasing storm flow velocity and channel erosion.

As with measures of impervious surface, high intensity developed increases surface water flow, or otherwise speeds water delivery to stream channels (e.g., storm water pipes), increasing the energy of flowing water and the potential to erode soils (on terrain and in stream channels), carry pollutants, and displace organisms. Expedited transport of water from a basin decreases groundwater recharge and amplifies both high and low flow extremes. Increased pollutant transport could include nutrients, organics, and/or inorganics from residential, commercial, and/or industrial activities associated with this land use. Reduction of available heterotrophic material could also shift trophic conditions in aquatic systems to more autotrophic conditions that could also alter biological community structure (Dodds 2007).

High intensity developed land use threshold values were determined at both scales by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, threshold values based on statistically significant relationships between biology and high intensity developed land use were identified in all three eco-regions (see Appendix A: Table A-17). Based on the results, thresholds were set at 1% for Highland, 4% for Eastern Piedmont, and 3% for Coastal at the watershed scale. Applying these threshold values to individual sites allows the determination of the *high % of high intensity developed in watershed* condition considered for the BSID.

Second, the same relationships were significant at the 60m buffer scale. Highland benthic integrity was very sensitive to even small percentages of high-intensity land cover near streams (see Appendix A: Table A-18). Based on the results, thresholds were set at 0.1% for Highland, and 1% for Eastern Piedmont and Coastal in the 60m buffer bordering watershed streams. Applying these threshold values to individual sites allows the determination of the *high % of high intensity developed in 60m stream buffer* condition considered for the BSID.

Medium Intensity Developed Land Use

Medium intensity developed land use represents the proportion of highly developed land and developed open space at two scales: within the entire drainage basin for each stream station (watershed scale), and within 60 meters of streams upstream from each stream station (60m buffer scale). Medium intensity developed areas are described as multifamily residential areas and townhome areas (Claggett et al. 2013). The watershed scale conveys the total system flow energy potential and developed proportions. The 60m buffer scale demonstrates the increased potential for pollutants to enter streams due to proximity and the corresponding lack of natural buffers to filter pollutants. High proportions also demonstrate the increased potential for encroachment of urban development on floodplains, which could reduce flow attenuation properties, thereby increasing storm flow velocity and channel erosion.

Medium intensity developed land use threshold values were determined by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, statistically significant relationships between biology and medium intensity developed land use were identified in all three eco-regions (see Appendix A: Table A-19). Based on the results, thresholds were set at 1% for Highland, 5% for Eastern Piedmont, and 15% for Coastal. Applying these threshold values to individual sites allows the determination of the *high % of medium intensity developed in watershed* condition considered for the BSID.

Second, the same relationships were significant at the 60m buffer scale (see Appendix A: Table A-20). Based on the results, thresholds were set at 1% for Highland, 2% for Eastern Piedmont, and 6% for Coastal in the 60m buffer bordering watershed streams. Applying these threshold values to individual sites allows the determination of the *high % of medium intensity developed 60m stream buffer* condition considered for the BSID.

Low Intensity Developed Land Use

Low intensity developed land use represents the proportion of low intensity developed land and developed open space at two scales: within the entire drainage basin for each stream station (watershed scale), and within 60 meters of streams upstream from each stream station (60m buffer scale). Low intensity development is described as half-acre residential lots; approximately 20% of the area is impervious. Areas including ball fields and parks comprise developed open space, which is approximately 6% impervious (Claggett et al. 2013). Pervious land cover in this category is dominated by deciduous trees, evergreen trees, mixed trees/forest, or recreational grasses. Pollutant types are expected to be similar to those associated with high intensity urban. Episodic acute loads may equal the magnitude of high intensity area due, for example, to potential seasonal application of lawn fertilizers/pesticides or random illegal dumping of pollutants. However, chronic pollutant loads are expected to be less than those in high intensity settings due to the implied presence of natural vegetation associated with this land use classification.

Low intensity developed land use threshold values were determined by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, threshold values based on statistically significant relationships between biology and low intensity development were identified in all three eco-regions (see Appendix A: Table A-21). Based on the results, threshold values were set at 10% for Highland, 30% for Eastern Piedmont, and 40% for Coastal sites. Applying these threshold values to individual sites allows the determination of the *high % of low-intensity developed in watershed* condition considered for the BSID.

Second, the same relationships were significant at the 60m buffer scale (see Appendix A: Table A-22). Based on the results, thresholds were set at 5% for Highland and 30% for Eastern Piedmont and Coastal sites. Applying these threshold values to individual sites allows the determination of the *high % of low-intensity urban land 60m stream buffer* condition considered for the BSID.

Residential Developed Land Use

Residential developed land use represents the proportion of highly developed land and developed open space at two scales: within the entire drainage basin for each stream station (watershed scale), and within 60 meters of streams upstream from each stream station (60m buffer scale). Residential or suburban developed areas are described as a combination of roads, and lawns and landscaped areas planted with herbs, shrubs, and trees (Claggett et al. 2013). Residential roads include roads that have low to moderate traffic volumes and permit car access. Service roads, highway access ramps, bridges, and tunnels are excluded. The watershed scale conveys the total system flow energy potential and developed proportions. The 60m buffer scale demonstrates the increased potential for pollutants to enter streams due to proximity and the corresponding lack of natural buffers to filter pollutants. High proportions also demonstrate the increased potential for encroachment of urban development on floodplains, which could reduce flow attenuation properties, thereby increasing storm flow velocity and channel erosion.

Residential developed land use threshold values were determined at both scales by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, threshold values based on statistically significant relationships between benthic biology and residential development were identified in Highland (2%) and Eastern Piedmont (4%) eco-regions (see Appendix A: Table A-23). Fish response trends were not significant or consistent. A threshold of 4% was assigned for the Coastal region based on the benthic response, which showed a visible trend, though insignificant. Applying these thresholds to individual sites allows the determination of the *high % of residential developed in watershed* condition considered for the BSID.

Second, at the 60m buffer scale, threshold values based on statistically significant relationships between benthic biology and residential development were identified in all three eco-regions (see Appendix A: Table A-24). Based on the results, thresholds were set at 1.5% for Highland and 2.5% for Eastern Piedmont and Coastal eco-regions. Applying these thresholds to individual sites allows the determination of the *high % of residential developed in 60m buffer* condition considered for the BSID.

Rural Developed Land Use

Rural developed land use represents the proportion of highly developed land and developed open space at two scales: within the entire drainage basin for each stream station (watershed scale), and within 60 meters of streams upstream from each stream station (60m buffer scale). Rural developed areas are described as all areas outside of the urban and suburban zones (Claggett et al. 2013). The watershed scale conveys the total system flow energy potential and developed proportions. The 60m buffer scale demonstrates the increased potential for pollutants to enter streams due to proximity and the corresponding lack of natural buffers to filter pollutants. High proportions also demonstrate the increased potential for encroachment of urban development on floodplains, which could reduce flow attenuation properties, thereby increasing storm flow velocity and channel erosion.

Rural developed land use threshold values were determined at both scales by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, a threshold value based on statistically significant relationships between biology and rural development was identified in the Highland eco-region (4%; see Appendix A: Table A-25). Due to the remaining inconsistent response trends for benthic and fish, regional upper confidence limits were assigned in Eastern Piedmont (7%) and Coastal (8%) eco-regions. Applying these thresholds to individual sites allows the determination of the *high % of rural developed in watershed* condition considered for the BSID.

Second, the same relationships were significant at the 60m buffer scale. A threshold value based on statistically significant relationships between biology and rural development was identified in the Highland eco-region (4%; see Appendix A: Table A-26). Regional upper confidence limits were assigned in Eastern Piedmont and Coastal eco-regions (4% each). Applying these threshold values to individual sites allows the determination of the *high % of rural developed in 60m stream buffer* condition considered for the BSID.

4.3.3. Agricultural Land Use

Agricultural Land

Agricultural land represents the proportion of land area used for pasture/hay and for row crops at two scales: within the drainage basin upstream of sample stations, and within 60 meters of streams upstream from sample stations. Possible stream consequences to large proportions of agricultural land may include increased loads of sediment, nutrients, and/or pesticides. This is an extremely variable land use classification that could represent conditions ranging from dense livestock feeding lots to broad hay fields with no exposed soils.

Agricultural land use threshold values were determined at both scales by comparing the 90th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, one threshold value based on a statistically significant relationship between benthic biology and agricultural land was identified in the Highland eco-region (55%; see Appendix A: Table A-27). In Eastern Piedmont (75%) and Coastal (90%) eco-regions, the relationships were insignificant and opposite, suggesting that agriculture is not a good indicator of IBI scores in this analysis. Regional upper confidence limits were therefore used as thresholds for each. Applying the threshold to individual sites will allow the determination of the *high % of agriculture in watershed* condition considered for the BSID.

Second, one threshold value based on statistically significant relationships between biology and agricultural land at the 60m buffer scale was identified in the Highland eco-region (50%; see Appendix A: Table A-28). As with the watershed scale, Eastern Piedmont (60%) and Highland (70%) eco-regions output insignificant and opposite relationships, suggesting that this is not a good indicator of IBI scores in this analysis. Regional upper confidence limits were therefore used for each. Applying these thresholds to individual sites allows the determination of the *high % of agriculture in 60m stream buffer* condition considered for the BSID.

4.3.4. Anthropogenic Land Use

Forest

Forested land is measured at two different scales: within a sample site's drainage basin (watershed scale) and within 60 meters of streams upstream from sample sites (60m buffer scale). The amount of forested land reveals the general extent of urban and agricultural development. Forested land use describes natural areas dominated by tree cover with an understory of natural plant material or ground cover. Due to processes such as evaporation, water uptake, and transpiration, watersheds with high forest proportions demonstrate natural hydrological regimes. High forest proportions also suggest that erosion will be limited due to canopies that reduce the impact of heavy rain events, along with roots and leaf litter that secure soils from transport in any overland water flow. Due to the retention of precipitation by living vegetation and leaf litter, less surface water flow means less chance for transport of pollutants (e.g., nutrients, organic, and inorganic contaminants). High forest proportion also suggests that heterotrophic material will be in abundance, and that autochthonous production will be minimal due to the presence of canopies over small water bodies. Thus, decreased amounts of forested land use will affect hydrological regimes, nutrient loads, trophic conditions, and inorganic pollutant contaminants on surface waters.

Forested land use threshold values were determined at both scales by comparing the 10th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, all watershed-scale relationships between biology and forested land use were statistically significant in all eco-regions (see Appendix A: Table A-29). Threshold values indicating levels below which biology is likely to be degraded were 20% for Highland, 15% for Eastern Piedmont, and 5% for Coastal. Applying these thresholds value to individual sites allows the determination of the *low % of forest in watershed* condition considered for the BSID.

Second, all relationships between biology and forested land use were also statistically significant at the 60m buffer scale (see Appendix A: Table A-30). Threshold values indicating levels below which biology is likely to be degraded were 20% for Highland, 30% for Eastern Piedmont, and 10% for Coastal. Applying these threshold values to individual sites allows the determination of the *low % of forest in 60m stream buffer* condition considered for the BSID.

Wetland

Wetland land use represents the proportion of wetlands at two scales: within the entire drainage basin for each stream station (watershed scale), and within 60 meters of streams upstream from each stream station (60m buffer scale). Wetlands are transitional areas between land and water, and are designated as such based on soils and vegetation in which the substrate is periodically saturated with or covered with water (Irani and Claggett 2010). The watershed scale conveys the total system flow energy potential proportions. The 60m buffer scale demonstrates the increased potential for pollutants to enter streams due to proximity and the corresponding lack of natural buffers to filter pollutants. Low proportions also demonstrate the increased potential for encroachment of urban development pollutants to streams, and increase storm flow velocity and erosion.

Wetland threshold values were determined at both scales by comparing the 10th percentile coverages among very poor, poor, fair, and good biological conditions (fish and benthic separately) in each eco-region. First, at the watershed scale, threshold values based on statistically significant relationships between biology and wetland were identified in Eastern Piedmont (0.1%) and most clearly in the Coastal (0.7%) eco-regions (see Appendix A: Table A-31). Though all 10th percentiles were low, the mountainous Highland region inherently has the least land classified as wetland and had no measured biological response. Highland was therefore assigned a threshold of zero. Applying these threshold values to individual sites allows the determination of the *low % of wetland in watershed* condition considered for the BSID.

Second, at the 60m buffer scale, a threshold value based on statistically significant relationships between biology and wetland was identified in the Coastal eco-region (1%; see Appendix A: Table A-32). Since Highland and Eastern Piedmont comparisons showed no measured response, they were each assigned thresholds of zero. Therefore, percentages less than 1% in the Coastal region may indicate biological degradation. Applying these threshold values to individual sites allows the determination of the *low % of wetland in 60m buffer* condition considered for the BSID.

4.3.5. Sources of Acidity - Acid Mine Drainage

Acid mine drainage (AMD) results from the oxidation of the mineral pyrite, which is found in mine spoils and abandoned mine shafts, and is known to cause extreme acidification of surface waters as well as affect stream physical substrate. Streams strongly affected by AMD exhibit high levels of sulfate, manganese, iron, aluminum, and conductivity. Highly acidic waters (pH < 3) can solubilize heavy metals and other toxic elements from soil and cause them to be transported into nearby surface waters. The high acidity of acid mine drainage and the high amounts of dissolved heavy metals (such as copper and zinc) generally make acid mine drainage extremely toxic to most organisms (Penreath 1994). AMD reflects a binary response (yes/no) for presence in a watershed and is contained in the principal dataset. Site samples are marked as affected by AMD if their sulfur concentrations exceed 500 µeq/L, their ANCs are less than 200 µeq/L, and they are in the North Branch Potomac River or Youghiogheny River 6-digit Maryland watershed (Roth et al. 1999). The condition considered for the BSID analysis is *AMD present.*

4.3.6. Sources of Acidity – Organic Acid Source

Natural decay of organic materials may contribute acidity in the form of organic anions, as in blackwater streams associated with bald cypress wetlands and boreal bogs. Streams dominated by organic sources are often characterized by high concentrations of dissolved organic carbon and organic anions. Organic acid source reflects a binary response (yes/no) for presence in a watershed and is contained in the principal dataset. It is marked as present if a site sample's ANC is less than 200 µeq/L and either: (1) organic ions (as a function of DOC and pH) dominate over sulfate and nitrate, or (2) DOC exceeds 8 mg/L (Roth et al. 1999; Oliver et al. 1983). The condition considered for the BSID analysis is *organic acid source present.*

4.3.7. Sources of Acidity – Agricultural Acid Source

Agricultural lands fertilized with high levels of nitrogen or other acidifying compounds are a source of acidification in surface waters. Agricultural activities in watersheds affect stream chemistry, adding both ANC from soil liming practices and strong acid anions from nitrogen fertilizers. Agricultural acid source reflects a binary response (yes/no) for presence in a watershed and is contained in the principal dataset. It is marked as present for sites with ANCs less than 200 µeq/L, nitrate concentrations greater than 100 µeq/L, and at least 50% agricultural land use (Roth et al. 1999). The condition considered for the BSID analysis is *agricultural acid source present.*

5. Statistical Methods

MDE has adopted a case-control, risk-based approach to evaluate associations between aquatic communities' biological integrity and various potential stressors. Odds ratios and attributable risks were used to objectively identify the most likely stressor(s) of biological condition in each watershed.

5.1. Biological and Stressor Components

The BSID is a two-way comparison of biology to stressors. First, the aquatic biology component is based on fish and benthic macroinvertebrate community scores, which are used to categorize each site sample as a case or a control. Cases are defined as sites with poor to very poor biological conditions within watersheds (Southerland et al. 2005b). Since MDE aims to protect both fish and benthic aquatic life, sites with either their fish IBI or benthic IBI less than 3 are categorized as cases. Conversely, controls are sites with fair to good biological conditions within the eco-region. If a site's IBIs are greater than or equal to 3, the site is a control. Unlike the cases, the controls are grouped within Maryland's three physiographic eco-regions: Highland, Eastern Piedmont, and Coastal Plain (Southerland et al. 2005b). Summarizing controls over this larger area means more robust results due to considerably larger sample sizes, while maintaining comparability within the control groups, or strata.

For habitat and sediment parameters, each of these three strata control groups was further subdivided into first order streams and second through fourth order streams. The rationale for this was that the extent or quality of habitat can vary naturally with stream order, and it is more appropriate to compare streams of similar size. This additional strata division resulted in a similar number of control sites per stratum. Also, due to sample size limitations, the second through fourth order streams were not subdivided into smaller groups.

Stressors are the second component of the comparison. As described in section 4.1 above, each parameter was assigned one stressor threshold per eco-region using existing guidelines when they were available, or using statistical analysis on grouped responses if they were not. Sequentially for each stressor, all site samples were then categorized into beyond-threshold or within-threshold stressor categories. This stressor component (beyond/within threshold) and the biological component (case/control) were then combined using odds ratios to quantify associations within each watershed.

5.2. Odds Ratios

The BSID analysis tests for the strength of association between stressor and biological components by determining whether there is an increased risk associated with the stressor surpassing the threshold. More specifically, the assessment compares the likelihood that biological condition is degraded, given that a stressor is beyond its threshold, by using the ratio of the prevalence within the case group as compared to the prevalence in the control group. These groupings and calculations are performed in SAS and R software (SAS Institute 2002-2010; R Core Team 2012; Canty and Ripley 2014, and Dorai-Raj 2009).

Calculation of odds ratios begins with the two-way contingency table setup. Commonly used in the field of epidemiology, two-way contingency tables report the frequency of cases and controls, as well as stressor levels beyond and within thresholds, for each assessment unit (Table *2*). The cells are populated with the number of site samples within each category, for each assessment unit.

Table 2. Layout of a Two-way Contingency Table.

	Cases (Sites with very poor to poor biological communities in watershed)	Controls (Sites with fair to good biological communities in strata)	Total
Stressor/Source Beyond Threshold	a	b	m_1
Stressor/Source Within Threshold	c	d	m_0
Total	n_1	n_0	n

Where

a = # of case sites with stressor/source beyond threshold
b = # of control sites with stressor/source beyond threshold
c = # of case sites with stressor/source within threshold
d = # of control sites with stressor/source within threshold
n_1 = Total # of cases
n_0 = Total # of controls
m_1 = Total # of sites with stressor beyond threshold
m_0 = Total # of sites with stressor within threshold
n = Total # of sites

The counts within these tables are then used to evaluate the strength of association using the odds ratio. The odds ratio is calculated as:

$$\text{Odds Ratio} = \frac{a}{b} \Big/ \frac{c}{d}\text{, which is equivalent to } \frac{a}{c} \Big/ \frac{b}{d} \text{ and } \frac{ad}{cb}$$

When a watershed's case sites span multiple geographic strata, it is important to compare each case with controls from its appropriate stratum. In this scenario, a common odds ratio is calculated by first developing a separate 2x2 table for each stratum, then combining these separate stratum tables using the Mantel-Haenszel (MH) approach. The MH odds ratio is calculated as follows:

$$\text{Odds Ratio}_{\text{MH}} = \sum_{g=1}^{G} \frac{a_g d_g}{n_g} \Bigg/ \sum_{g=1}^{G} \frac{b_g c_g}{n_g}$$

Where

Odds Ratio$_{\text{MH}}$ = the Mantel-Haenszel common odds ratio
g = identifier used to denote the stratum
G = the total number of strata (6 for habitat and sediment stressors; 3 for all others)

The common odds ratio confidence interval was calculated to determine if the odds ratio was significantly greater than one, within a 90% confidence level. The confidence limits were estimated using the MH (1959) approach and based on the exact method due to small sample sizes for cases. A common odds ratio confidence interval greater than one indicates that there is a statistically significant higher likelihood that a stream's biological conditions will be poor to very poor (case) when a stressor is beyond the threshold than when a stressor is within the threshold (Szklo and Nieto 2007). This statistically significant, positive association between the stressor and poor to very poor biological conditions is the measure used to identify potential stressors.

5.3. Attributable Risks

Once potential stressors are identified (odds ratios significantly greater than one), the risk attributable to each identified stressor is quantified for all case sites in the watershed. The attributable risk (AR) is defined herein as the excess likelihood of beyond-threshold stressor levels at sites with poor to very poor biological conditions. The AR is calculated as the difference between the proportion of case sites with the stressor beyond threshold and the proportion of control sites with the stressor beyond threshold (Levin and Bertell 1978). The equation is as follows.

$$AR = R_{cases} - R_{controls}$$

Where

AR = attributable risk
R_{cases} = absolute risk (percent) of case sites with stressor above threshold
$R_{controls}$ = absolute risk (percent) of control sites with stressor above threshold

When multiple strata are present and the data are from a case control study, Bruzzi et al. (1985) stated that the AR can be estimated using the cases alone once the relative risk is known. Instead of using the relative risk, it is possible to sum the AR for each case over all the cases. The assumption is that each case site has its own absolute risk. If the stressor is beyond the threshold, the absolute risk is unity, whereas if the stressor is within the threshold, the absolute risk is zero. The absolute risk of the stressor among the controls, for the specific case site, is determined based on the physiographic region of the case site and includes stream order if the stressor is related to habitat or sediment condition. The following equation is used to determine the AR of a stressor when considering multiple strata:

$$AR = \frac{\sum_{g=1}^{G} \sum_{i=1}^{n_{ig}} \left[Rcase_{ig} - Rcontrols_{g} \right]}{G \cdot n_g}$$

Where

AR = Attributable risk of a stressor for a population of sites within a watershed
$Rcase_{ig}$ = absolute risk of stressor for case *i* in stratum *g* (0 or 1)
$Rcontrols_g$ = absolute risk of stressor among controls for stratum *g*
G = total number of strata
n_g = number of cases within stratum *g*
$G \cdot n_g$ = total number of cases

Once the AR is defined for each possible stressor, the AR for groups of stressors is calculated. Similar to the AR calculation for each stressor, the AR calculation for a group of stressors is also summed over the case sites using the individual site characteristics (i.e., stressors present at that site). The only difference is that the absolute risk for the controls at each site is estimated based on the stressor present at the site that has the lowest absolute risk among the controls. For example, if high embeddedness and poor epifaunal substrate were present at the site and the absolute risk among the controls were 0.25 and 0.15 respectively, then a value of 0.15 would be used since it has the lowest risk among the controls and would produce the highest AR. The equation for estimating AR for groups of stressors is as follows:

$$AR_{group} = \frac{\sum_{g=1}^{G} \sum_{i=1}^{n_{ig}} \max_j \left[Rcase_{jig} - Rcontrols_{jg} \right]}{G \cdot n_g}$$

Where

AR_{group} = Attributable risk of a group of stressors for a population of sites within a watershed
$Rcase_{jig}$ = absolute risk of stressor j for case i in stratum g (0 or 1)
$Rcontrols_{jg}$ = absolute risk of stressor j among controls for stratum g
G = total number of strata
n_g = number of cases within stratum g
$G \cdot n_g$ = total number of cases

After determining the AR for each stressor and the AR for groups of stressors, the AR for all potential stressors is calculated. This value represents the excess prevalence of all potential stressors in cases, sites in the watershed with poor to very poor biological conditions, beyond the prevalence of stressors in controls. The purpose of this metric is to determine whether stressors have been identified for an acceptable proportion of cases. While there is not a reported acceptable value for this metric, it is recommended that a limit be selected based on the number of cases in the watershed and consideration for the biological listing methodology.

To assist in determining potential sources of the stressors, the above described statistical methods are also applied to all source parameters (e.g. land use, acid sources, etc.).

6. Conclusion

The BSID process evaluates each biologically impaired watershed to determine potential stressors and sources. Interpretation of the BSID analysis results is based upon components of Hill's Postulates (1965), which propose a set of standards that could be used to judge when an association might be causal. The components applied are: 1) the strength of association which is assessed using the odds ratio; 2) the specificity of the association for a specific stressor (risk among controls); 3) the presence of a biological gradient; 4) ecological plausibility which is illustrated through final causal models; and 5) experimental evidence gathered through literature reviews to help support the causal linkage.

The BSID process uses general causal scenarios to aid in the interpretation of how land-use conditions might generate instream stressors, and how the resulting impacts can alter the biological community and structure. Appendix B contains four general causal scenario models MDE uses to aid in the interpretation of results. With the general understanding of ecological processes within casual scenarios, knowledge of impaired watersheds, and results from the BSID analysis, MDE can determine possible causes of degraded biological conditions.

Ecologically plausible causal models will be developed specifically for each watershed based on BSID analysis results. Once the BSID analysis is completed and a final causal model is developed, a number of stressors (pollutants) may be identified as the cause of the poor to very poor biological condition within the Maryland 8-digit watershed. If there are multiple stressors (pollutants) then the process will evaluate the AR for each stressor and rank them appropriately.

Finally, water quality limited segments with degraded biological condition linked to specific stressor(s) (e.g., sediment, nutrients) are compared to the current Integrated Report listing categories for the 8-digit watershed. The BSID analysis results can be used together with a variety of water quality analyses to update and/or support the probable stressors and sources of biological impairment in the Integrated Report.

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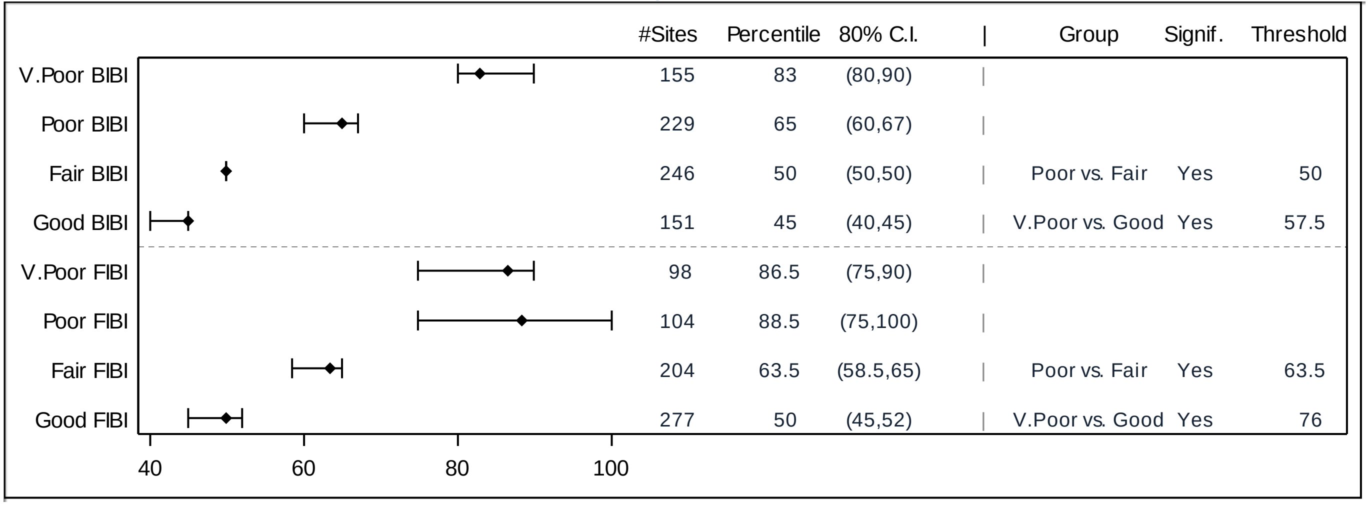
Appendix A

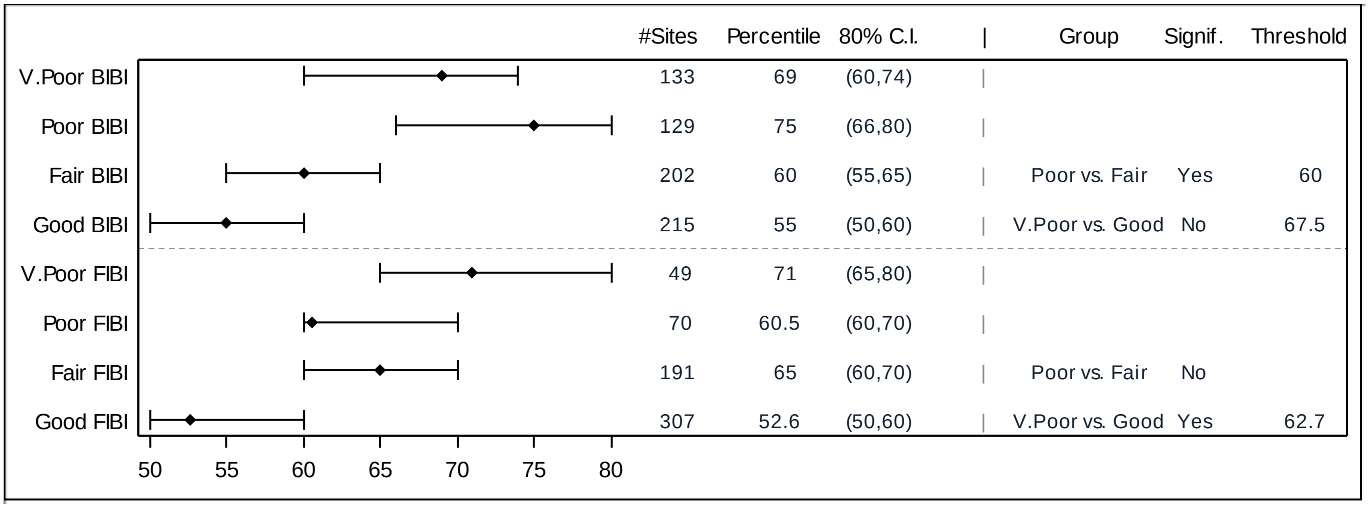
Table A-1. Physiographic Eco-region Analysis for High Embeddedness

Highland



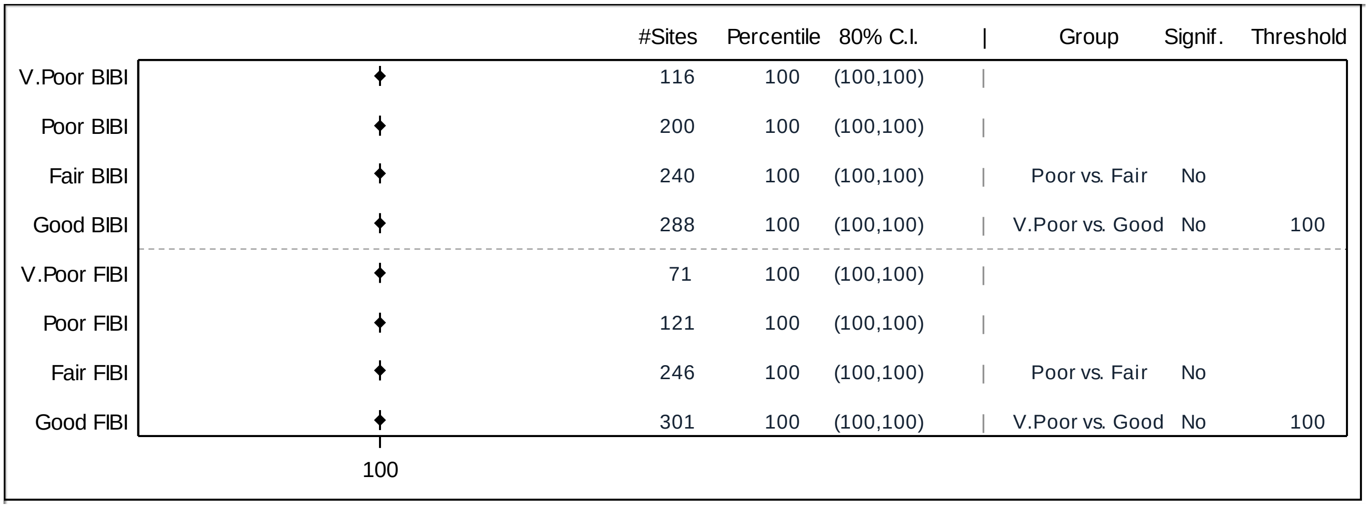
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	155	83	(80,90)			
Poor BIBI	229	65	(60,67)			
Fair BIBI	246	50	(50,50)	Poor vs. Fair	Yes	50
Good BIBI	151	45	(40,45)	V.Poor vs. Good	Yes	57.5
V.Poor FIBI	98	86.5	(75,90)			
Poor FIBI	104	88.5	(75,100)			
Fair FIBI	204	63.5	(58.5,65)	Poor vs. Fair	Yes	63.5
Good FIBI	277	50	(45,52)	V.Poor vs. Good	Yes	76

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	69	(60,74)			
Poor BIBI	129	75	(66,80)			
Fair BIBI	202	60	(55,65)	Poor vs. Fair	Yes	60
Good BIBI	215	55	(50,60)	V.Poor vs. Good	No	67.5
V.Poor FIBI	49	71	(65,80)			
Poor FIBI	70	60.5	(60,70)			
Fair FIBI	191	65	(60,70)	Poor vs. Fair	No	
Good FIBI	307	52.6	(50,60)	V.Poor vs. Good	Yes	62.7

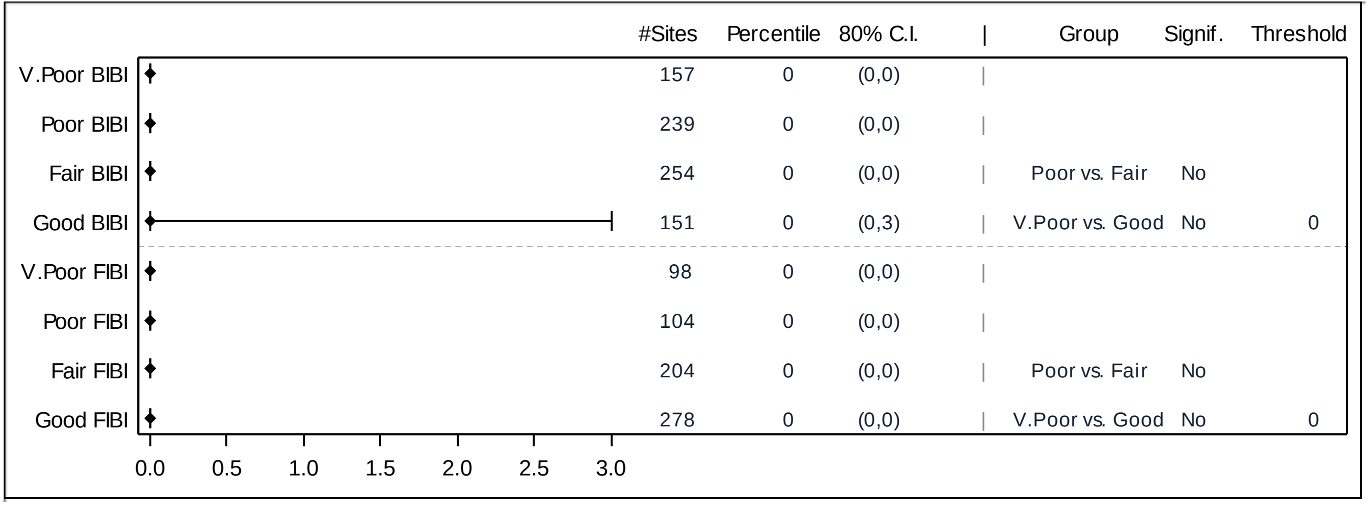
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	116	100	(100,100)			
Poor BIBI	200	100	(100,100)			
Fair BIBI	240	100	(100,100)	Poor vs. Fair	No	
Good BIBI	288	100	(100,100)	V.Poor vs. Good	No	100
V.Poor FIBI	71	100	(100,100)			
Poor FIBI	121	100	(100,100)			
Fair FIBI	246	100	(100,100)	Poor vs. Fair	No	
Good FIBI	301	100	(100,100)	V.Poor vs. Good	No	100

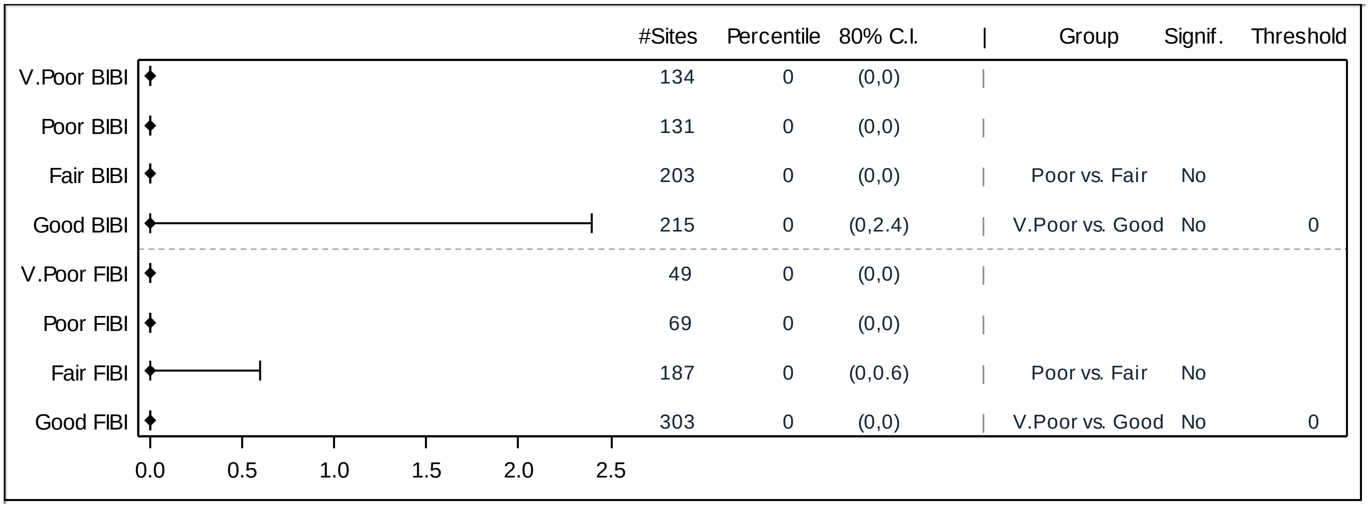
Table A-2. Physiographic Eco-region Analysis for No Riparian Buffer

Highland



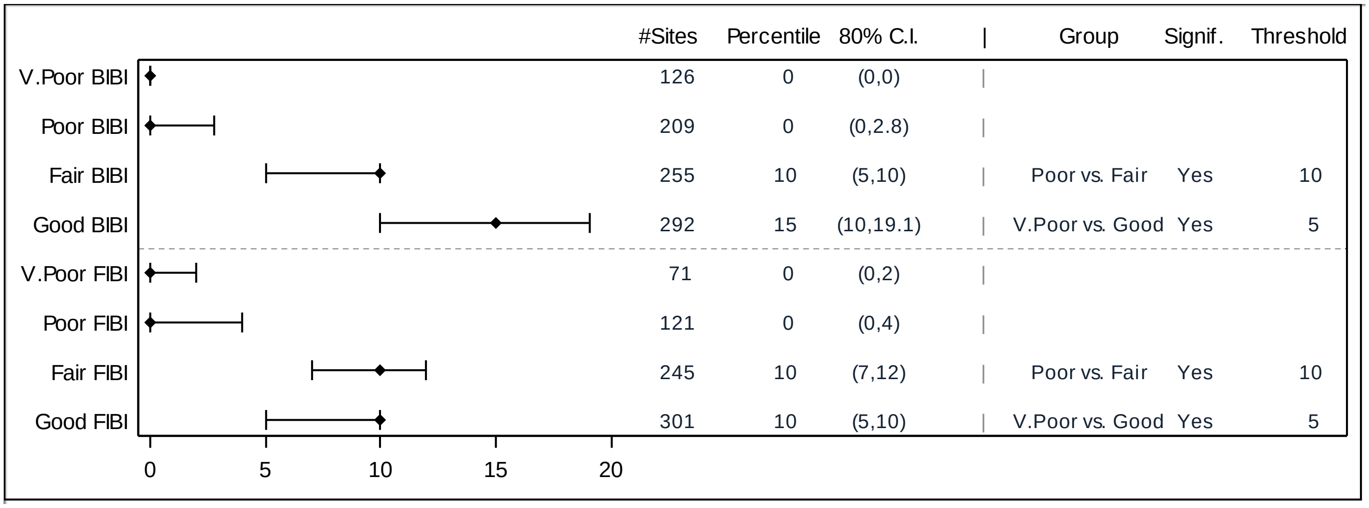
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	157	0	(0,0)			
Poor BIBI	239	0	(0,0)			
Fair BIBI	254	0	(0,0)	Poor vs. Fair	No	
Good BIBI	151	0	(0,3)	V.Poor vs. Good	No	0
V.Poor FIBI	98	0	(0,0)			
Poor FIBI	104	0	(0,0)			
Fair FIBI	204	0	(0,0)	Poor vs. Fair	No	
Good FIBI	278	0	(0,0)	V.Poor vs. Good	No	0

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0	(0,0)			
Poor BIBI	131	0	(0,0)			
Fair BIBI	203	0	(0,0)	Poor vs. Fair	No	
Good BIBI	215	0	(0,2.4)	V.Poor vs. Good	No	0
V.Poor FIBI	49	0	(0,0)			
Poor FIBI	69	0	(0,0)			
Fair FIBI	187	0	(0,0.6)	Poor vs. Fair	No	
Good FIBI	303	0	(0,0)	V.Poor vs. Good	No	0

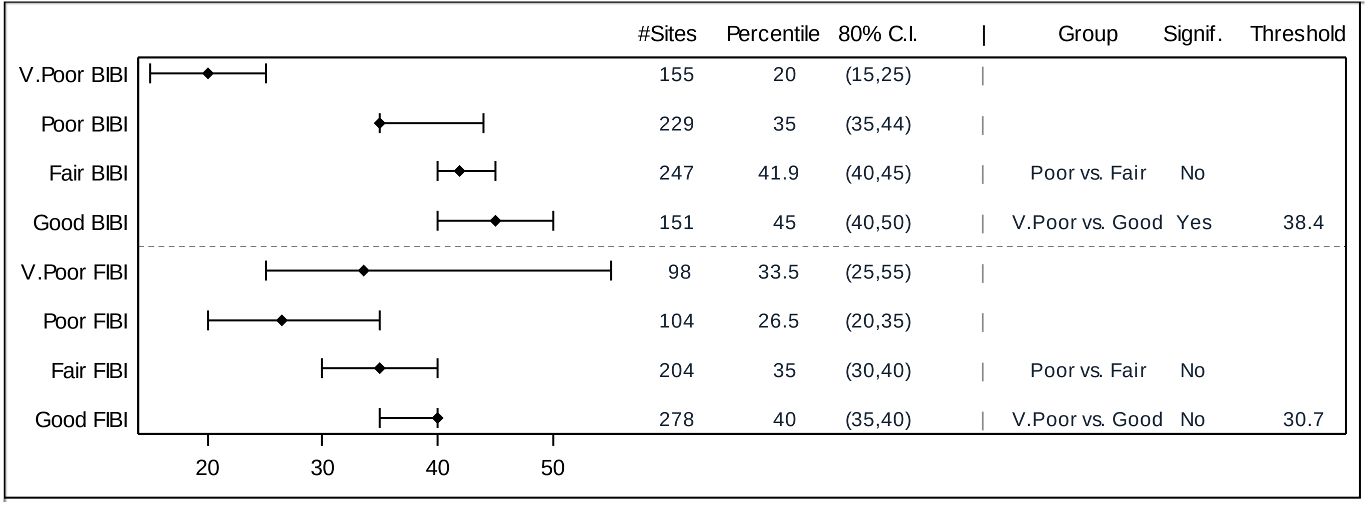
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	126	0	(0,0)			
Poor BIBI	209	0	(0,2.8)			
Fair BIBI	255	10	(5,10)	Poor vs. Fair	Yes	10
Good BIBI	292	15	(10,19.1)	V.Poor vs. Good	Yes	5
V.Poor FIBI	71	0	(0,2)			
Poor FIBI	121	0	(0,4)			
Fair FIBI	245	10	(7,12)	Poor vs. Fair	Yes	10
Good FIBI	301	10	(5,10)	V.Poor vs. Good	Yes	5

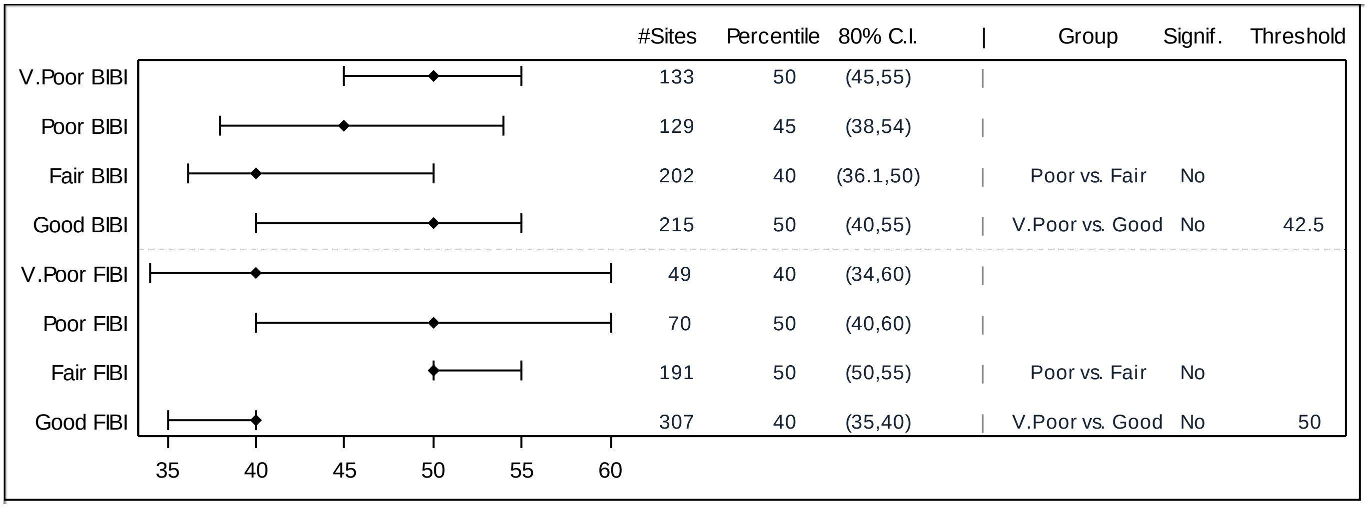
Table A-3. Physiographic Eco-region Analysis for Low Shading

Highland



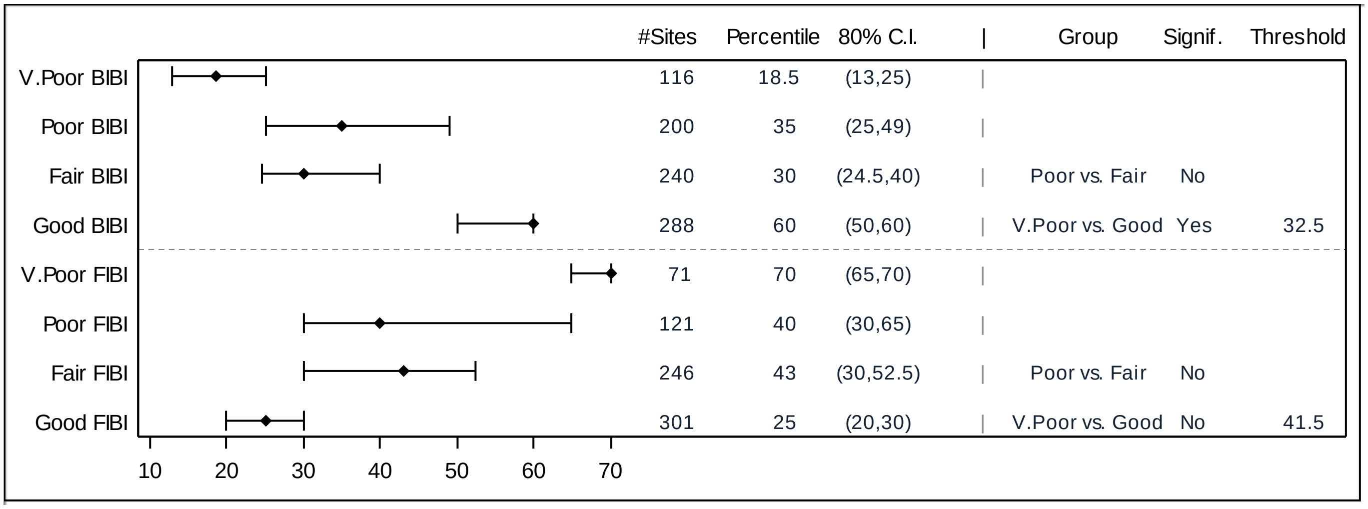
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	155	20	(15,25)			
Poor BIBI	229	35	(35,44)			
Fair BIBI	247	41.9	(40,45)	Poor vs. Fair	No	
Good BIBI	151	45	(40,50)	V.Poor vs. Good	Yes	38.4
V.Poor FIBI	98	33.5	(25,55)			
Poor FIBI	104	26.5	(20,35)			
Fair FIBI	204	35	(30,40)	Poor vs. Fair	No	
Good FIBI	278	40	(35,40)	V.Poor vs. Good	No	30.7

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	50	(45,55)			
Poor BIBI	129	45	(38,54)			
Fair BIBI	202	40	(36.1,50)	Poor vs. Fair	No	
Good BIBI	215	50	(40,55)	V.Poor vs. Good	No	42.5
V.Poor FIBI	49	40	(34,60)			
Poor FIBI	70	50	(40,60)			
Fair FIBI	191	50	(50,55)	Poor vs. Fair	No	
Good FIBI	307	40	(35,40)	V.Poor vs. Good	No	50

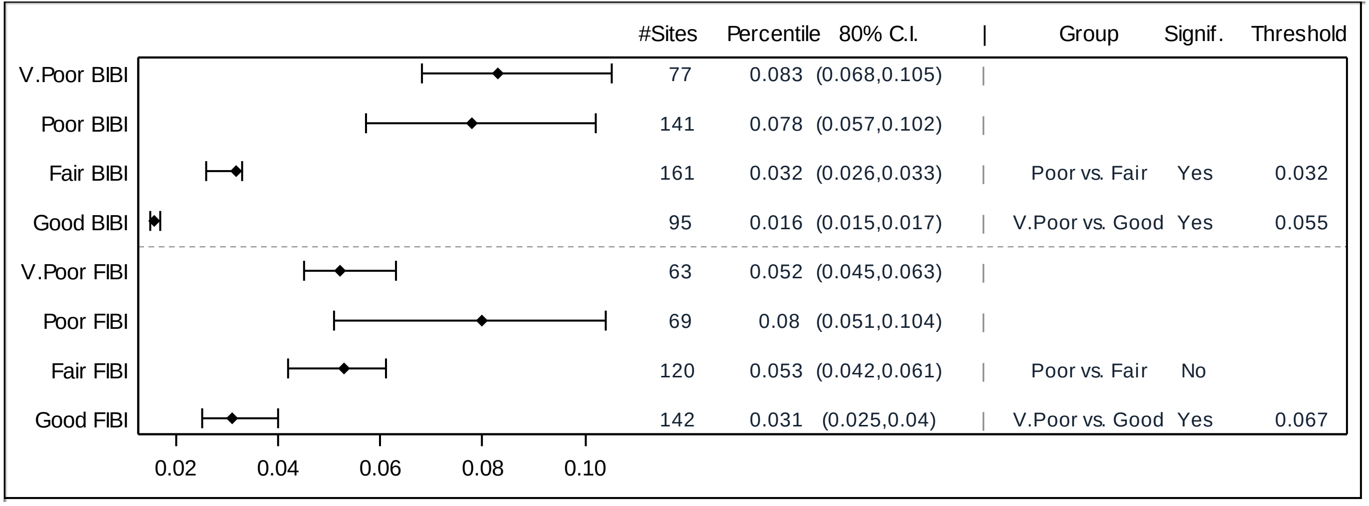
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	116	18.5	(13,25)			
Poor BIBI	200	35	(25,49)			
Fair BIBI	240	30	(24.5,40)	Poor vs. Fair	No	
Good BIBI	288	60	(50,60)	V.Poor vs. Good	Yes	32.5
V.Poor FIBI	71	70	(65,70)			
Poor FIBI	121	40	(30,65)			
Fair FIBI	246	43	(30,52.5)	Poor vs. Fair	No	
Good FIBI	301	25	(20,30)	V.Poor vs. Good	No	41.5

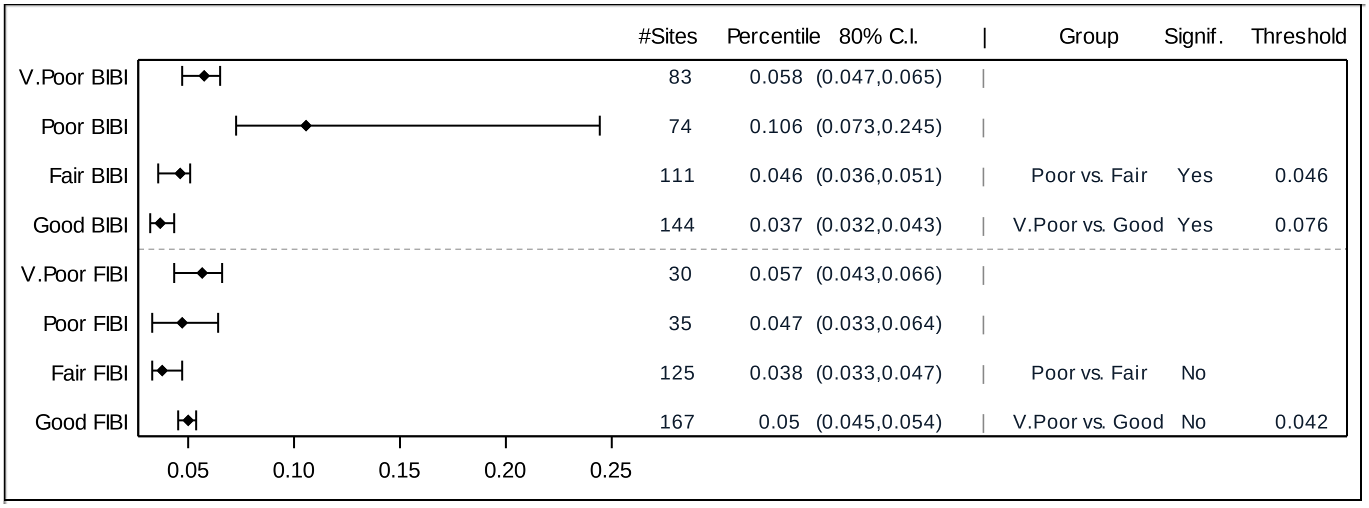
Table A-4. Physiographic Eco-region Analysis for High Total Phosphorus

Highland



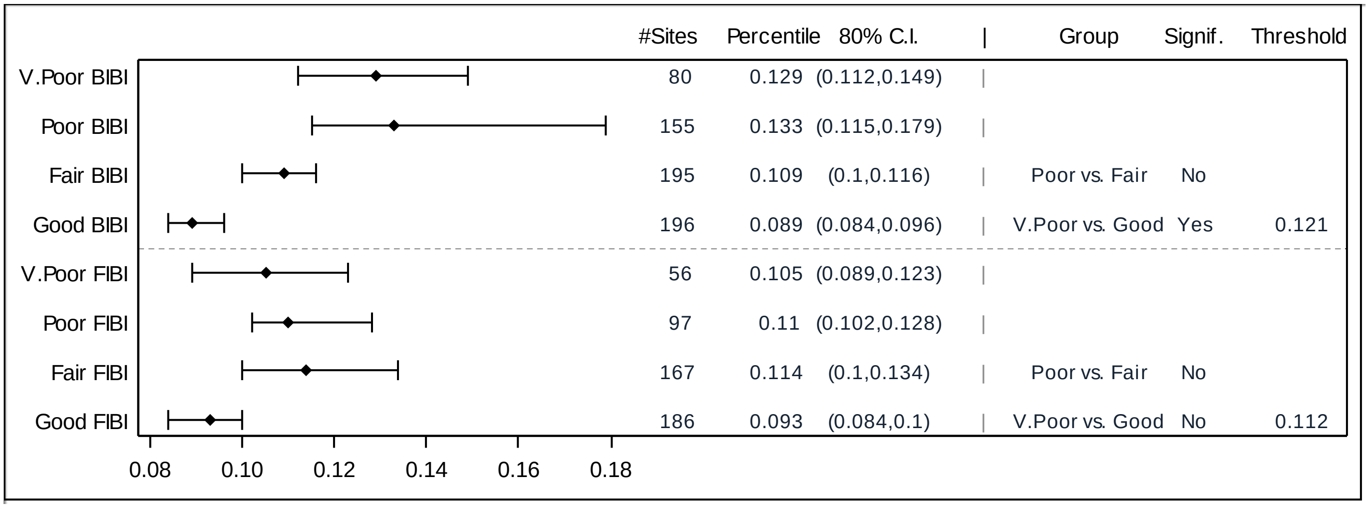
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	77	0.083	(0.068,0.105)			
Poor BIBI	141	0.078	(0.057,0.102)			
Fair BIBI	161	0.032	(0.026,0.033)	Poor vs. Fair	Yes	0.032
Good BIBI	95	0.016	(0.015,0.017)	V.Poor vs. Good	Yes	0.055
V.Poor FIBI	63	0.052	(0.045,0.063)			
Poor FIBI	69	0.08	(0.051,0.104)			
Fair FIBI	120	0.053	(0.042,0.061)	Poor vs. Fair	No	
Good FIBI	142	0.031	(0.025,0.04)	V.Poor vs. Good	Yes	0.067

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	83	0.058	(0.047,0.065)			
Poor BIBI	74	0.106	(0.073,0.245)			
Fair BIBI	111	0.046	(0.036,0.051)	Poor vs. Fair	Yes	0.046
Good BIBI	144	0.037	(0.032,0.043)	V.Poor vs. Good	Yes	0.076
V.Poor FIBI	30	0.057	(0.043,0.066)			
Poor FIBI	35	0.047	(0.033,0.064)			
Fair FIBI	125	0.038	(0.033,0.047)	Poor vs. Fair	No	
Good FIBI	167	0.05	(0.045,0.054)	V.Poor vs. Good	No	0.042

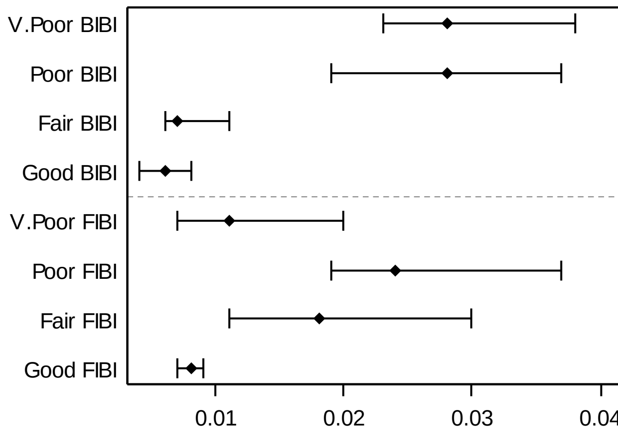
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	80	0.129	(0.112,0.149)			
Poor BIBI	155	0.133	(0.115,0.179)			
Fair BIBI	195	0.109	(0.1,0.116)	Poor vs. Fair	No	
Good BIBI	196	0.089	(0.084,0.096)	V.Poor vs. Good	Yes	0.121
V.Poor FIBI	56	0.105	(0.089,0.123)			
Poor FIBI	97	0.11	(0.102,0.128)			
Fair FIBI	167	0.114	(0.1,0.134)	Poor vs. Fair	No	
Good FIBI	186	0.093	(0.084,0.1)	V.Poor vs. Good	No	0.112

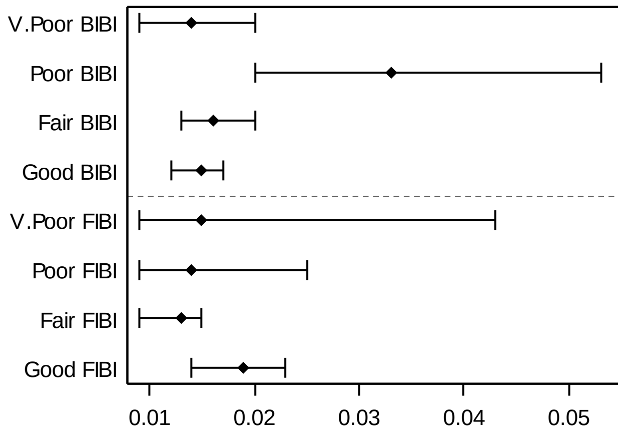
Table A-5. Physiographic Eco-region Analysis for High Orthophosphate

Highland



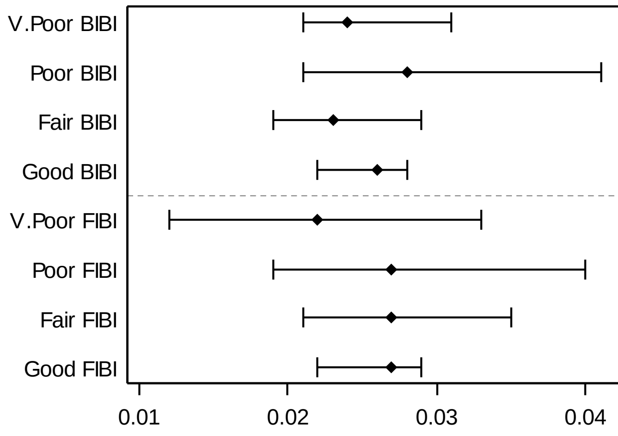
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	77	0.028	(0.023,0.038)			
Poor BIBI	141	0.028	(0.019,0.037)			
Fair BIBI	161	0.007	(0.006,0.011)	Poor vs. Fair	Yes	0.007
Good BIBI	95	0.006	(0.004,0.008)	V.Poor vs. Good	Yes	0.018
V.Poor FIBI	63	0.011	(0.007,0.02)			
Poor FIBI	69	0.024	(0.019,0.037)			
Fair FIBI	120	0.018	(0.011,0.03)	Poor vs. Fair	No	
Good FIBI	142	0.008	(0.007,0.009)	V.Poor vs. Good	No	0.021

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	83	0.014	(0.009,0.02)			
Poor BIBI	74	0.033	(0.02,0.053)			
Fair BIBI	111	0.016	(0.013,0.02)	Poor vs. Fair	Yes	0.016
Good BIBI	144	0.015	(0.012,0.017)	V.Poor vs. Good	No	0.024
V.Poor FIBI	30	0.015	(0.009,0.043)			
Poor FIBI	35	0.014	(0.009,0.025)			
Fair FIBI	125	0.013	(0.009,0.015)	Poor vs. Fair	No	
Good FIBI	167	0.019	(0.014,0.023)	V.Poor vs. Good	No	0.013

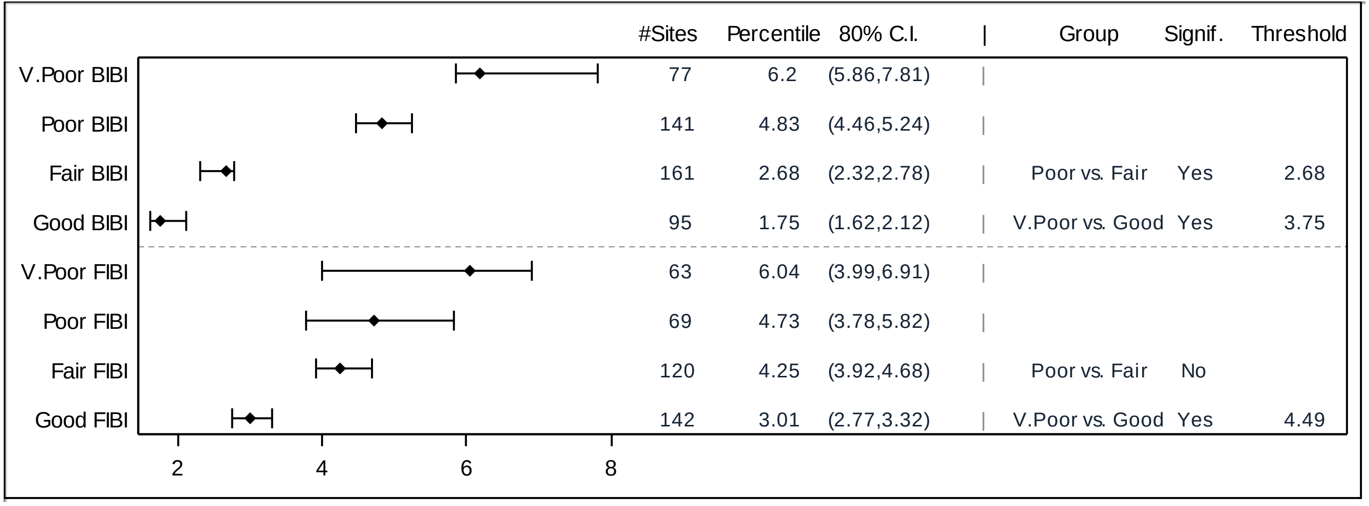
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	80	0.024	(0.021,0.031)			
Poor BIBI	155	0.028	(0.021,0.041)			
Fair BIBI	195	0.023	(0.019,0.029)	Poor vs. Fair	No	
Good BIBI	196	0.026	(0.022,0.028)	V.Poor vs. Good	No	0.025
V.Poor FIBI	56	0.022	(0.012,0.033)			
Poor FIBI	97	0.027	(0.019,0.04)			
Fair FIBI	167	0.027	(0.021,0.035)	Poor vs. Fair	No	
Good FIBI	186	0.027	(0.022,0.029)	V.Poor vs. Good	No	0.027

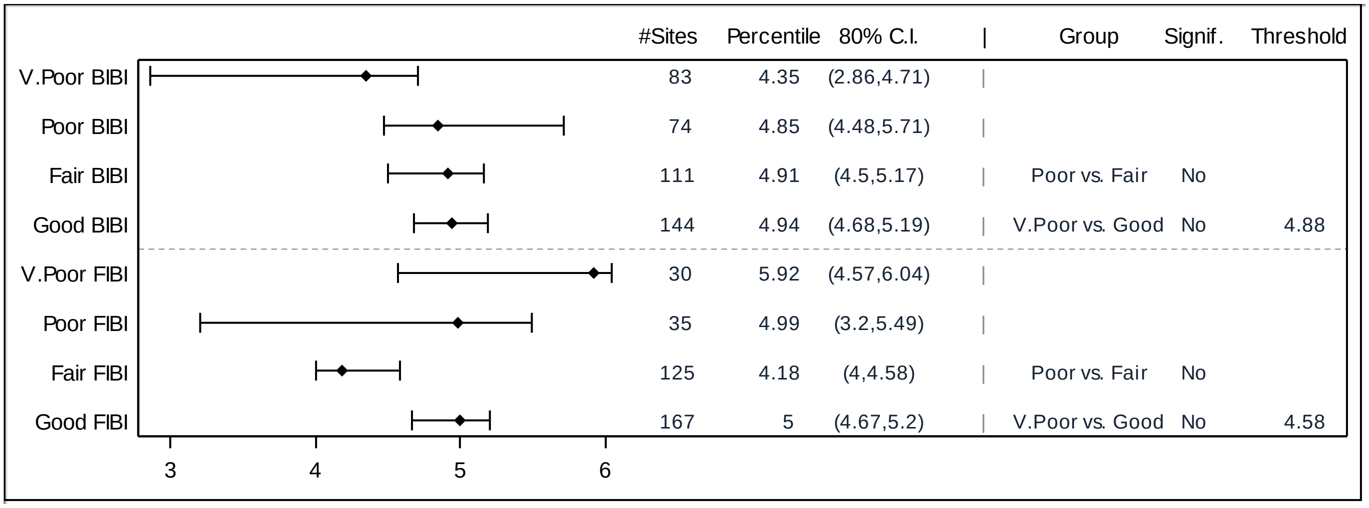
Table A-6. Physiographic Eco-region Analysis for High Total Nitrogen

Highland



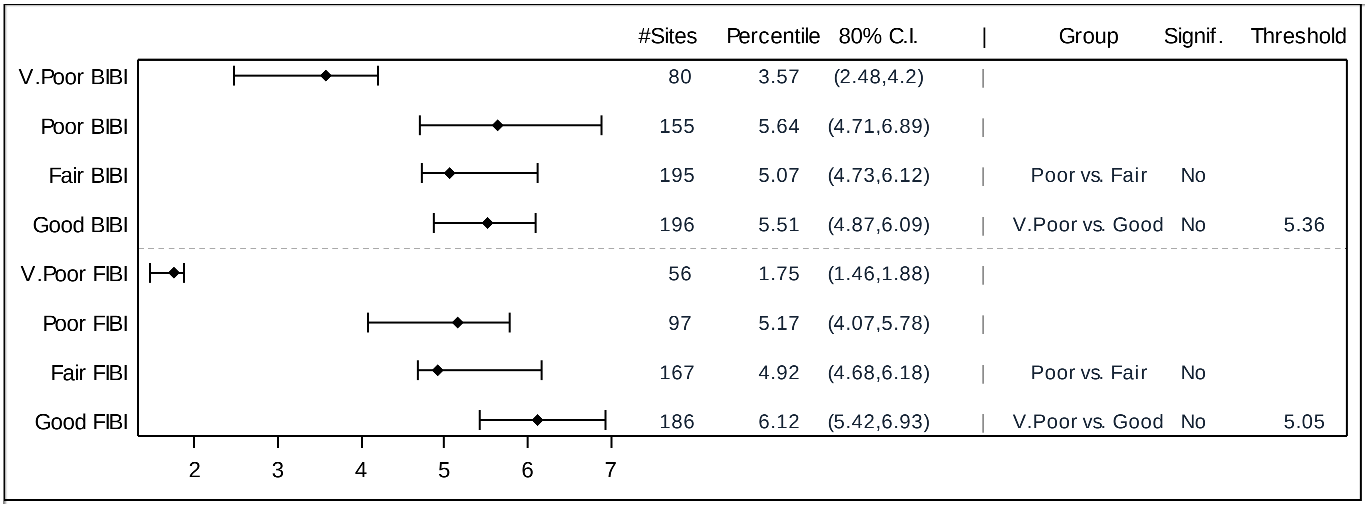
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	77	6.2	(5.86,7.81)			
Poor BIBI	141	4.83	(4.46,5.24)			
Fair BIBI	161	2.68	(2.32,2.78)	Poor vs. Fair	Yes	2.68
Good BIBI	95	1.75	(1.62,2.12)	V.Poor vs. Good	Yes	3.75
V.Poor FIBI	63	6.04	(3.99,6.91)			
Poor FIBI	69	4.73	(3.78,5.82)			
Fair FIBI	120	4.25	(3.92,4.68)	Poor vs. Fair	No	
Good FIBI	142	3.01	(2.77,3.32)	V.Poor vs. Good	Yes	4.49

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	83	4.35	(2.86,4.71)			
Poor BIBI	74	4.85	(4.48,5.71)			
Fair BIBI	111	4.91	(4.5,5.17)	Poor vs. Fair	No	
Good BIBI	144	4.94	(4.68,5.19)	V.Poor vs. Good	No	4.88
V.Poor FIBI	30	5.92	(4.57,6.04)			
Poor FIBI	35	4.99	(3.2,5.49)			
Fair FIBI	125	4.18	(4,4.58)	Poor vs. Fair	No	
Good FIBI	167	5	(4.67,5.2)	V.Poor vs. Good	No	4.58

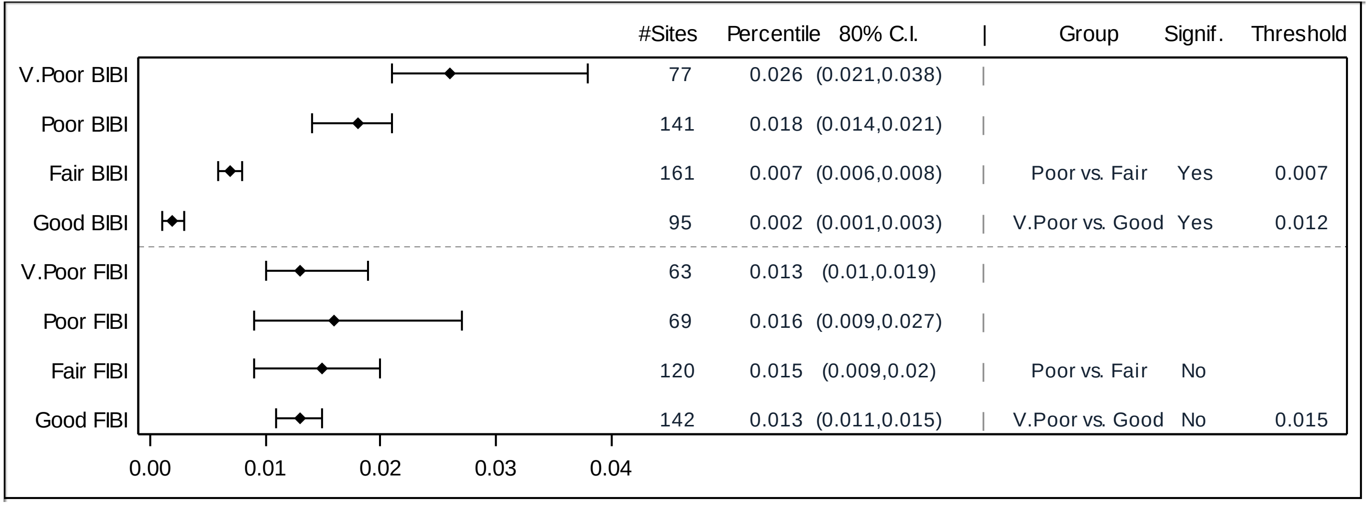
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	80	3.57	(2.48,4.2)			
Poor BIBI	155	5.64	(4.71,6.89)			
Fair BIBI	195	5.07	(4.73,6.12)	Poor vs. Fair	No	
Good BIBI	196	5.51	(4.87,6.09)	V.Poor vs. Good	No	5.36
V.Poor FIBI	56	1.75	(1.46,1.88)			
Poor FIBI	97	5.17	(4.07,5.78)			
Fair FIBI	167	4.92	(4.68,6.18)	Poor vs. Fair	No	
Good FIBI	186	6.12	(5.42,6.93)	V.Poor vs. Good	No	5.05

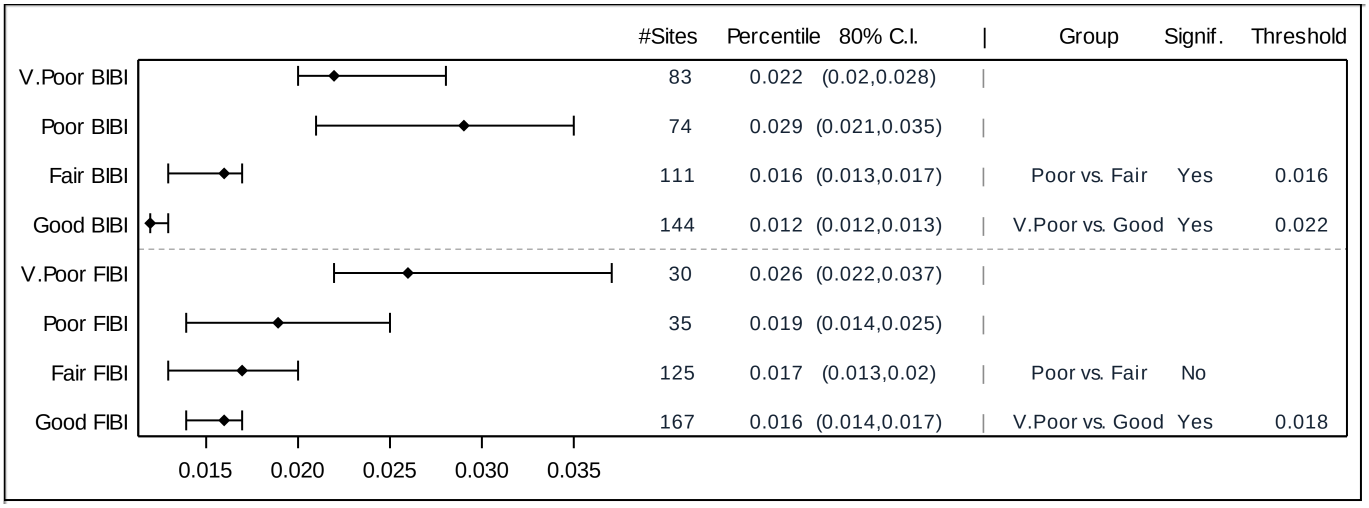
Table A-7. Physiographic Eco-region Analysis for High Nitrites

Highland



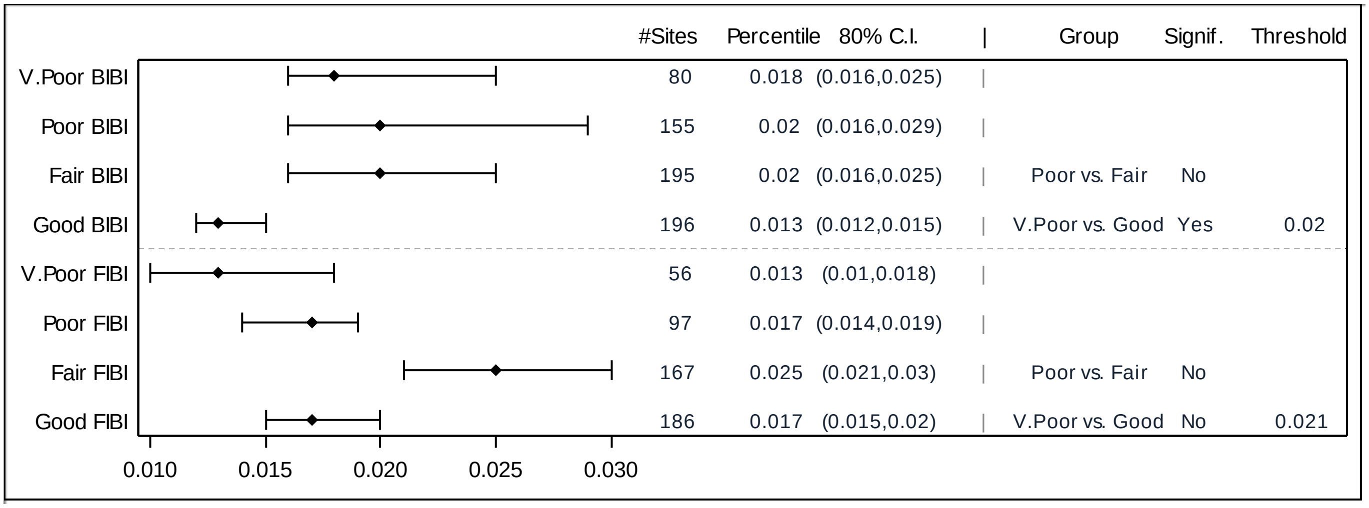
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	77	0.026	(0.021,0.038)			
Poor BIBI	141	0.018	(0.014,0.021)			
Fair BIBI	161	0.007	(0.006,0.008)	Poor vs. Fair	Yes	0.007
Good BIBI	95	0.002	(0.001,0.003)	V.Poor vs. Good	Yes	0.012
V.Poor FIBI	63	0.013	(0.01,0.019)			
Poor FIBI	69	0.016	(0.009,0.027)			
Fair FIBI	120	0.015	(0.009,0.02)	Poor vs. Fair	No	
Good FIBI	142	0.013	(0.011,0.015)	V.Poor vs. Good	No	0.015

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	83	0.022	(0.02,0.028)			
Poor BIBI	74	0.029	(0.021,0.035)			
Fair BIBI	111	0.016	(0.013,0.017)	Poor vs. Fair	Yes	0.016
Good BIBI	144	0.012	(0.012,0.013)	V.Poor vs. Good	Yes	0.022
V.Poor FIBI	30	0.026	(0.022,0.037)			
Poor FIBI	35	0.019	(0.014,0.025)			
Fair FIBI	125	0.017	(0.013,0.02)	Poor vs. Fair	No	
Good FIBI	167	0.016	(0.014,0.017)	V.Poor vs. Good	Yes	0.018

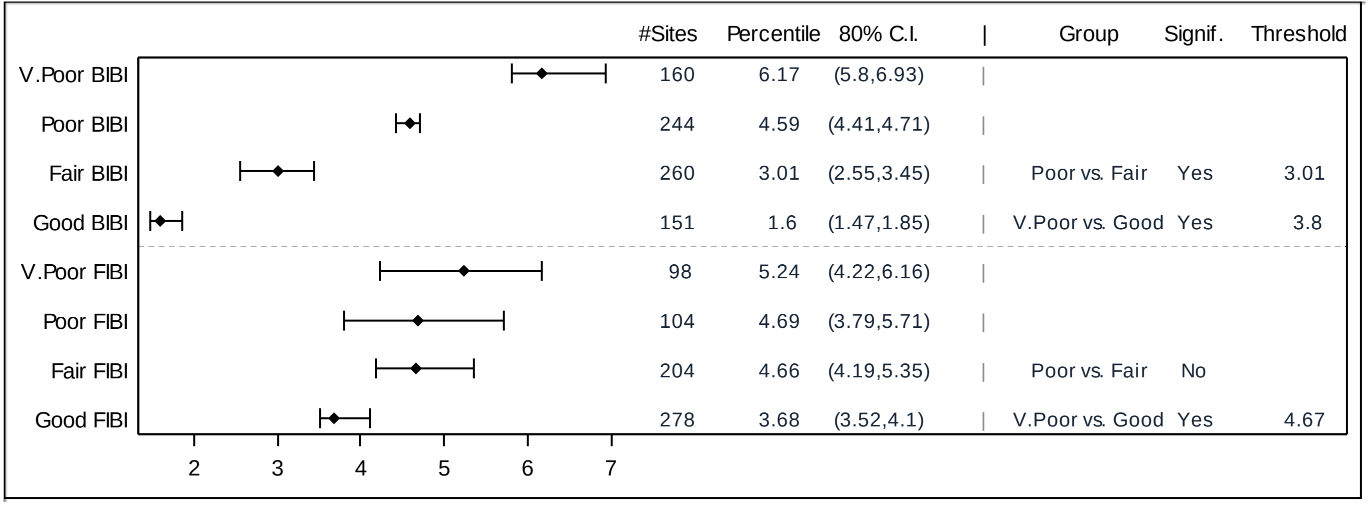
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	80	0.018	(0.016,0.025)			
Poor BIBI	155	0.02	(0.016,0.029)			
Fair BIBI	195	0.02	(0.016,0.025)	Poor vs. Fair	No	
Good BIBI	196	0.013	(0.012,0.015)	V.Poor vs. Good	Yes	0.02
V.Poor FIBI	56	0.013	(0.01,0.018)			
Poor FIBI	97	0.017	(0.014,0.019)			
Fair FIBI	167	0.025	(0.021,0.03)	Poor vs. Fair	No	
Good FIBI	186	0.017	(0.015,0.02)	V.Poor vs. Good	No	0.021

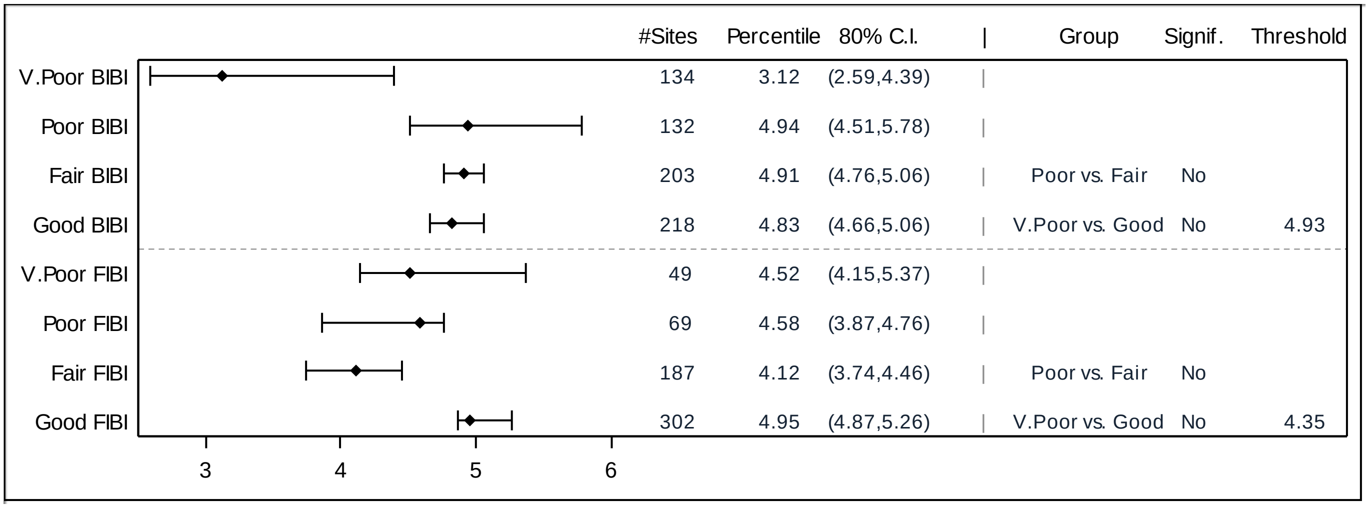
Table A-8. Physiographic Eco-region Analysis for High Nitrates

Highland



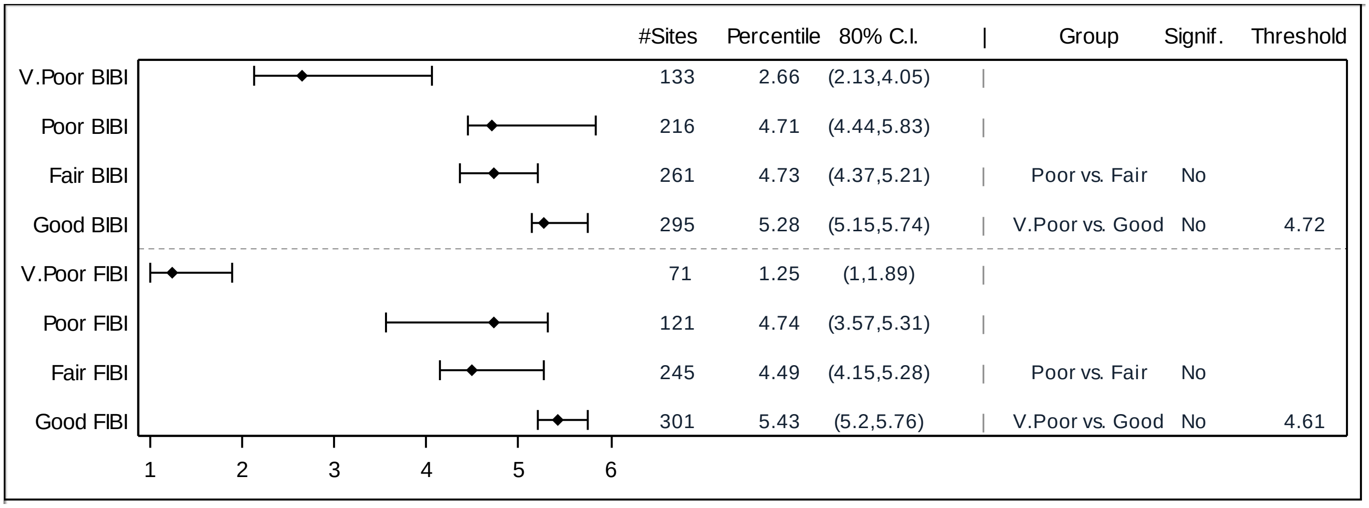
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	6.17	(5.8,6.93)			
Poor BIBI	244	4.59	(4.41,4.71)			
Fair BIBI	260	3.01	(2.55,3.45)	Poor vs. Fair	Yes	3.01
Good BIBI	151	1.6	(1.47,1.85)	V.Poor vs. Good	Yes	3.8
V.Poor FIBI	98	5.24	(4.22,6.16)			
Poor FIBI	104	4.69	(3.79,5.71)			
Fair FIBI	204	4.66	(4.19,5.35)	Poor vs. Fair	No	
Good FIBI	278	3.68	(3.52,4.1)	V.Poor vs. Good	Yes	4.67

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	3.12	(2.59,4.39)			
Poor BIBI	132	4.94	(4.51,5.78)			
Fair BIBI	203	4.91	(4.76,5.06)	Poor vs. Fair	No	
Good BIBI	218	4.83	(4.66,5.06)	V.Poor vs. Good	No	4.93
V.Poor FIBI	49	4.52	(4.15,5.37)			
Poor FIBI	69	4.58	(3.87,4.76)			
Fair FIBI	187	4.12	(3.74,4.46)	Poor vs. Fair	No	
Good FIBI	302	4.95	(4.87,5.26)	V.Poor vs. Good	No	4.35

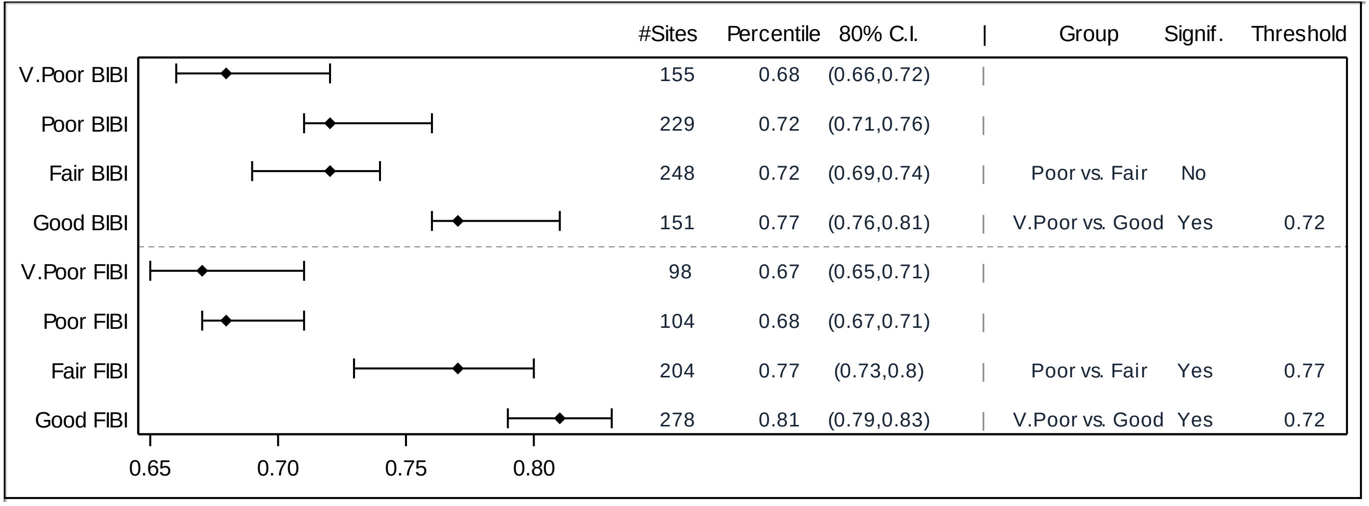
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	2.66	(2.13,4.05)			
Poor BIBI	216	4.71	(4.44,5.83)			
Fair BIBI	261	4.73	(4.37,5.21)	Poor vs. Fair	No	
Good BIBI	295	5.28	(5.15,5.74)	V.Poor vs. Good	No	4.72
V.Poor FIBI	71	1.25	(1,1.89)			
Poor FIBI	121	4.74	(3.57,5.31)			
Fair FIBI	245	4.49	(4.15,5.28)	Poor vs. Fair	No	
Good FIBI	301	5.43	(5.2,5.76)	V.Poor vs. Good	No	4.61

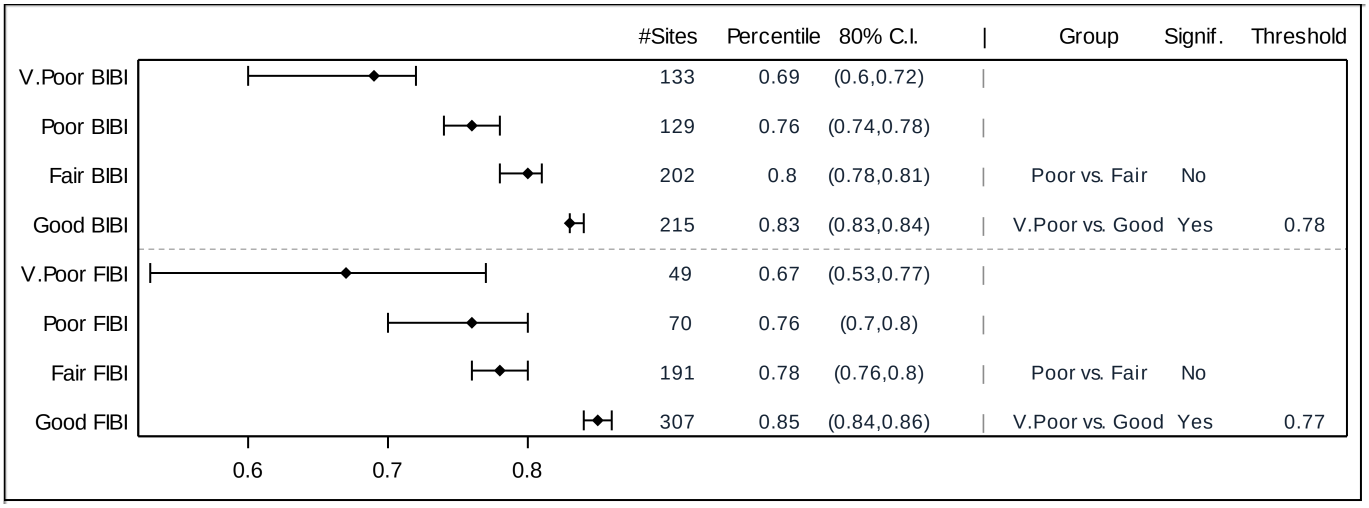
Table A-9. Physiographic Eco-region Analysis for Low Dissolved Oxygen Saturation

Highland



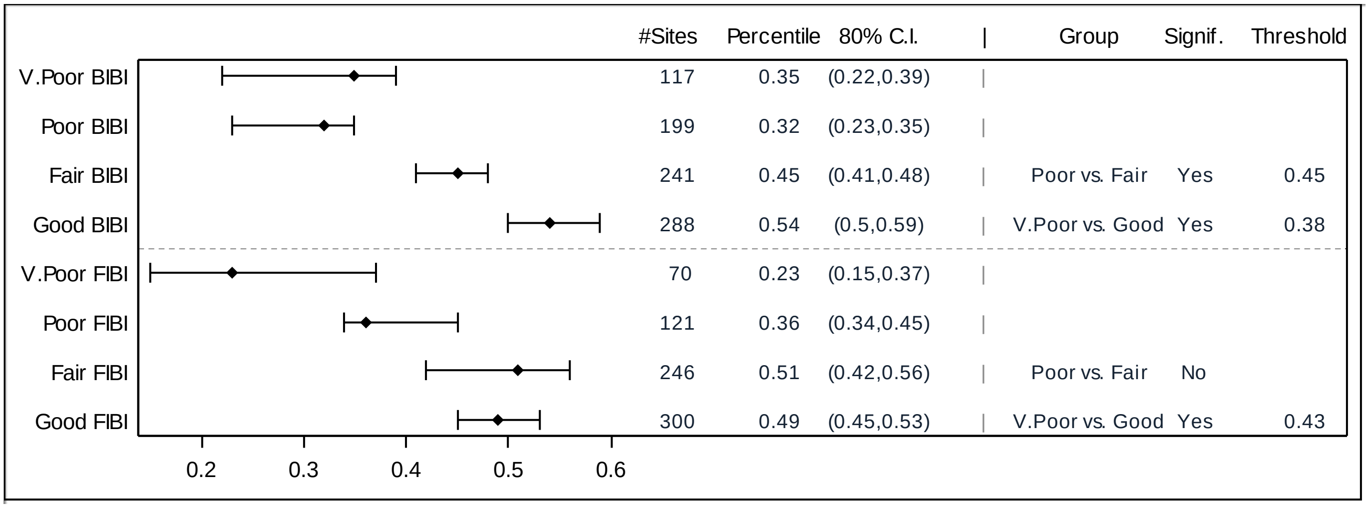
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	155	0.68	(0.66,0.72)			
Poor BIBI	229	0.72	(0.71,0.76)			
Fair BIBI	248	0.72	(0.69,0.74)	Poor vs. Fair	No	
Good BIBI	151	0.77	(0.76,0.81)	V.Poor vs. Good	Yes	0.72
V.Poor FIBI	98	0.67	(0.65,0.71)			
Poor FIBI	104	0.68	(0.67,0.71)			
Fair FIBI	204	0.77	(0.73,0.8)	Poor vs. Fair	Yes	0.77
Good FIBI	278	0.81	(0.79,0.83)	V.Poor vs. Good	Yes	0.72

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	0.69	(0.6,0.72)			
Poor BIBI	129	0.76	(0.74,0.78)			
Fair BIBI	202	0.8	(0.78,0.81)	Poor vs. Fair	No	
Good BIBI	215	0.83	(0.83,0.84)	V.Poor vs. Good	Yes	0.78
V.Poor FIBI	49	0.67	(0.53,0.77)			
Poor FIBI	70	0.76	(0.7,0.8)			
Fair FIBI	191	0.78	(0.76,0.8)	Poor vs. Fair	No	
Good FIBI	307	0.85	(0.84,0.86)	V.Poor vs. Good	Yes	0.77

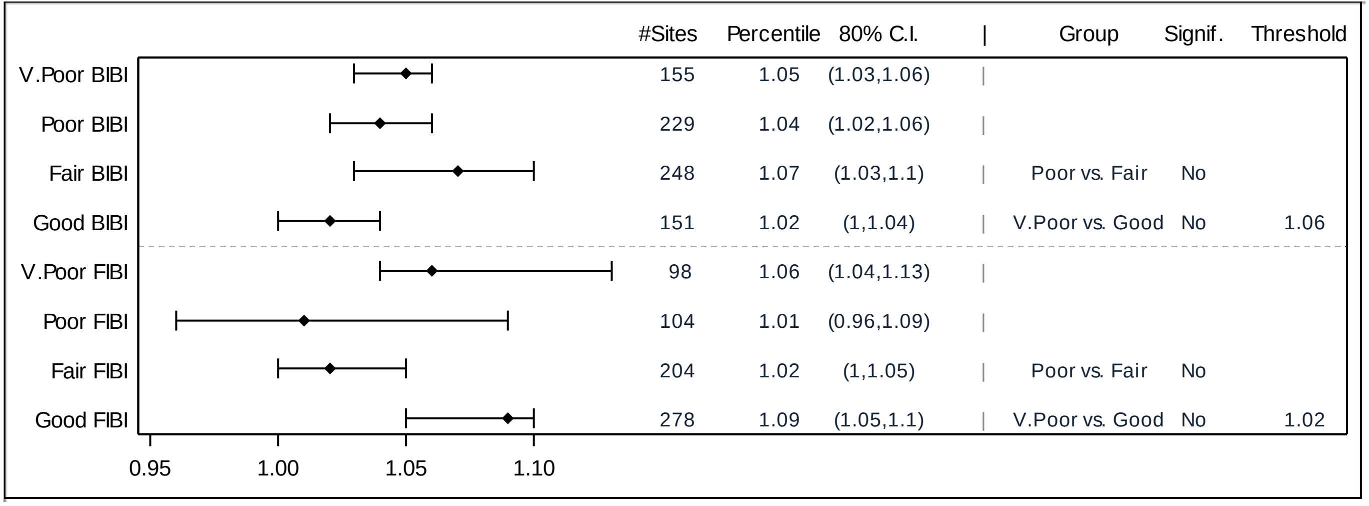
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	117	0.35	(0.22,0.39)			
Poor BIBI	199	0.32	(0.23,0.35)			
Fair BIBI	241	0.45	(0.41,0.48)	Poor vs. Fair	Yes	0.45
Good BIBI	288	0.54	(0.5,0.59)	V.Poor vs. Good	Yes	0.38
V.Poor FIBI	70	0.23	(0.15,0.37)			
Poor FIBI	121	0.36	(0.34,0.45)			
Fair FIBI	246	0.51	(0.42,0.56)	Poor vs. Fair	No	
Good FIBI	300	0.49	(0.45,0.53)	V.Poor vs. Good	Yes	0.43

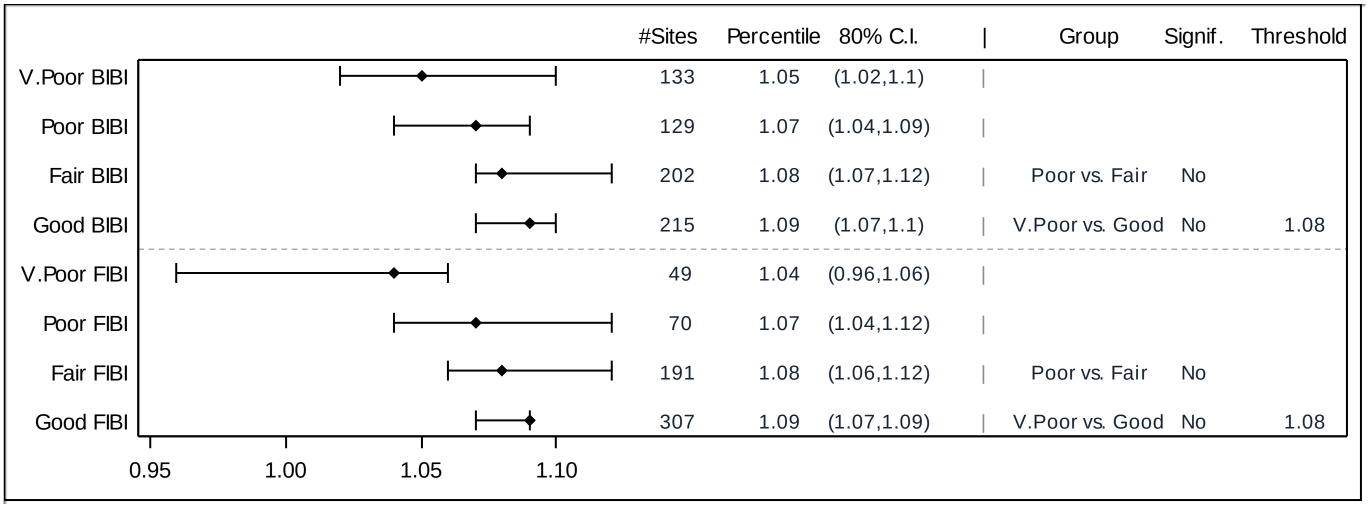
Table A-10. Physiographic Eco-region Analysis for High Dissolved Oxygen Saturation

Highland



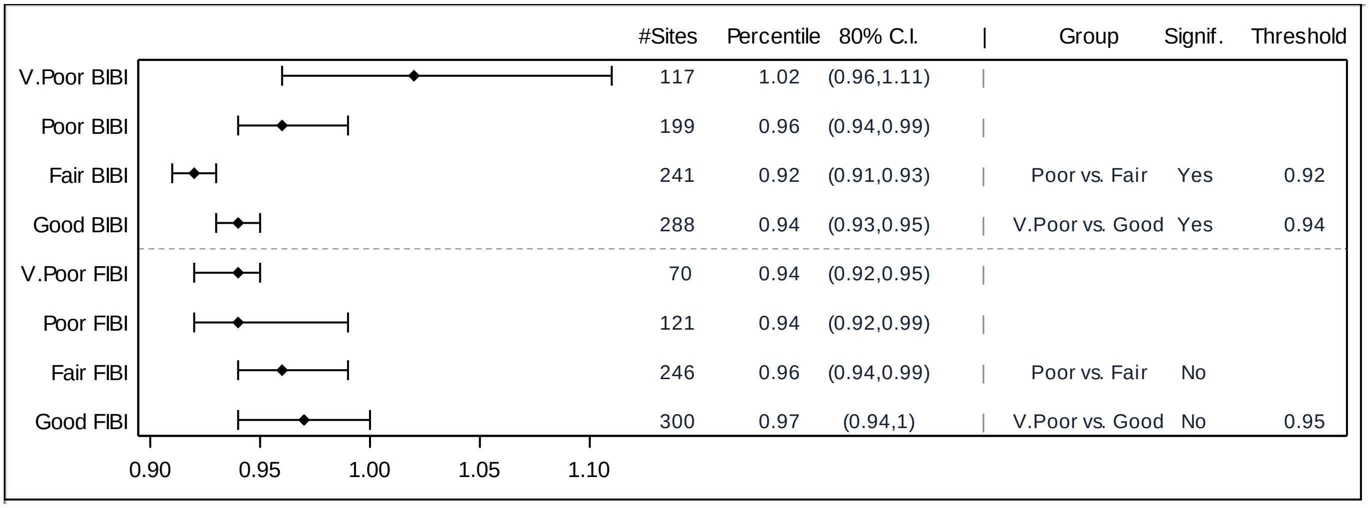
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	155	1.05	(1.03,1.06)			
Poor BIBI	229	1.04	(1.02,1.06)			
Fair BIBI	248	1.07	(1.03,1.1)	Poor vs. Fair	No	
Good BIBI	151	1.02	(1,1.04)	V.Poor vs. Good	No	1.06
V.Poor FIBI	98	1.06	(1.04,1.13)			
Poor FIBI	104	1.01	(0.96,1.09)			
Fair FIBI	204	1.02	(1,1.05)	Poor vs. Fair	No	
Good FIBI	278	1.09	(1.05,1.1)	V.Poor vs. Good	No	1.02

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	1.05	(1.02,1.1)			
Poor BIBI	129	1.07	(1.04,1.09)			
Fair BIBI	202	1.08	(1.07,1.12)	Poor vs. Fair	No	
Good BIBI	215	1.09	(1.07,1.1)	V.Poor vs. Good	No	1.08
V.Poor FIBI	49	1.04	(0.96,1.06)			
Poor FIBI	70	1.07	(1.04,1.12)			
Fair FIBI	191	1.08	(1.06,1.12)	Poor vs. Fair	No	
Good FIBI	307	1.09	(1.07,1.09)	V.Poor vs. Good	No	1.08

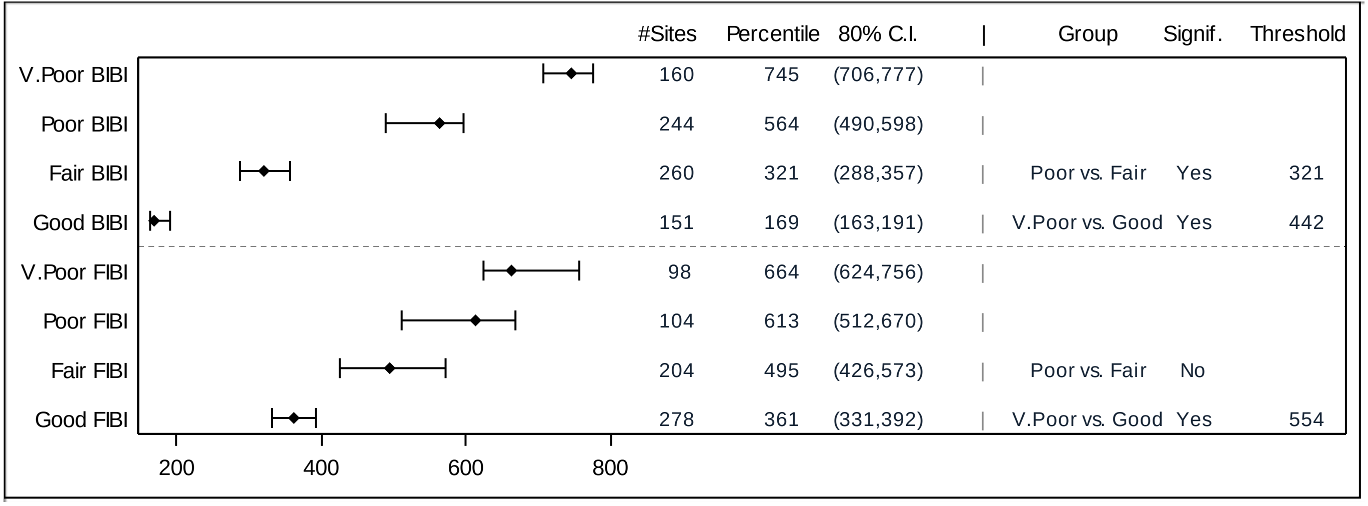
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	117	1.02	(0.96,1.11)			
Poor BIBI	199	0.96	(0.94,0.99)			
Fair BIBI	241	0.92	(0.91,0.93)	Poor vs. Fair	Yes	0.92
Good BIBI	288	0.94	(0.93,0.95)	V.Poor vs. Good	Yes	0.94
V.Poor FIBI	70	0.94	(0.92,0.95)			
Poor FIBI	121	0.94	(0.92,0.99)			
Fair FIBI	246	0.96	(0.94,0.99)	Poor vs. Fair	No	
Good FIBI	300	0.97	(0.94,1)	V.Poor vs. Good	No	0.95

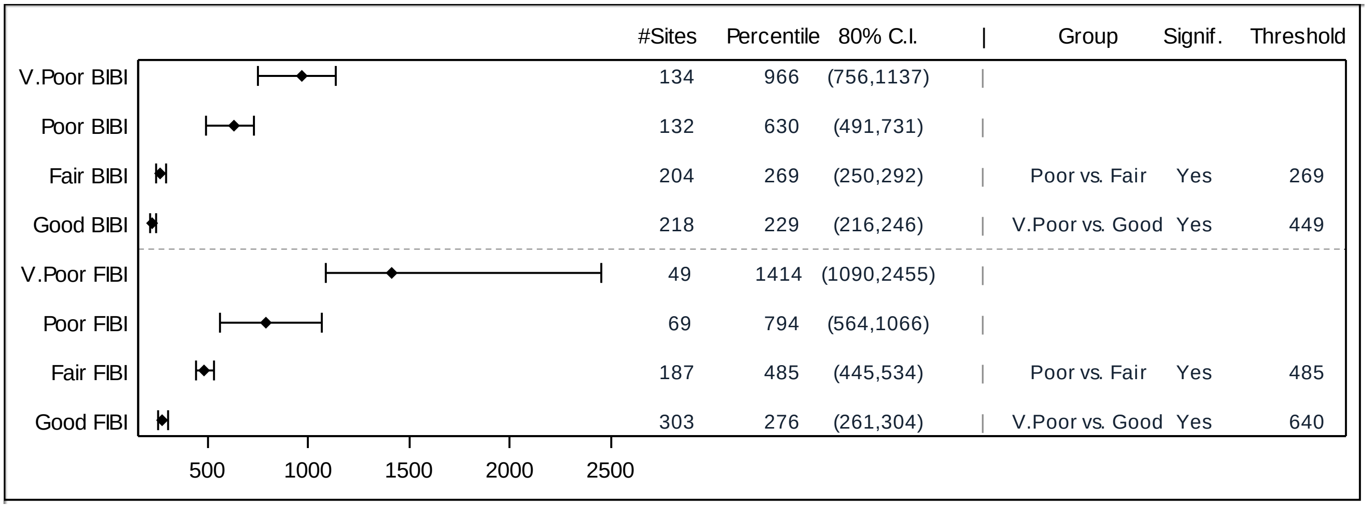
Table A-11. Physiographic Eco-region Analysis for High Conductivity

Highland



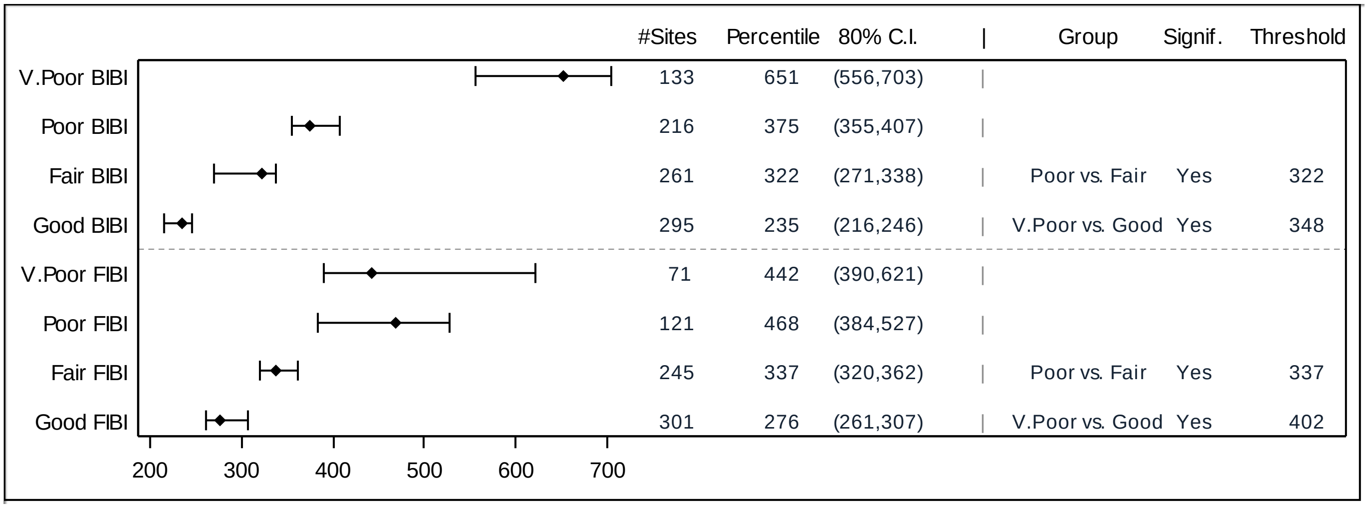
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	745	(706,777)			
Poor BIBI	244	564	(490,598)			
Fair BIBI	260	321	(288,357)	Poor vs. Fair	Yes	321
Good BIBI	151	169	(163,191)	V.Poor vs. Good	Yes	442
V.Poor FIBI	98	664	(624,756)			
Poor FIBI	104	613	(512,670)			
Fair FIBI	204	495	(426,573)	Poor vs. Fair	No	
Good FIBI	278	361	(331,392)	V.Poor vs. Good	Yes	554

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	966	(756,1137)			
Poor BIBI	132	630	(491,731)			
Fair BIBI	204	269	(250,292)	Poor vs. Fair	Yes	269
Good BIBI	218	229	(216,246)	V.Poor vs. Good	Yes	449
V.Poor FIBI	49	1414	(1090,2455)			
Poor FIBI	69	794	(564,1066)			
Fair FIBI	187	485	(445,534)	Poor vs. Fair	Yes	485
Good FIBI	303	276	(261,304)	V.Poor vs. Good	Yes	640

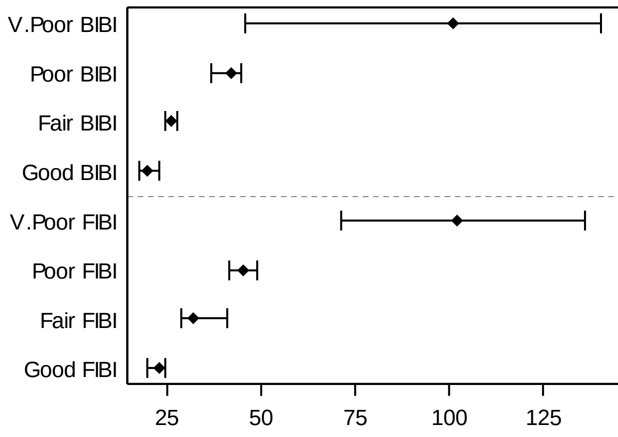
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	651	(556,703)			
Poor BIBI	216	375	(355,407)			
Fair BIBI	261	322	(271,338)	Poor vs. Fair	Yes	322
Good BIBI	295	235	(216,246)	V.Poor vs. Good	Yes	348
V.Poor FIBI	71	442	(390,621)			
Poor FIBI	121	468	(384,527)			
Fair FIBI	245	337	(320,362)	Poor vs. Fair	Yes	337
Good FIBI	301	276	(261,307)	V.Poor vs. Good	Yes	402

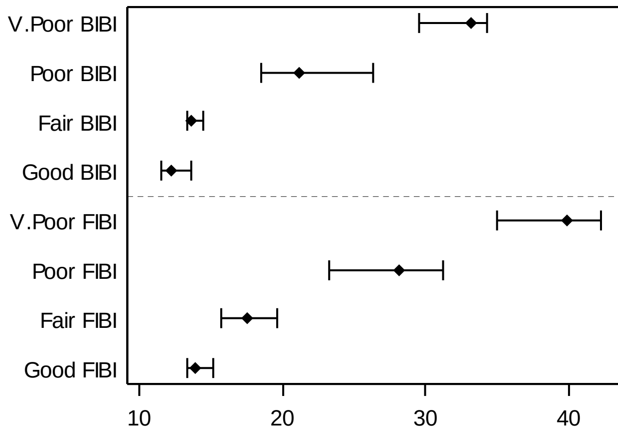
Table A-12. Physiographic Eco-region Analysis for High Sulfates

Highland



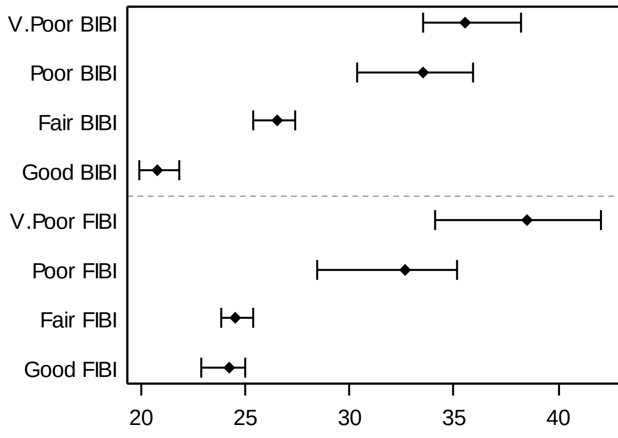
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	101	(45.9,140)			
Poor BIBI	244	42.1	(37,44.9)			
Fair BIBI	260	26	(24.3,28)	Poor vs. Fair	Yes	26
Good BIBI	151	19.9	(17.6,23.2)	V.Poor vs. Good	Yes	34
V.Poor FIBI	98	102	(71.2,136)			
Poor FIBI	104	45	(41.6,49.2)			
Fair FIBI	204	31.9	(28.9,41.2)	Poor vs. Fair	Yes	31.9
Good FIBI	278	23.1	(19.9,24.8)	V.Poor vs. Good	Yes	38.4

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	33.2	(29.6,34.3)			
Poor BIBI	132	21.2	(18.5,26.3)			
Fair BIBI	203	13.7	(13.3,14.5)	Poor vs. Fair	Yes	13.7
Good BIBI	218	12.2	(11.6,13.6)	V.Poor vs. Good	Yes	17.4
V.Poor FIBI	49	39.9	(35,42.2)			
Poor FIBI	69	28.2	(23.3,31.3)			
Fair FIBI	187	17.6	(15.8,19.7)	Poor vs. Fair	Yes	17.6
Good FIBI	302	13.9	(13.4,15.2)	V.Poor vs. Good	Yes	22.9

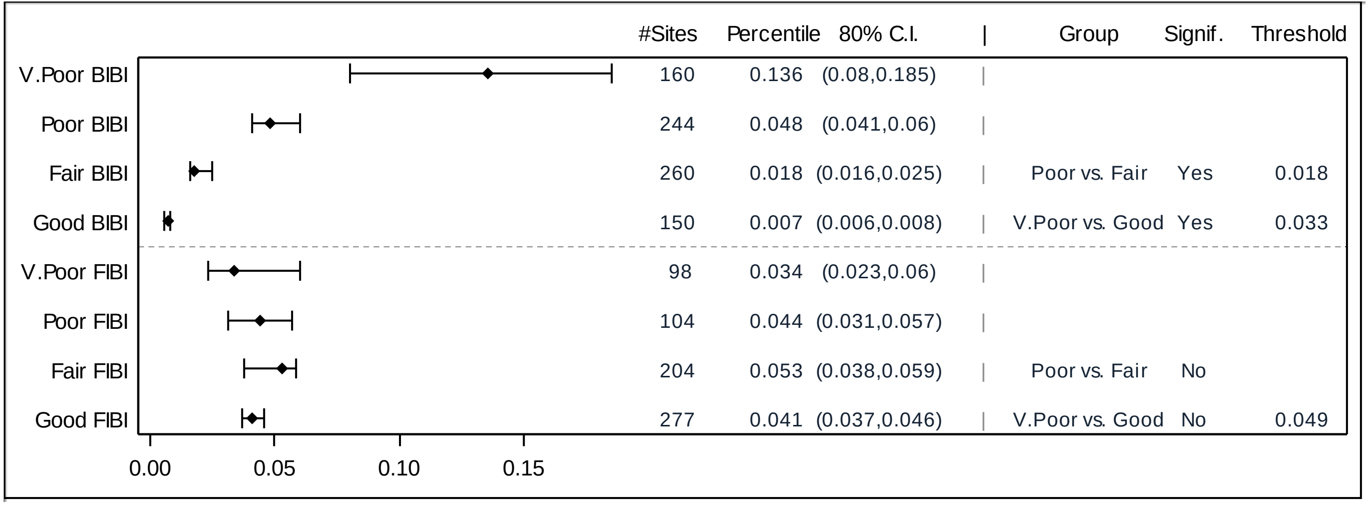
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	133	35.5	(33.5,38.2)			
Poor BIBI	216	33.5	(30.4,35.9)			
Fair BIBI	261	26.5	(25.4,27.4)	Poor vs. Fair	Yes	26.5
Good BIBI	295	20.8	(19.9,21.8)	V.Poor vs. Good	Yes	30
V.Poor FIBI	71	38.5	(34.1,42)			
Poor FIBI	121	32.7	(28.4,35.1)			
Fair FIBI	245	24.5	(23.8,25.4)	Poor vs. Fair	Yes	24.5
Good FIBI	301	24.2	(22.9,25)	V.Poor vs. Good	Yes	28.6

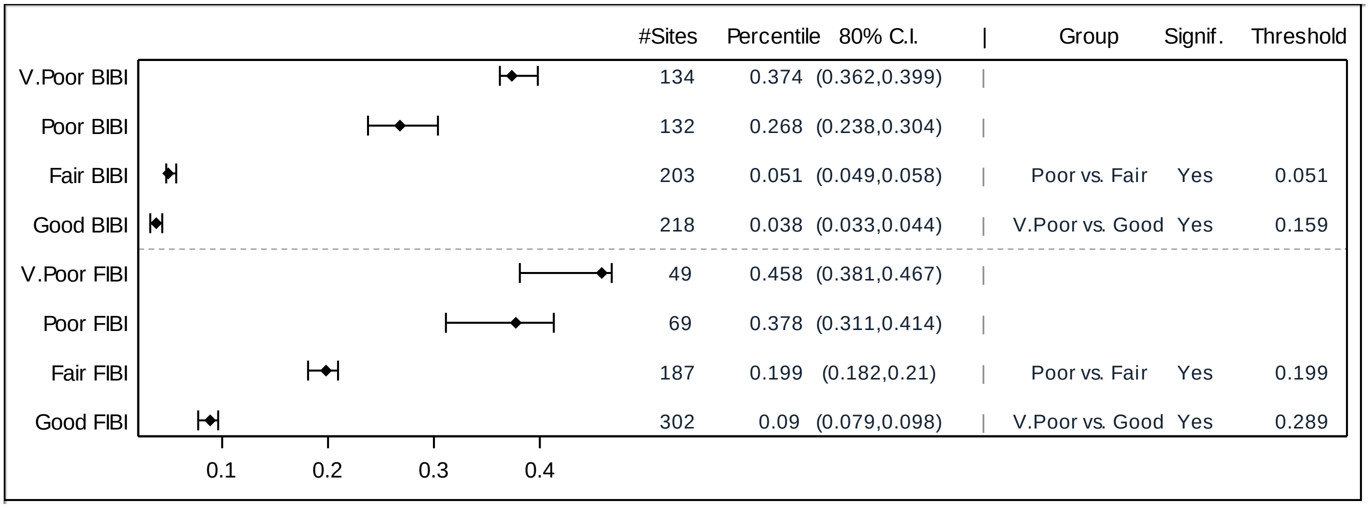
Table A-13. Physiographic Eco-region Analysis for High % of Impervious Surface in Watershed

Highland



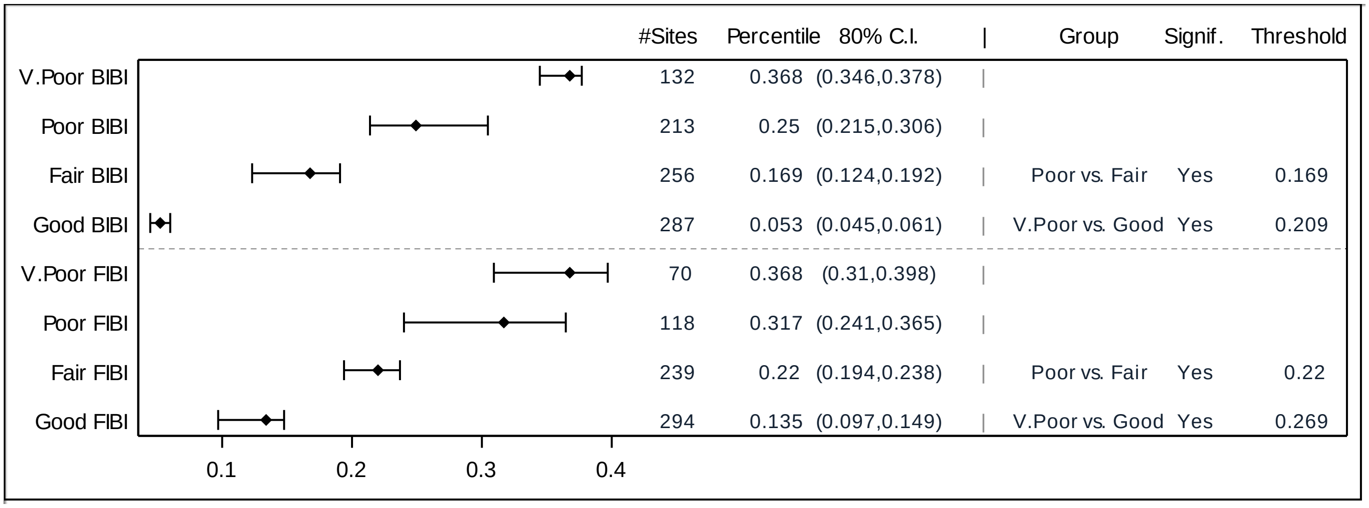
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.136	(0.08,0.185)			
Poor BIBI	244	0.048	(0.041,0.06)			
Fair BIBI	260	0.018	(0.016,0.025)	Poor vs. Fair	Yes	0.018
Good BIBI	150	0.007	(0.006,0.008)	V.Poor vs. Good	Yes	0.033
V.Poor FIBI	98	0.034	(0.023,0.06)			
Poor FIBI	104	0.044	(0.031,0.057)			
Fair FIBI	204	0.053	(0.038,0.059)	Poor vs. Fair	No	
Good FIBI	277	0.041	(0.037,0.046)	V.Poor vs. Good	No	0.049

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.374	(0.362,0.399)			
Poor BIBI	132	0.268	(0.238,0.304)			
Fair BIBI	203	0.051	(0.049,0.058)	Poor vs. Fair	Yes	0.051
Good BIBI	218	0.038	(0.033,0.044)	V.Poor vs. Good	Yes	0.159
V.Poor FIBI	49	0.458	(0.381,0.467)			
Poor FIBI	69	0.378	(0.311,0.414)			
Fair FIBI	187	0.199	(0.182,0.21)	Poor vs. Fair	Yes	0.199
Good FIBI	302	0.09	(0.079,0.098)	V.Poor vs. Good	Yes	0.289

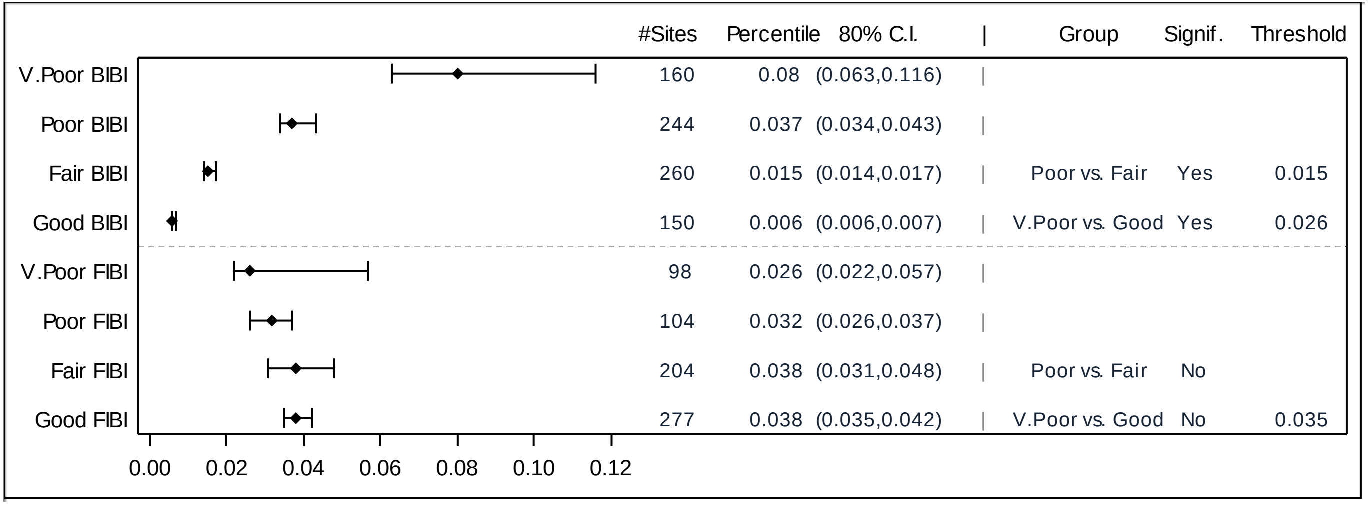
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.368	(0.346,0.378)			
Poor BIBI	213	0.25	(0.215,0.306)			
Fair BIBI	256	0.169	(0.124,0.192)	Poor vs. Fair	Yes	0.169
Good BIBI	287	0.053	(0.045,0.061)	V.Poor vs. Good	Yes	0.209
V.Poor FIBI	70	0.368	(0.31,0.398)			
Poor FIBI	118	0.317	(0.241,0.365)			
Fair FIBI	239	0.22	(0.194,0.238)	Poor vs. Fair	Yes	0.22
Good FIBI	294	0.135	(0.097,0.149)	V.Poor vs. Good	Yes	0.269

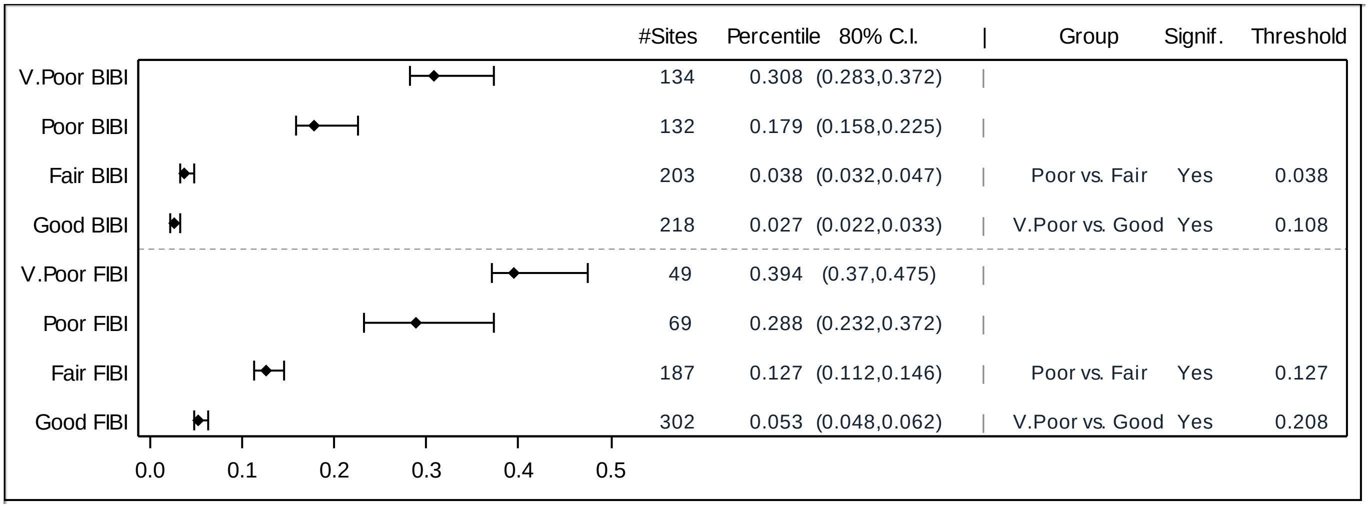
Table A-14. Physiographic Eco-region Analysis for High % of Impervious Surface in 60m Buffer

Highland



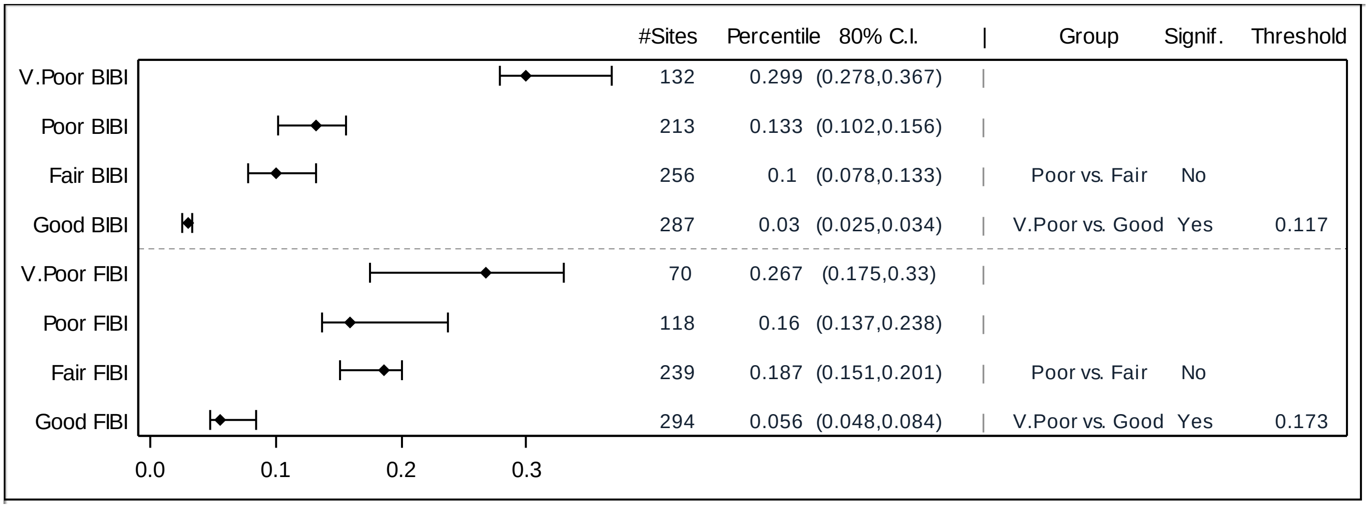
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.08	(0.063,0.116)			
Poor BIBI	244	0.037	(0.034,0.043)			
Fair BIBI	260	0.015	(0.014,0.017)	Poor vs. Fair	Yes	0.015
Good BIBI	150	0.006	(0.006,0.007)	V.Poor vs. Good	Yes	0.026
V.Poor FIBI	98	0.026	(0.022,0.057)			
Poor FIBI	104	0.032	(0.026,0.037)			
Fair FIBI	204	0.038	(0.031,0.048)	Poor vs. Fair	No	
Good FIBI	277	0.038	(0.035,0.042)	V.Poor vs. Good	No	0.035

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.308	(0.283,0.372)			
Poor BIBI	132	0.179	(0.158,0.225)			
Fair BIBI	203	0.038	(0.032,0.047)	Poor vs. Fair	Yes	0.038
Good BIBI	218	0.027	(0.022,0.033)	V.Poor vs. Good	Yes	0.108
V.Poor FIBI	49	0.394	(0.37,0.475)			
Poor FIBI	69	0.288	(0.232,0.372)			
Fair FIBI	187	0.127	(0.112,0.146)	Poor vs. Fair	Yes	0.127
Good FIBI	302	0.053	(0.048,0.062)	V.Poor vs. Good	Yes	0.208

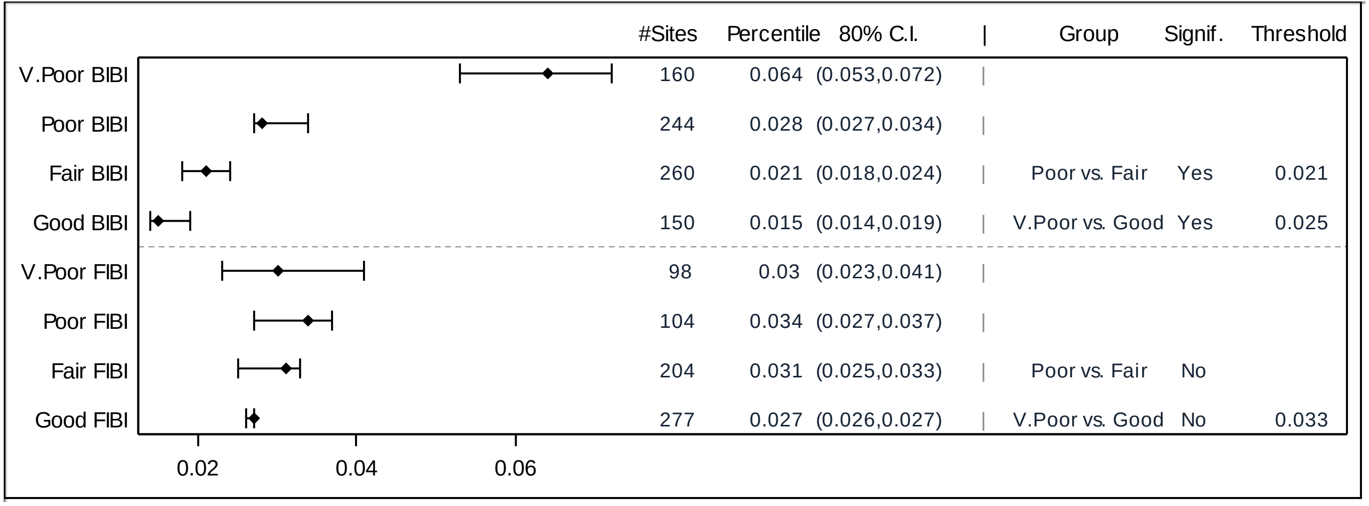
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.299	(0.278,0.367)			
Poor BIBI	213	0.133	(0.102,0.156)			
Fair BIBI	256	0.1	(0.078,0.133)	Poor vs. Fair	No	
Good BIBI	287	0.03	(0.025,0.034)	V.Poor vs. Good	Yes	0.117
V.Poor FIBI	70	0.267	(0.175,0.33)			
Poor FIBI	118	0.16	(0.137,0.238)			
Fair FIBI	239	0.187	(0.151,0.201)	Poor vs. Fair	No	
Good FIBI	294	0.056	(0.048,0.084)	V.Poor vs. Good	Yes	0.173

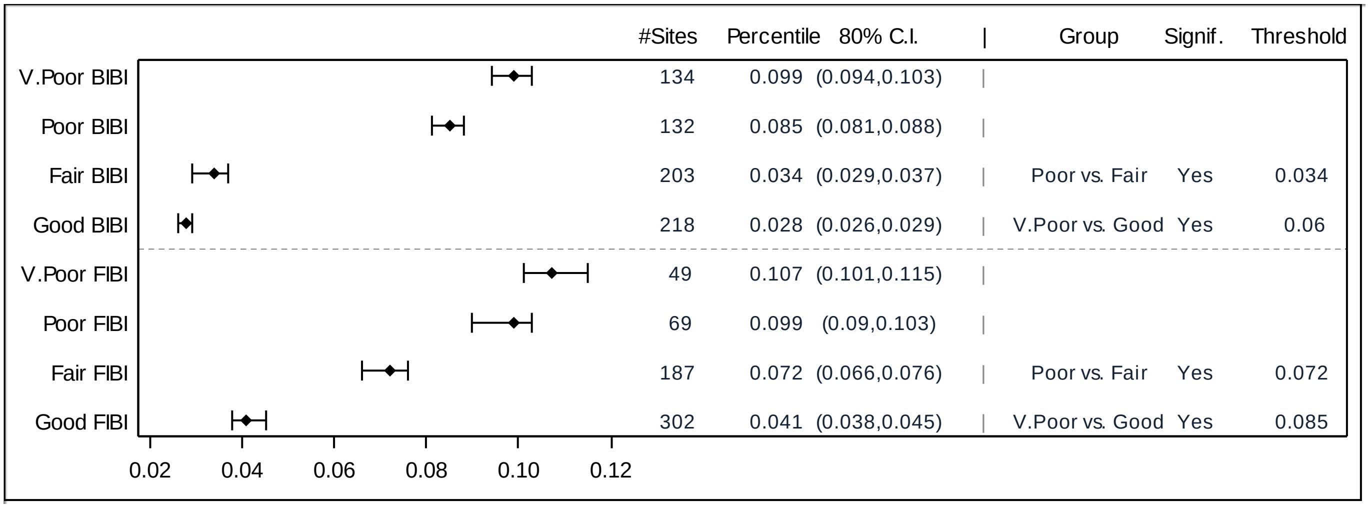
Table A-15. Physiographic Eco-region Analysis for High % of Roads in Watershed

Highland



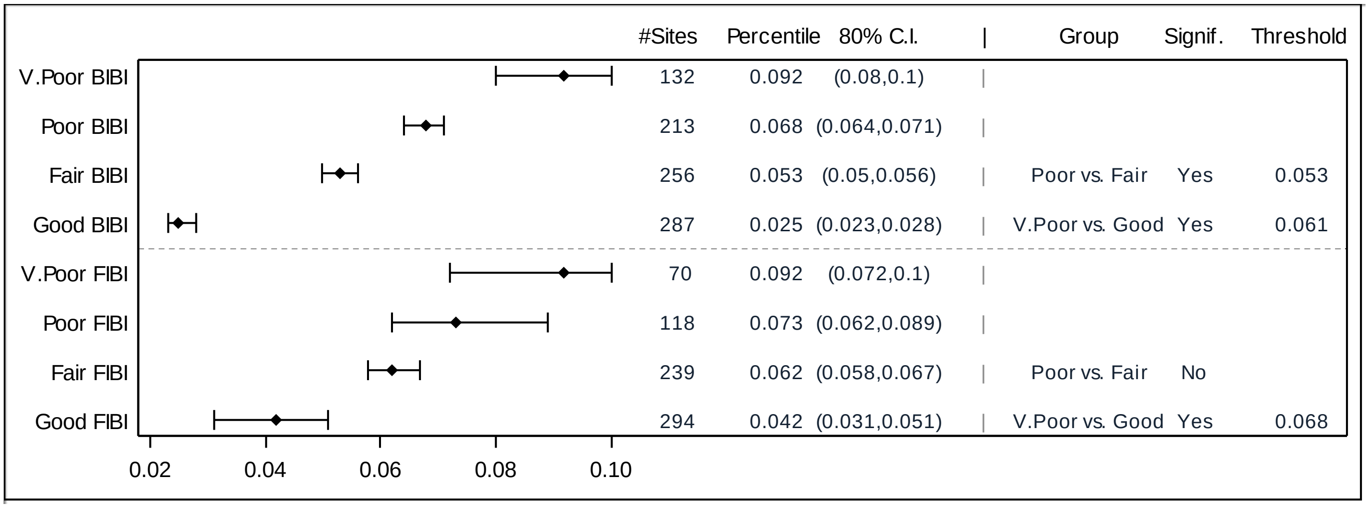
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.064	(0.053,0.072)			
Poor BIBI	244	0.028	(0.027,0.034)			
Fair BIBI	260	0.021	(0.018,0.024)	Poor vs. Fair	Yes	0.021
Good BIBI	150	0.015	(0.014,0.019)	V.Poor vs. Good	Yes	0.025
V.Poor FIBI	98	0.03	(0.023,0.041)			
Poor FIBI	104	0.034	(0.027,0.037)			
Fair FIBI	204	0.031	(0.025,0.033)	Poor vs. Fair	No	
Good FIBI	277	0.027	(0.026,0.027)	V.Poor vs. Good	No	0.033

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.099	(0.094,0.103)			
Poor BIBI	132	0.085	(0.081,0.088)			
Fair BIBI	203	0.034	(0.029,0.037)	Poor vs. Fair	Yes	0.034
Good BIBI	218	0.028	(0.026,0.029)	V.Poor vs. Good	Yes	0.06
V.Poor FIBI	49	0.107	(0.101,0.115)			
Poor FIBI	69	0.099	(0.09,0.103)			
Fair FIBI	187	0.072	(0.066,0.076)	Poor vs. Fair	Yes	0.072
Good FIBI	302	0.041	(0.038,0.045)	V.Poor vs. Good	Yes	0.085

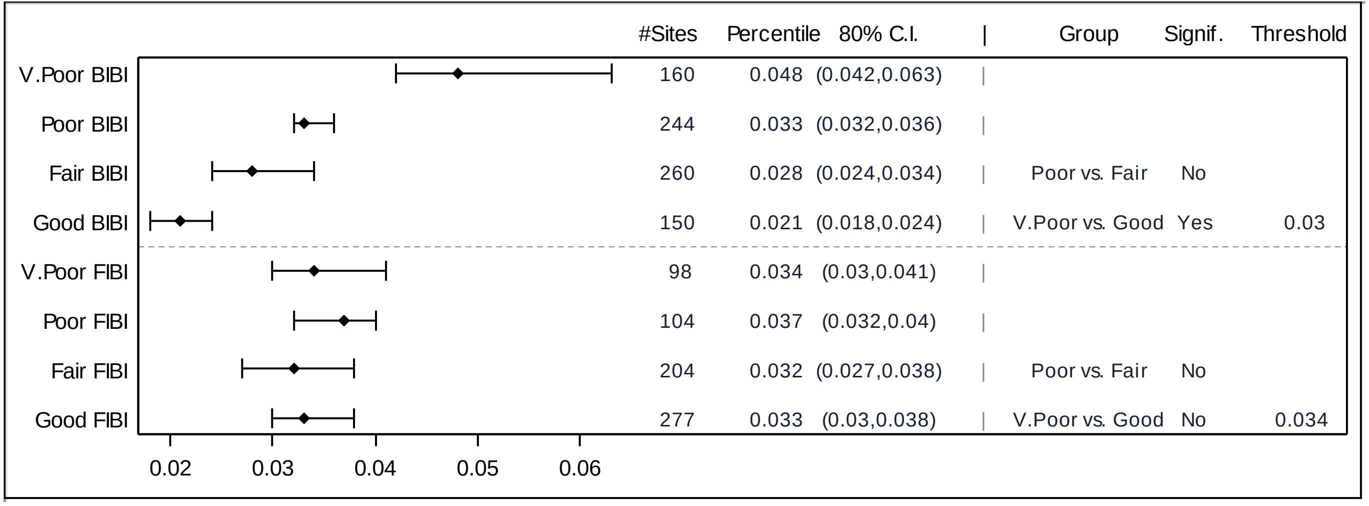
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.092	(0.08,0.1)			
Poor BIBI	213	0.068	(0.064,0.071)			
Fair BIBI	256	0.053	(0.05,0.056)	Poor vs. Fair	Yes	0.053
Good BIBI	287	0.025	(0.023,0.028)	V.Poor vs. Good	Yes	0.061
V.Poor FIBI	70	0.092	(0.072,0.1)			
Poor FIBI	118	0.073	(0.062,0.089)			
Fair FIBI	239	0.062	(0.058,0.067)	Poor vs. Fair	No	
Good FIBI	294	0.042	(0.031,0.051)	V.Poor vs. Good	Yes	0.068

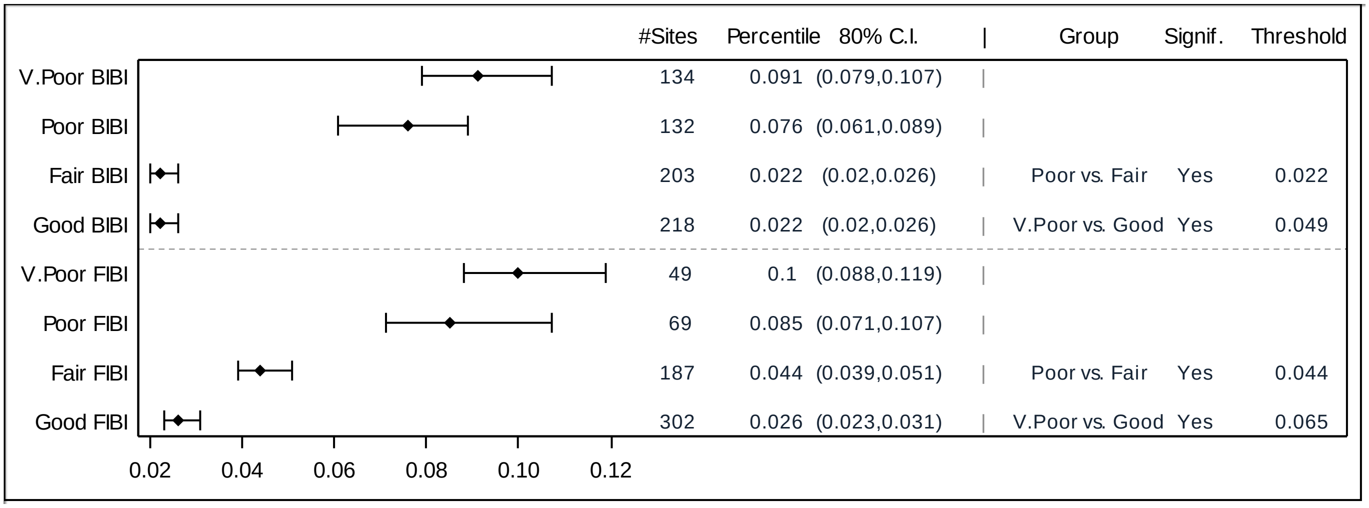
Table A-16. Physiographic Eco-region Analysis for High % of Roads in 60m Buffer

Highland



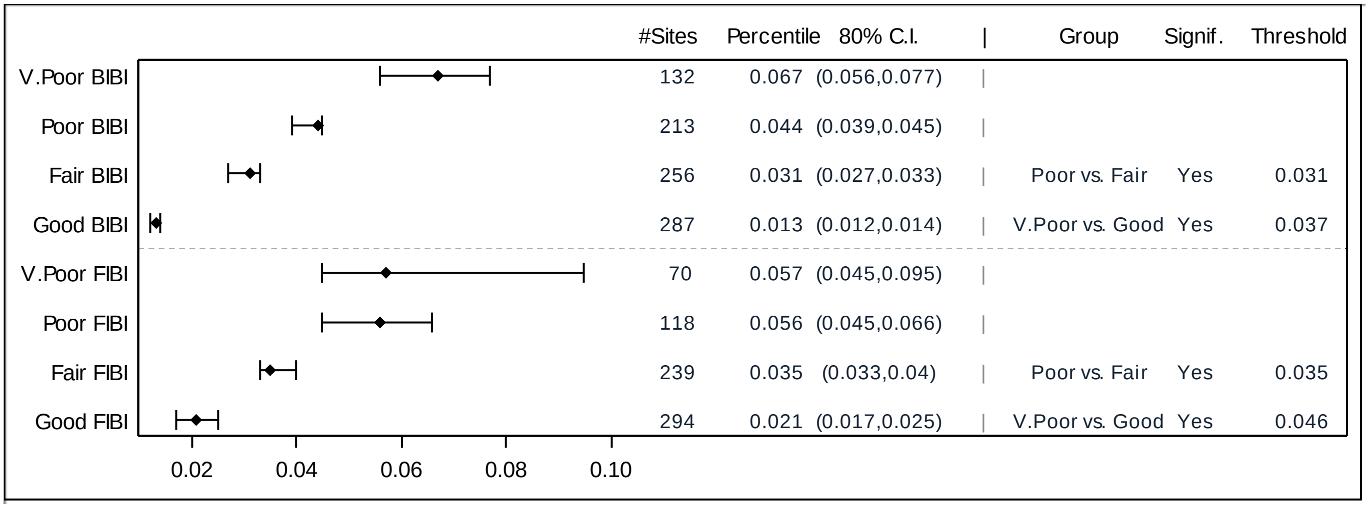
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.048	(0.042,0.063)			
Poor BIBI	244	0.033	(0.032,0.036)			
Fair BIBI	260	0.028	(0.024,0.034)	Poor vs. Fair	No	
Good BIBI	150	0.021	(0.018,0.024)	V.Poor vs. Good	Yes	0.03
V.Poor FIBI	98	0.034	(0.03,0.041)			
Poor FIBI	104	0.037	(0.032,0.04)			
Fair FIBI	204	0.032	(0.027,0.038)	Poor vs. Fair	No	
Good FIBI	277	0.033	(0.03,0.038)	V.Poor vs. Good	No	0.034

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.091	(0.079,0.107)			
Poor BIBI	132	0.076	(0.061,0.089)			
Fair BIBI	203	0.022	(0.02,0.026)	Poor vs. Fair	Yes	0.022
Good BIBI	218	0.022	(0.02,0.026)	V.Poor vs. Good	Yes	0.049
V.Poor FIBI	49	0.1	(0.088,0.119)			
Poor FIBI	69	0.085	(0.071,0.107)			
Fair FIBI	187	0.044	(0.039,0.051)	Poor vs. Fair	Yes	0.044
Good FIBI	302	0.026	(0.023,0.031)	V.Poor vs. Good	Yes	0.065

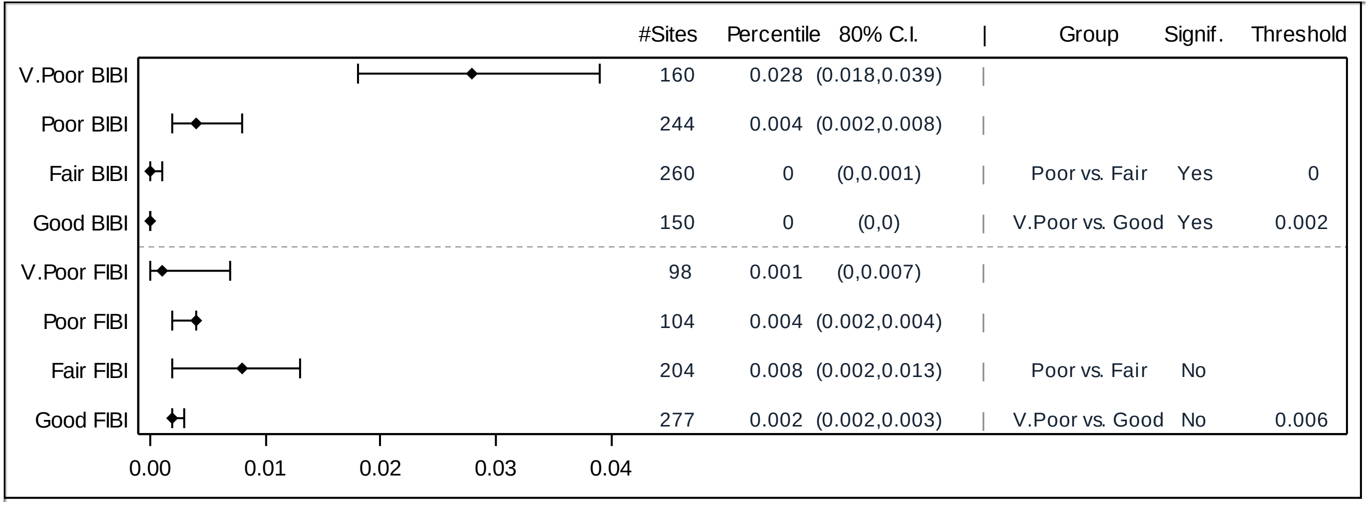
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.067	(0.056,0.077)			
Poor BIBI	213	0.044	(0.039,0.045)			
Fair BIBI	256	0.031	(0.027,0.033)	Poor vs. Fair	Yes	0.031
Good BIBI	287	0.013	(0.012,0.014)	V.Poor vs. Good	Yes	0.037
V.Poor FIBI	70	0.057	(0.045,0.095)			
Poor FIBI	118	0.056	(0.045,0.066)			
Fair FIBI	239	0.035	(0.033,0.04)	Poor vs. Fair	Yes	0.035
Good FIBI	294	0.021	(0.017,0.025)	V.Poor vs. Good	Yes	0.046

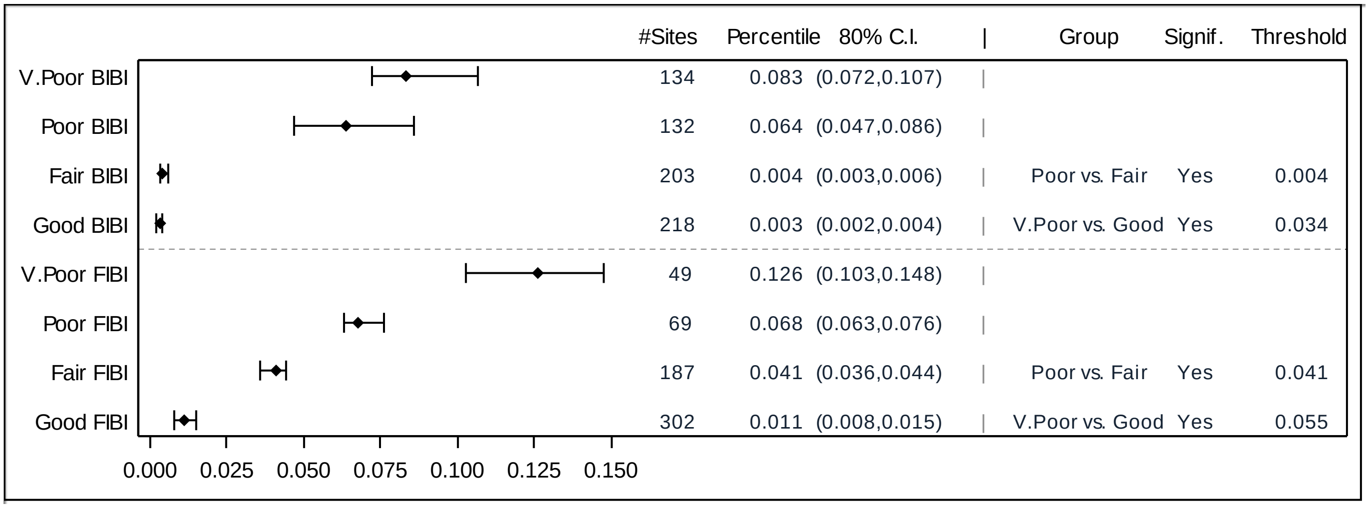
Table A-17. Physiographic Eco-region Analysis for High % of High-Intensity Developed in Watershed

Highland



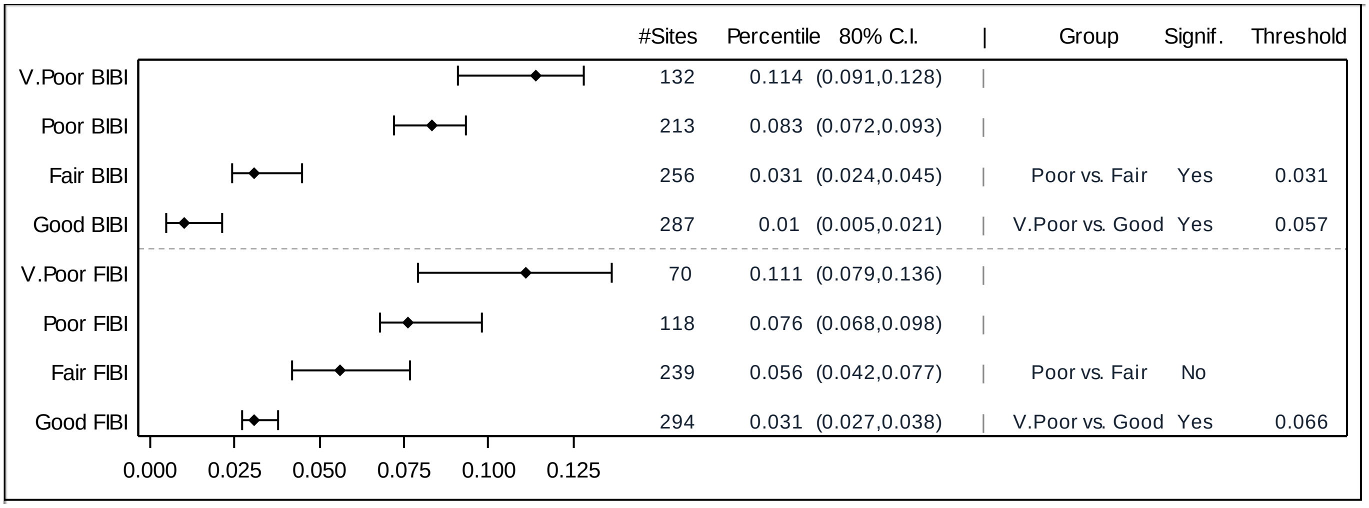
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.028	(0.018,0.039)			
Poor BIBI	244	0.004	(0.002,0.008)			
Fair BIBI	260	0	(0,0.001)	Poor vs. Fair	Yes	0
Good BIBI	150	0	(0,0)	V.Poor vs. Good	Yes	0.002
V.Poor FIBI	98	0.001	(0,0.007)			
Poor FIBI	104	0.004	(0.002,0.004)			
Fair FIBI	204	0.008	(0.002,0.013)	Poor vs. Fair	No	
Good FIBI	277	0.002	(0.002,0.003)	V.Poor vs. Good	No	0.006

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.083	(0.072,0.107)			
Poor BIBI	132	0.064	(0.047,0.086)			
Fair BIBI	203	0.004	(0.003,0.006)	Poor vs. Fair	Yes	0.004
Good BIBI	218	0.003	(0.002,0.004)	V.Poor vs. Good	Yes	0.034
V.Poor FIBI	49	0.126	(0.103,0.148)			
Poor FIBI	69	0.068	(0.063,0.076)			
Fair FIBI	187	0.041	(0.036,0.044)	Poor vs. Fair	Yes	0.041
Good FIBI	302	0.011	(0.008,0.015)	V.Poor vs. Good	Yes	0.055

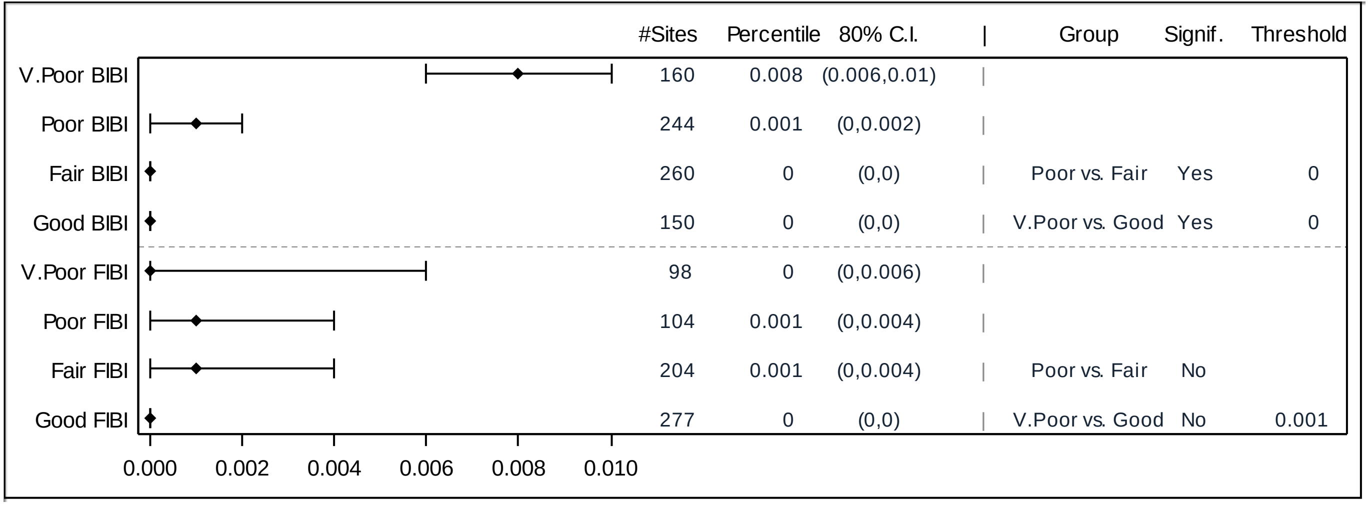
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.114	(0.091,0.128)			
Poor BIBI	213	0.083	(0.072,0.093)			
Fair BIBI	256	0.031	(0.024,0.045)	Poor vs. Fair	Yes	0.031
Good BIBI	287	0.01	(0.005,0.021)	V.Poor vs. Good	Yes	0.057
V.Poor FIBI	70	0.111	(0.079,0.136)			
Poor FIBI	118	0.076	(0.068,0.098)			
Fair FIBI	239	0.056	(0.042,0.077)	Poor vs. Fair	No	
Good FIBI	294	0.031	(0.027,0.038)	V.Poor vs. Good	Yes	0.066

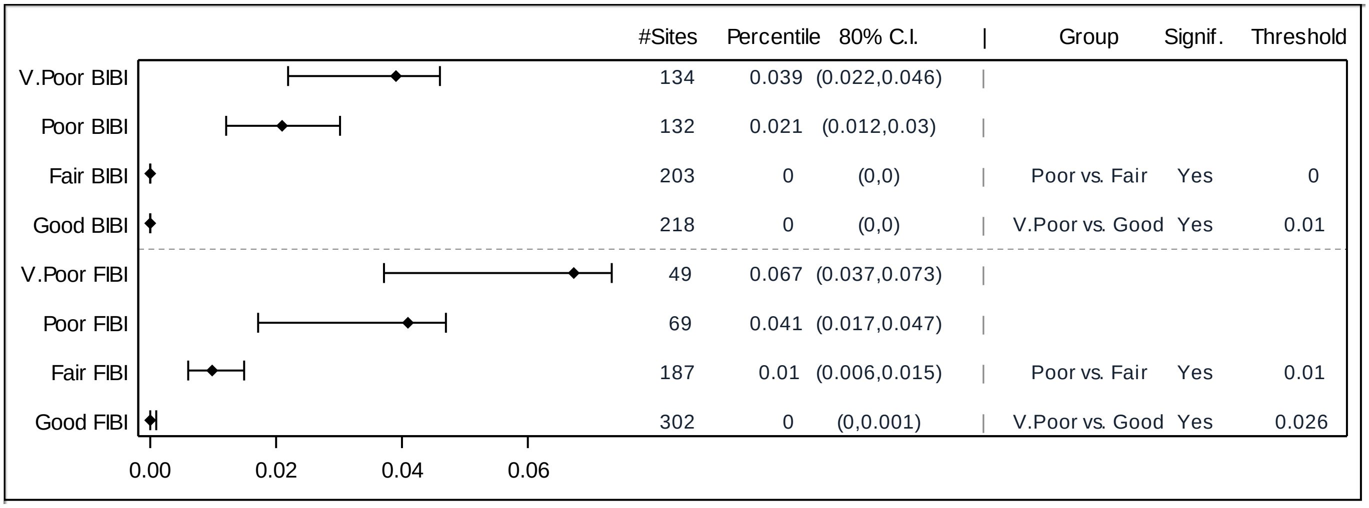
Table A-18. Physiographic Eco-region Analysis for High % of High-Intensity Developed in 60m Buffer

Highland



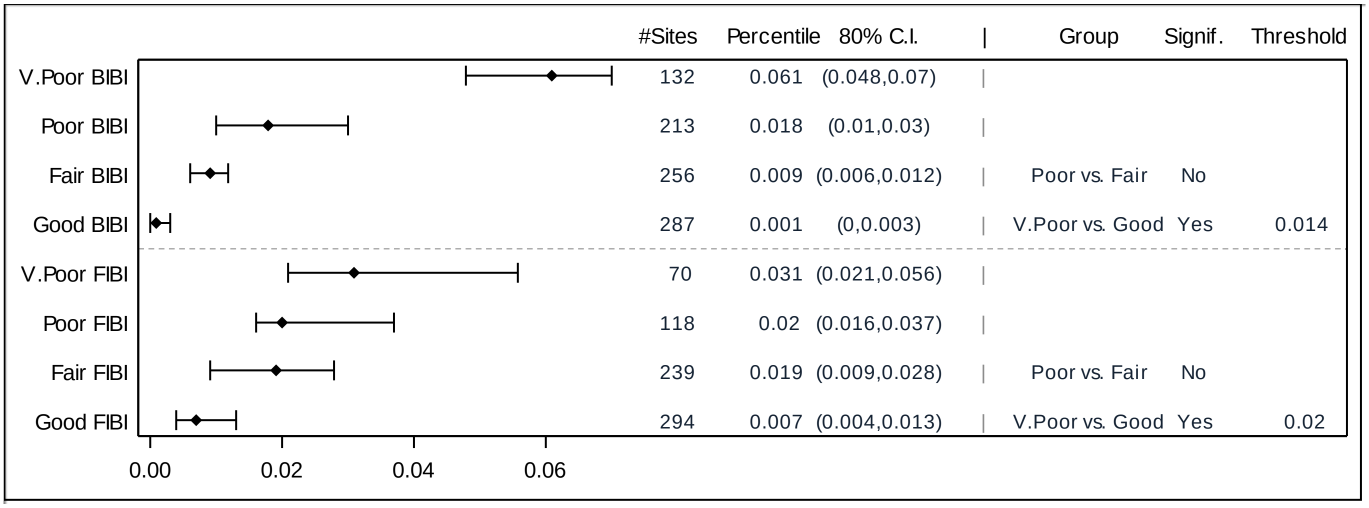
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.008	(0.006,0.01)			
Poor BIBI	244	0.001	(0,0.002)			
Fair BIBI	260	0	(0,0)	Poor vs. Fair	Yes	0
Good BIBI	150	0	(0,0)	V.Poor vs. Good	Yes	0
V.Poor FIBI	98	0	(0,0.006)			
Poor FIBI	104	0.001	(0,0.004)			
Fair FIBI	204	0.001	(0,0.004)	Poor vs. Fair	No	
Good FIBI	277	0	(0,0)	V.Poor vs. Good	No	0.001

Eastern Piedmont



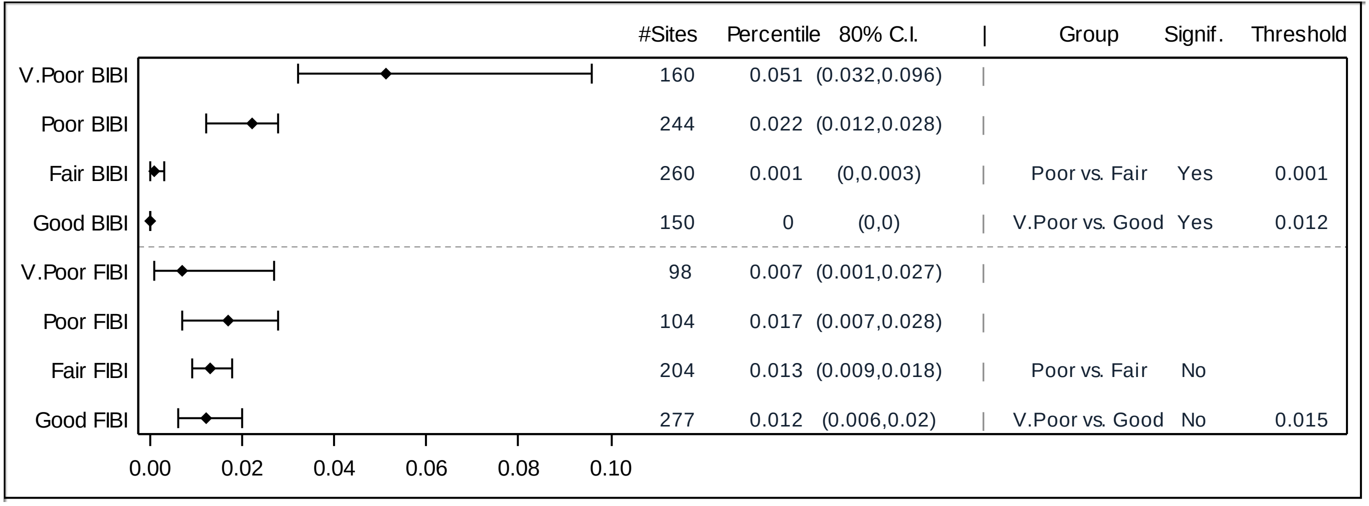
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.039	(0.022,0.046)			
Poor BIBI	132	0.021	(0.012,0.03)			
Fair BIBI	203	0	(0,0)	Poor vs. Fair	Yes	0
Good BIBI	218	0	(0,0)	V.Poor vs. Good	Yes	0.01
V.Poor FIBI	49	0.067	(0.037,0.073)			
Poor FIBI	69	0.041	(0.017,0.047)			
Fair FIBI	187	0.01	(0.006,0.015)	Poor vs. Fair	Yes	0.01
Good FIBI	302	0	(0,0.001)	V.Poor vs. Good	Yes	0.026

Coastal



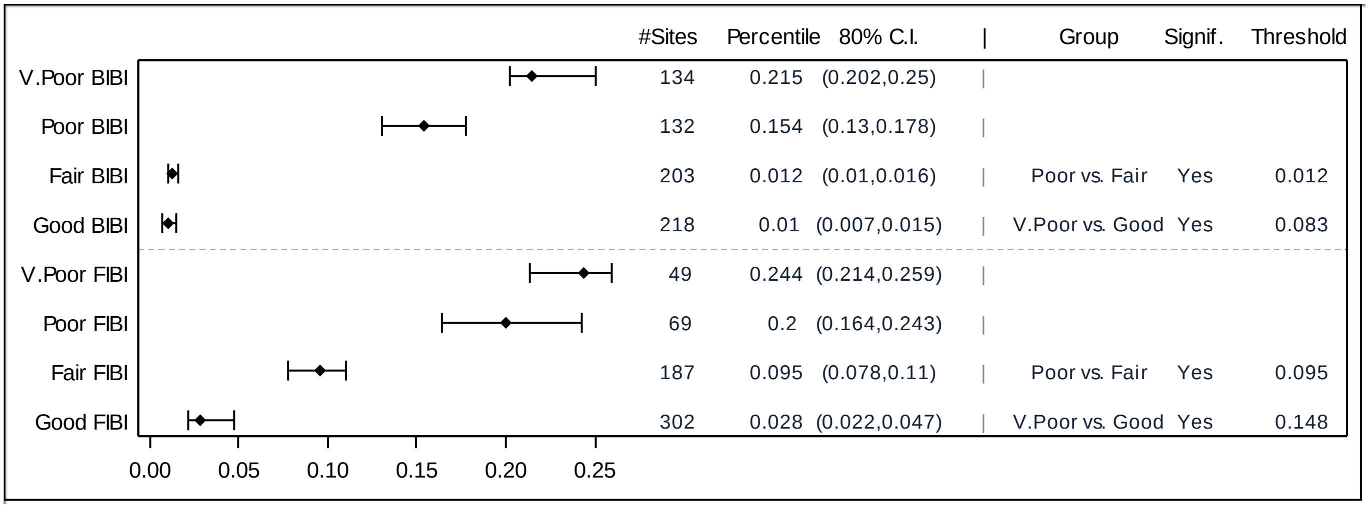
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.061	(0.048,0.07)			
Poor BIBI	213	0.018	(0.01,0.03)			
Fair BIBI	256	0.009	(0.006,0.012)	Poor vs. Fair	No	
Good BIBI	287	0.001	(0,0.003)	V.Poor vs. Good	Yes	0.014
V.Poor FIBI	70	0.031	(0.021,0.056)			
Poor FIBI	118	0.02	(0.016,0.037)			
Fair FIBI	239	0.019	(0.009,0.028)	Poor vs. Fair	No	
Good FIBI	294	0.007	(0.004,0.013)	V.Poor vs. Good	Yes	0.02

Table A-19. Physiographic Eco-region Analysis for High % of Medium-Intensity Developed in Watershed



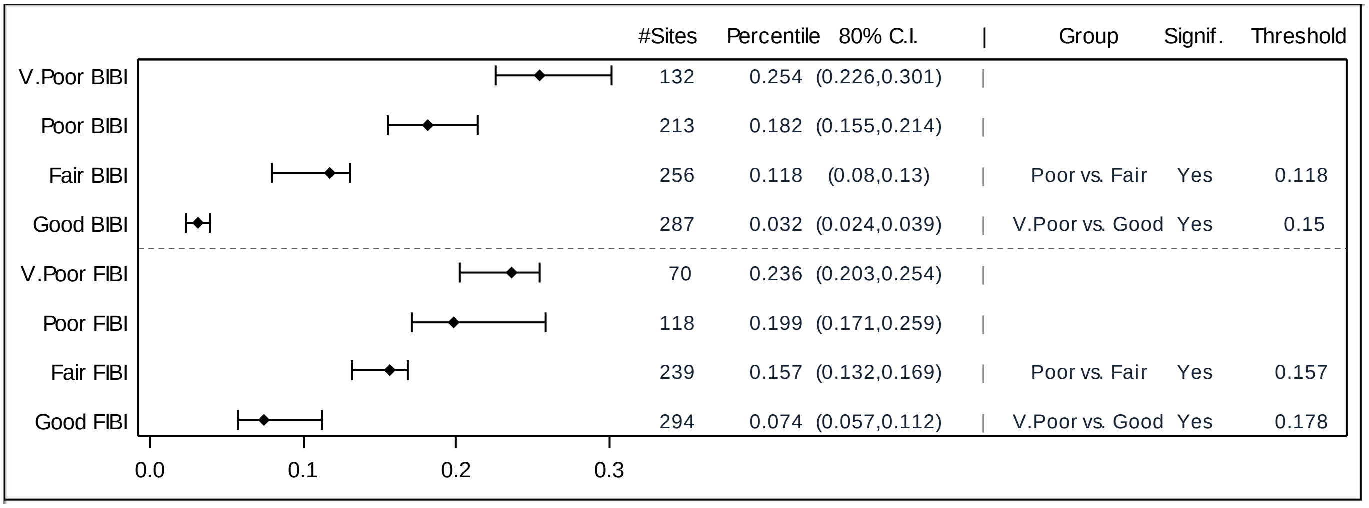
Highland

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.051	(0.032,0.096)			
Poor BIBI	244	0.022	(0.012,0.028)			
Fair BIBI	260	0.001	(0,0.003)	Poor vs. Fair	Yes	0.001
Good BIBI	150	0	(0,0)	V.Poor vs. Good	Yes	0.012
V.Poor FIBI	98	0.007	(0.001,0.027)			
Poor FIBI	104	0.017	(0.007,0.028)			
Fair FIBI	204	0.013	(0.009,0.018)	Poor vs. Fair	No	
Good FIBI	277	0.012	(0.006,0.02)	V.Poor vs. Good	No	0.015



Eastern Piedmont

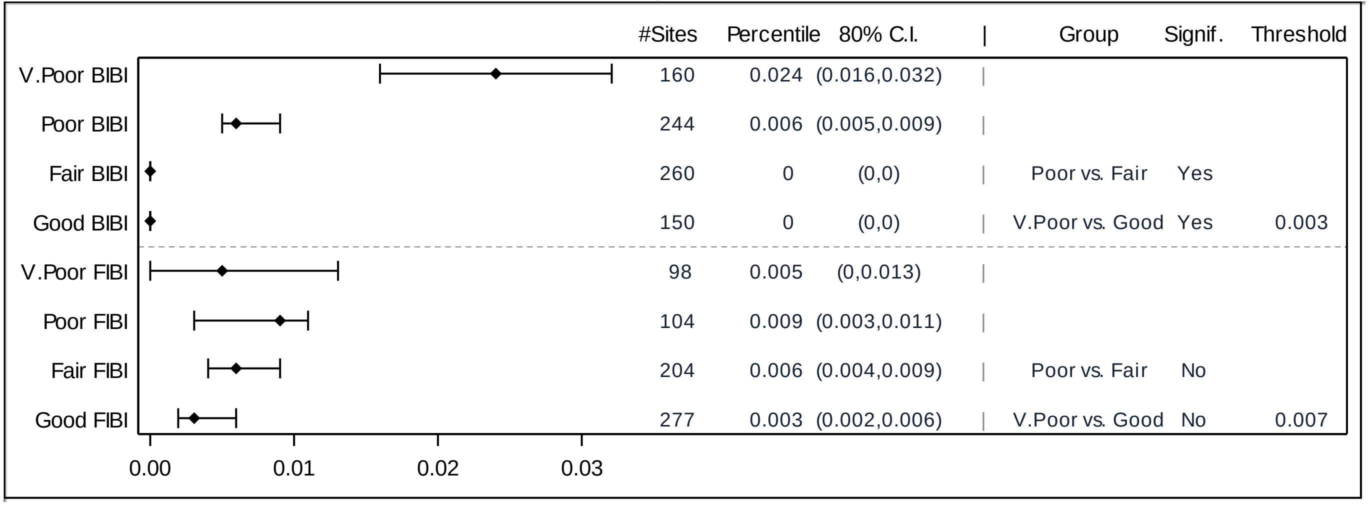
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.215	(0.202,0.25)			
Poor BIBI	132	0.154	(0.13,0.178)			
Fair BIBI	203	0.012	(0.01,0.016)	Poor vs. Fair	Yes	0.012
Good BIBI	218	0.01	(0.007,0.015)	V.Poor vs. Good	Yes	0.083
V.Poor FIBI	49	0.244	(0.214,0.259)			
Poor FIBI	69	0.2	(0.164,0.243)			
Fair FIBI	187	0.095	(0.078,0.11)	Poor vs. Fair	Yes	0.095
Good FIBI	302	0.028	(0.022,0.047)	V.Poor vs. Good	Yes	0.148



Coastal

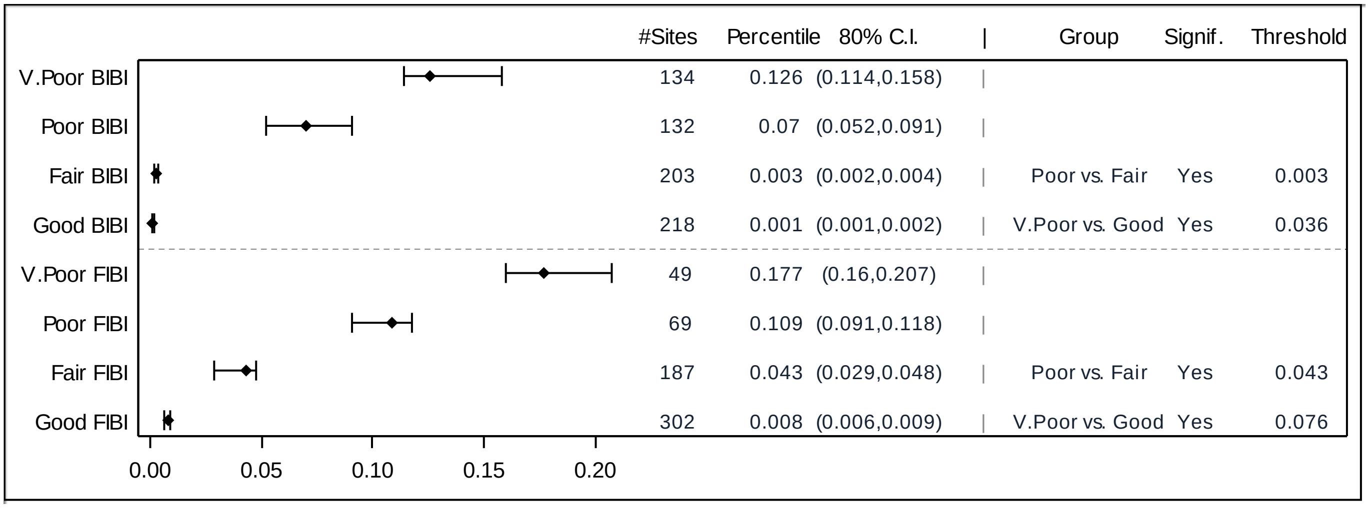
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.254	(0.226,0.301)			
Poor BIBI	213	0.182	(0.155,0.214)			
Fair BIBI	256	0.118	(0.08,0.13)	Poor vs. Fair	Yes	0.118
Good BIBI	287	0.032	(0.024,0.039)	V.Poor vs. Good	Yes	0.15
V.Poor FIBI	70	0.236	(0.203,0.254)			
Poor FIBI	118	0.199	(0.171,0.259)			
Fair FIBI	239	0.157	(0.132,0.169)	Poor vs. Fair	Yes	0.157
Good FIBI	294	0.074	(0.057,0.112)	V.Poor vs. Good	Yes	0.178

Table A-20. Physiographic Eco-region Analysis for High % of Medium-Intensity Developed in 60m Buffer



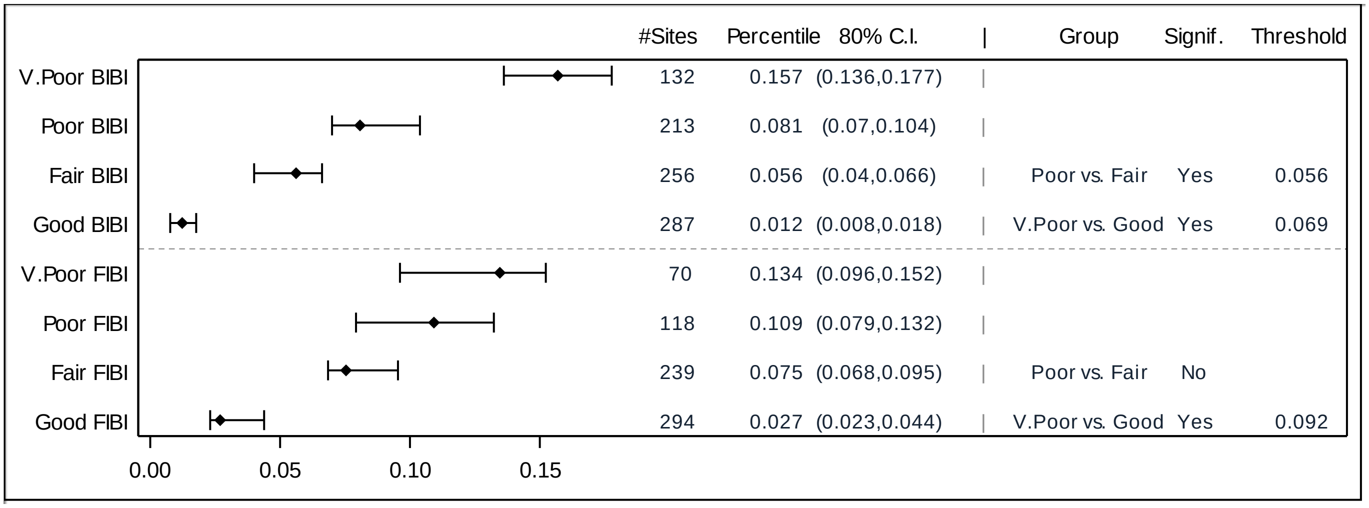
Highland

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.024	(0.016,0.032)			
Poor BIBI	244	0.006	(0.005,0.009)			
Fair BIBI	260	0	(0,0)	Poor vs. Fair	Yes	
Good BIBI	150	0	(0,0)	V.Poor vs. Good	Yes	0.003
V.Poor FIBI	98	0.005	(0,0.013)			
Poor FIBI	104	0.009	(0.003,0.011)			
Fair FIBI	204	0.006	(0.004,0.009)	Poor vs. Fair	No	
Good FIBI	277	0.003	(0.002,0.006)	V.Poor vs. Good	No	0.007



Eastern Piedmont

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.126	(0.114,0.158)			
Poor BIBI	132	0.07	(0.052,0.091)			
Fair BIBI	203	0.003	(0.002,0.004)	Poor vs. Fair	Yes	0.003
Good BIBI	218	0.001	(0.001,0.002)	V.Poor vs. Good	Yes	0.036
V.Poor FIBI	49	0.177	(0.16,0.207)			
Poor FIBI	69	0.109	(0.091,0.118)			
Fair FIBI	187	0.043	(0.029,0.048)	Poor vs. Fair	Yes	0.043
Good FIBI	302	0.008	(0.006,0.009)	V.Poor vs. Good	Yes	0.076

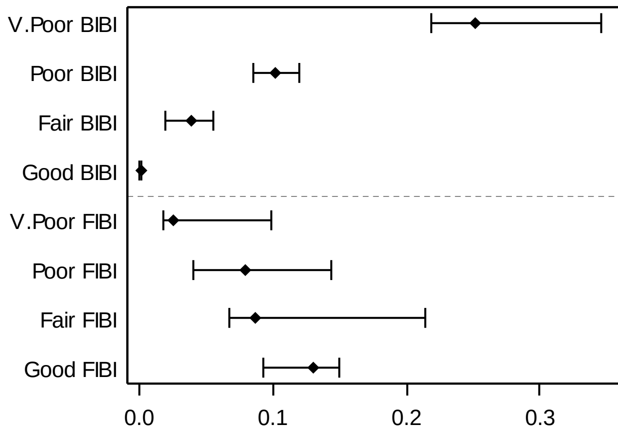


Coastal

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.157	(0.136,0.177)			
Poor BIBI	213	0.081	(0.07,0.104)			
Fair BIBI	256	0.056	(0.04,0.066)	Poor vs. Fair	Yes	0.056
Good BIBI	287	0.012	(0.008,0.018)	V.Poor vs. Good	Yes	0.069
V.Poor FIBI	70	0.134	(0.096,0.152)			
Poor FIBI	118	0.109	(0.079,0.132)			
Fair FIBI	239	0.075	(0.068,0.095)	Poor vs. Fair	No	
Good FIBI	294	0.027	(0.023,0.044)	V.Poor vs. Good	Yes	0.092

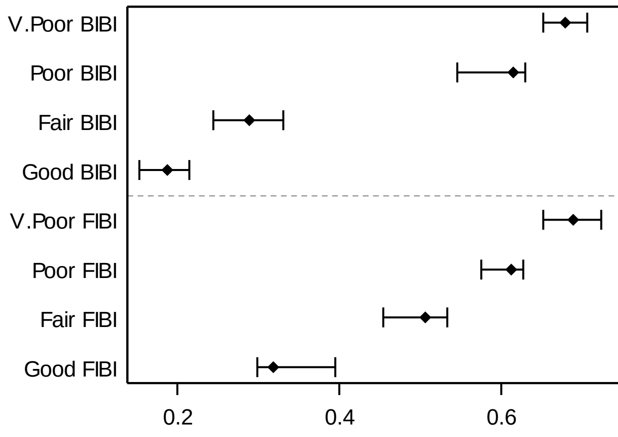
Table A-21. Physiographic Eco-region Analysis for High % of Low-Intensity Developed in Watershed

Highland



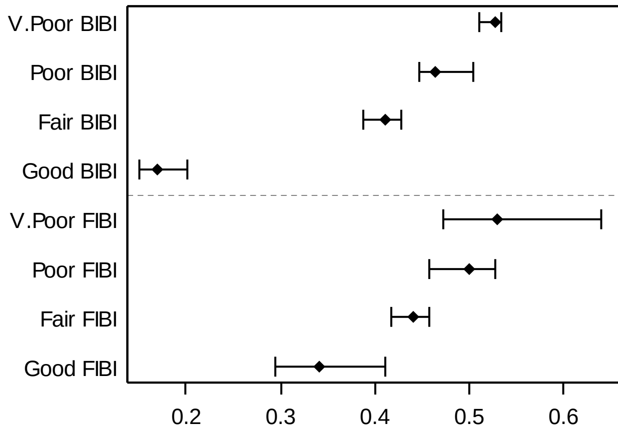
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.252	(0.218,0.345)			
Poor BIBI	244	0.102	(0.085,0.12)			
Fair BIBI	260	0.039	(0.02,0.055)	Poor vs. Fair	Yes	0.039
Good BIBI	150	0.001	(0,0.002)	V.Poor vs. Good	Yes	0.071
V.Poor FIBI	98	0.026	(0.018,0.099)			
Poor FIBI	104	0.079	(0.041,0.144)			
Fair FIBI	204	0.087	(0.067,0.214)	Poor vs. Fair	No	
Good FIBI	277	0.13	(0.093,0.149)	V.Poor vs. Good	No	0.083

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.679	(0.651,0.707)			
Poor BIBI	132	0.615	(0.545,0.628)			
Fair BIBI	203	0.287	(0.243,0.329)	Poor vs. Fair	Yes	0.287
Good BIBI	218	0.188	(0.152,0.215)	V.Poor vs. Good	Yes	0.451
V.Poor FIBI	49	0.688	(0.651,0.722)			
Poor FIBI	69	0.612	(0.574,0.626)			
Fair FIBI	187	0.505	(0.454,0.532)	Poor vs. Fair	Yes	0.505
Good FIBI	302	0.317	(0.297,0.394)	V.Poor vs. Good	Yes	0.559

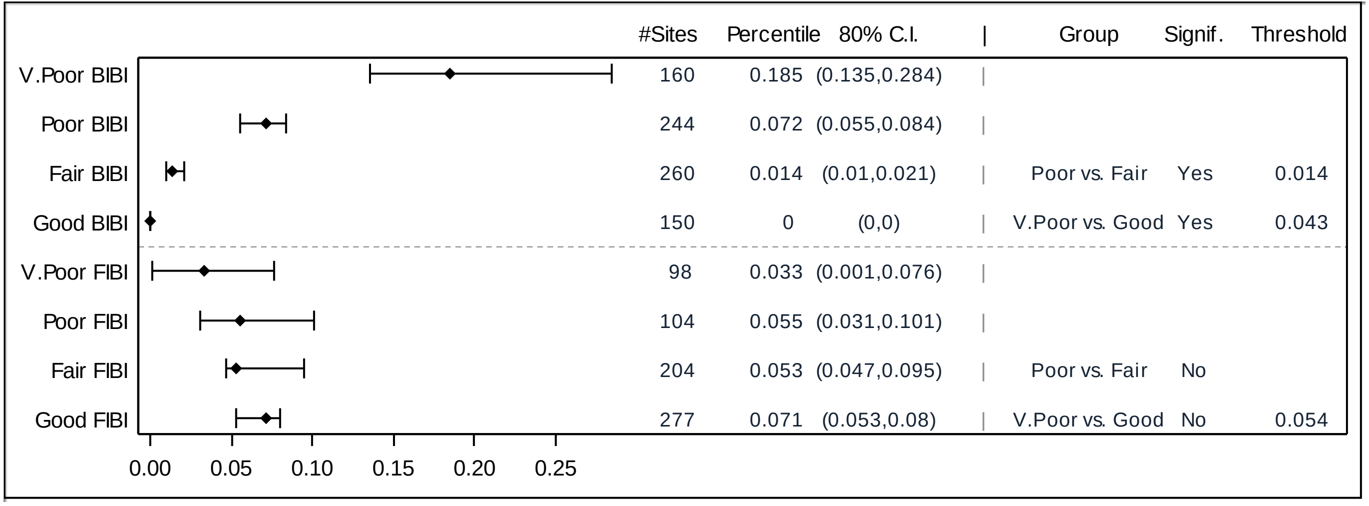
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.527	(0.511,0.534)			
Poor BIBI	213	0.463	(0.446,0.504)			
Fair BIBI	256	0.411	(0.387,0.427)	Poor vs. Fair	Yes	0.411
Good BIBI	287	0.169	(0.15,0.202)	V.Poor vs. Good	Yes	0.437
V.Poor FIBI	70	0.529	(0.473,0.639)			
Poor FIBI	118	0.5	(0.457,0.528)			
Fair FIBI	239	0.441	(0.418,0.458)	Poor vs. Fair	No	
Good FIBI	294	0.34	(0.294,0.411)	V.Poor vs. Good	Yes	0.47

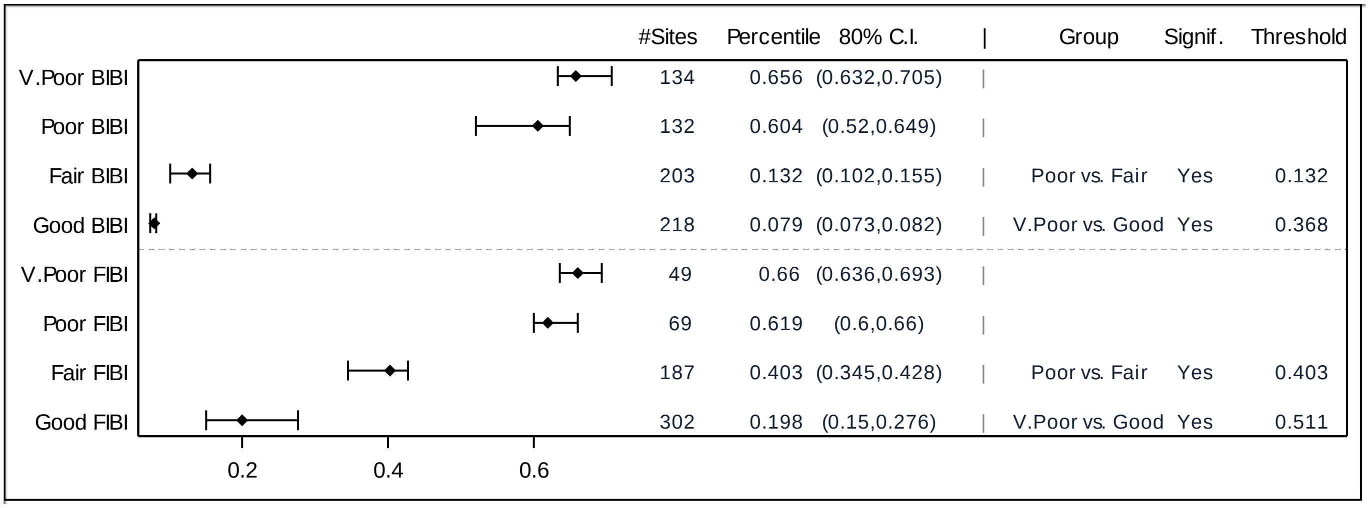
Table A-22. Physiographic Eco-region Analysis for High % of Low-Intensity Developed in 60m Buffer

Highland



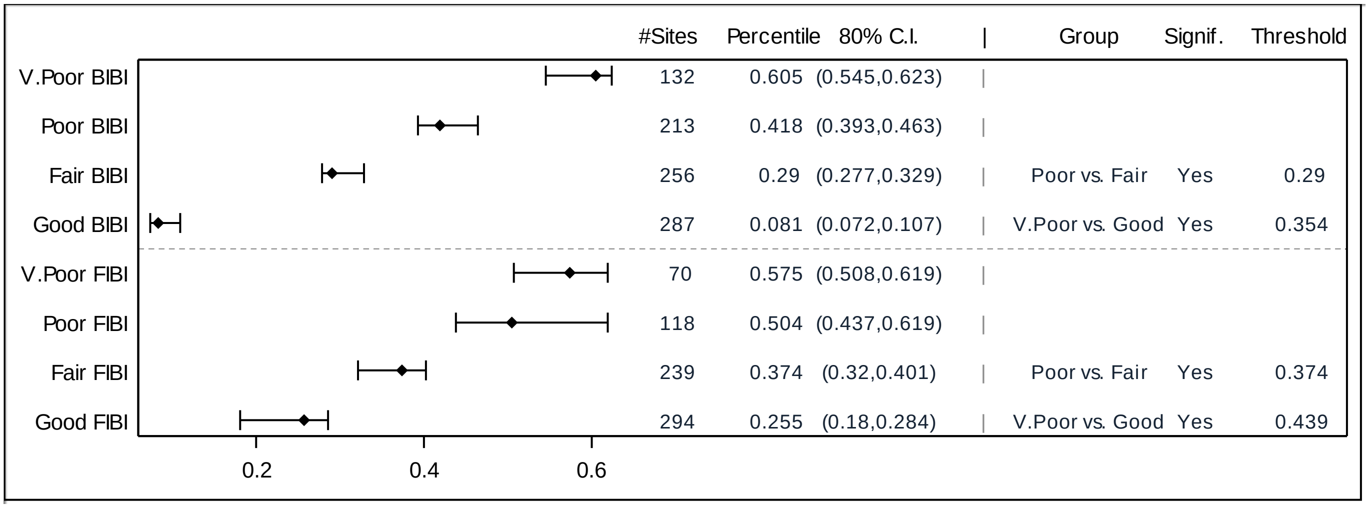
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.185	(0.135,0.284)			
Poor BIBI	244	0.072	(0.055,0.084)			
Fair BIBI	260	0.014	(0.01,0.021)	Poor vs. Fair	Yes	0.014
Good BIBI	150	0	(0,0)	V.Poor vs. Good	Yes	0.043
V.Poor FIBI	98	0.033	(0.001,0.076)			
Poor FIBI	104	0.055	(0.031,0.101)			
Fair FIBI	204	0.053	(0.047,0.095)	Poor vs. Fair	No	
Good FIBI	277	0.071	(0.053,0.08)	V.Poor vs. Good	No	0.054

Eastern Piedmont



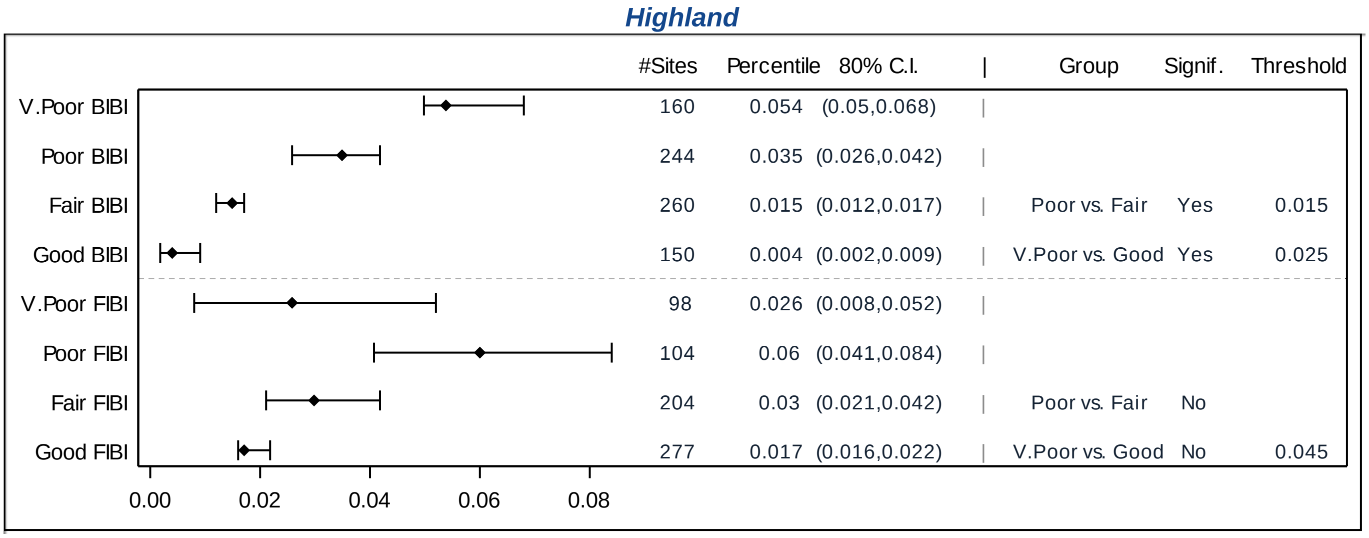
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.656	(0.632,0.705)			
Poor BIBI	132	0.604	(0.52,0.649)			
Fair BIBI	203	0.132	(0.102,0.155)	Poor vs. Fair	Yes	0.132
Good BIBI	218	0.079	(0.073,0.082)	V.Poor vs. Good	Yes	0.368
V.Poor FIBI	49	0.66	(0.636,0.693)			
Poor FIBI	69	0.619	(0.6,0.66)			
Fair FIBI	187	0.403	(0.345,0.428)	Poor vs. Fair	Yes	0.403
Good FIBI	302	0.198	(0.15,0.276)	V.Poor vs. Good	Yes	0.511

Coastal



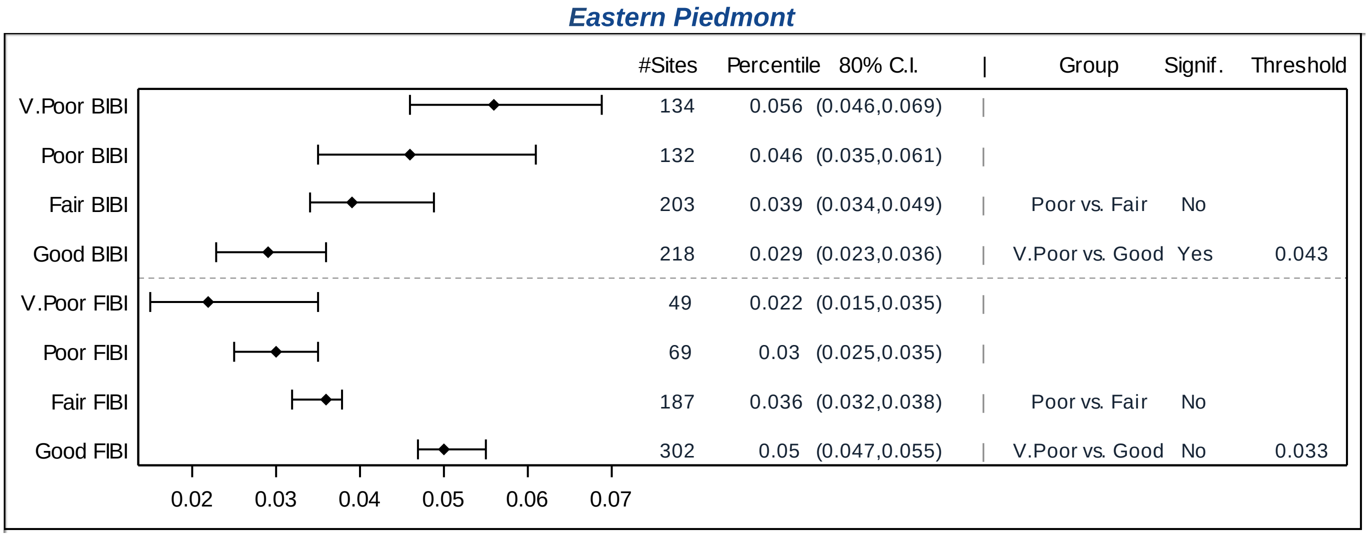
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.605	(0.545,0.623)			
Poor BIBI	213	0.418	(0.393,0.463)			
Fair BIBI	256	0.29	(0.277,0.329)	Poor vs. Fair	Yes	0.29
Good BIBI	287	0.081	(0.072,0.107)	V.Poor vs. Good	Yes	0.354
V.Poor FIBI	70	0.575	(0.508,0.619)			
Poor FIBI	118	0.504	(0.437,0.619)			
Fair FIBI	239	0.374	(0.32,0.401)	Poor vs. Fair	Yes	0.374
Good FIBI	294	0.255	(0.18,0.284)	V.Poor vs. Good	Yes	0.439

Table A-23. Physiographic Eco-region Analysis for High % of Residential Developed in Watershed



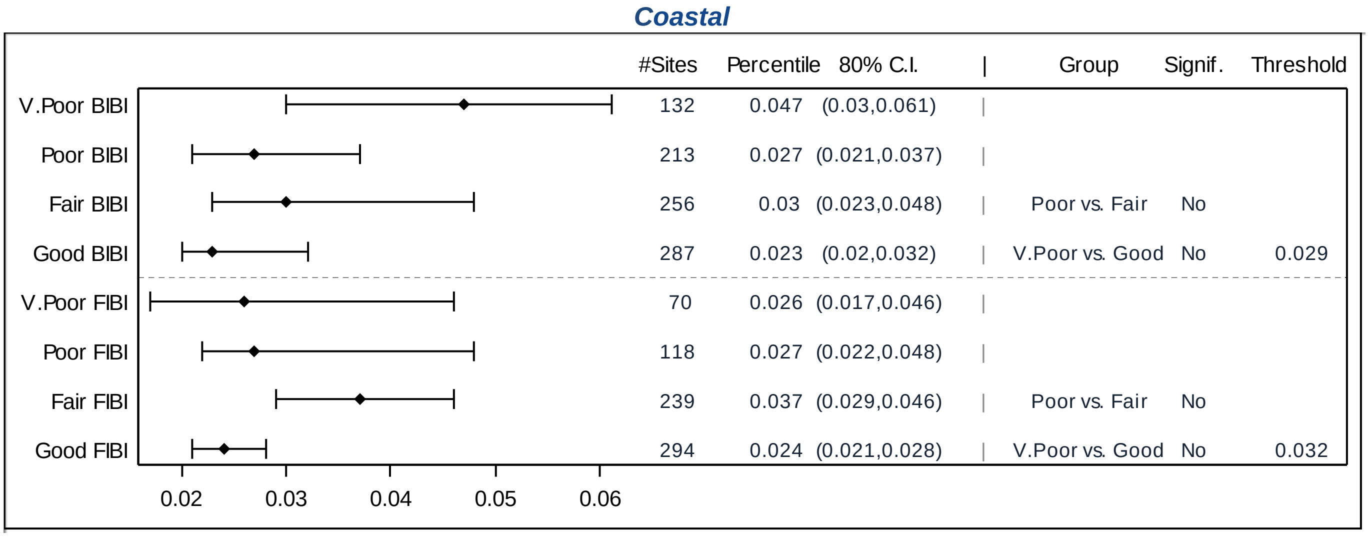
Highland

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.054	(0.05,0.068)			
Poor BIBI	244	0.035	(0.026,0.042)			
Fair BIBI	260	0.015	(0.012,0.017)	Poor vs. Fair	Yes	0.015
Good BIBI	150	0.004	(0.002,0.009)	V.Poor vs. Good	Yes	0.025
V.Poor FIBI	98	0.026	(0.008,0.052)			
Poor FIBI	104	0.06	(0.041,0.084)			
Fair FIBI	204	0.03	(0.021,0.042)	Poor vs. Fair	No	
Good FIBI	277	0.017	(0.016,0.022)	V.Poor vs. Good	No	0.045



Eastern Piedmont

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.056	(0.046,0.069)			
Poor BIBI	132	0.046	(0.035,0.061)			
Fair BIBI	203	0.039	(0.034,0.049)	Poor vs. Fair	No	
Good BIBI	218	0.029	(0.023,0.036)	V.Poor vs. Good	Yes	0.043
V.Poor FIBI	49	0.022	(0.015,0.035)			
Poor FIBI	69	0.03	(0.025,0.035)			
Fair FIBI	187	0.036	(0.032,0.038)	Poor vs. Fair	No	
Good FIBI	302	0.05	(0.047,0.055)	V.Poor vs. Good	No	0.033

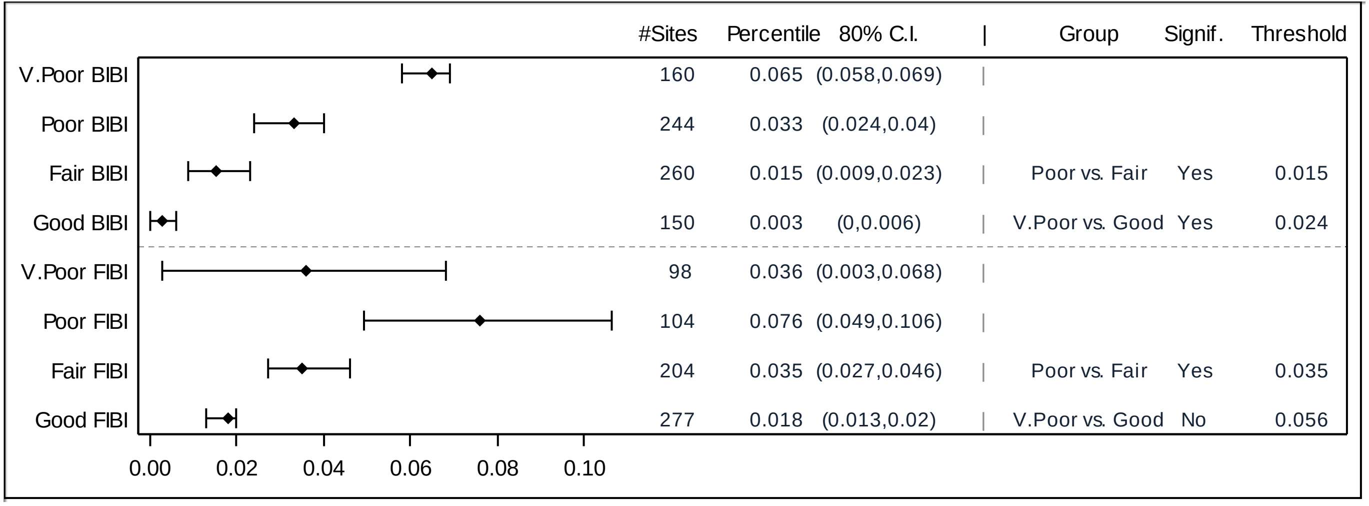


Coastal

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.047	(0.03,0.061)			
Poor BIBI	213	0.027	(0.021,0.037)			
Fair BIBI	256	0.03	(0.023,0.048)	Poor vs. Fair	No	
Good BIBI	287	0.023	(0.02,0.032)	V.Poor vs. Good	No	0.029
V.Poor FIBI	70	0.026	(0.017,0.046)			
Poor FIBI	118	0.027	(0.022,0.048)			
Fair FIBI	239	0.037	(0.029,0.046)	Poor vs. Fair	No	
Good FIBI	294	0.024	(0.021,0.028)	V.Poor vs. Good	No	0.032

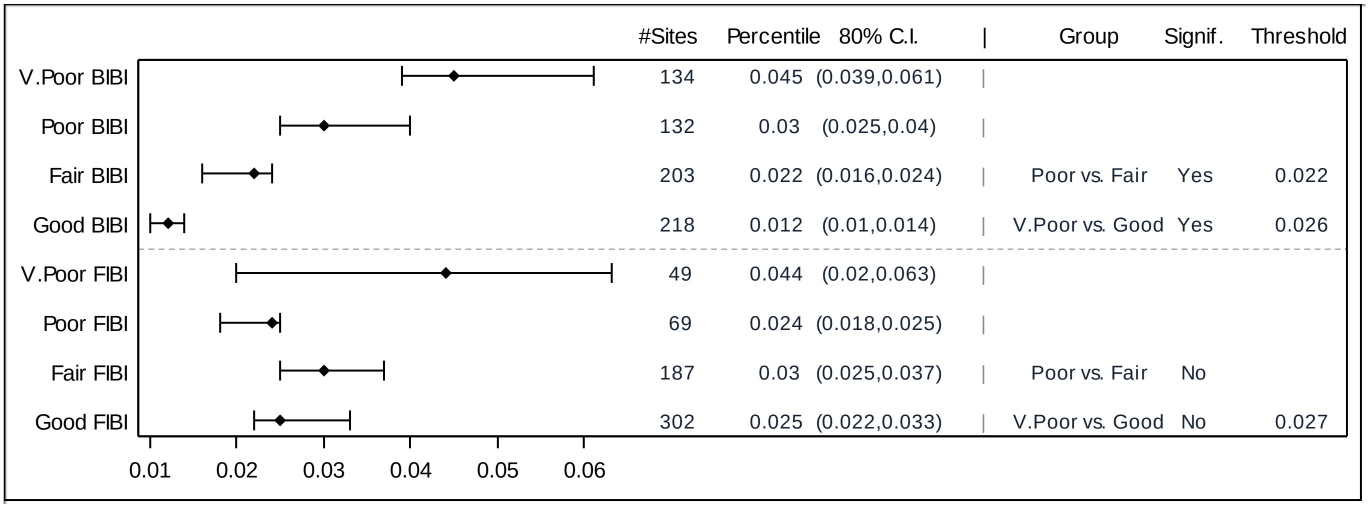
Table A-24. Physiographic Eco-region Analysis for High % of Residential Developed in 60m Buffer

Highland



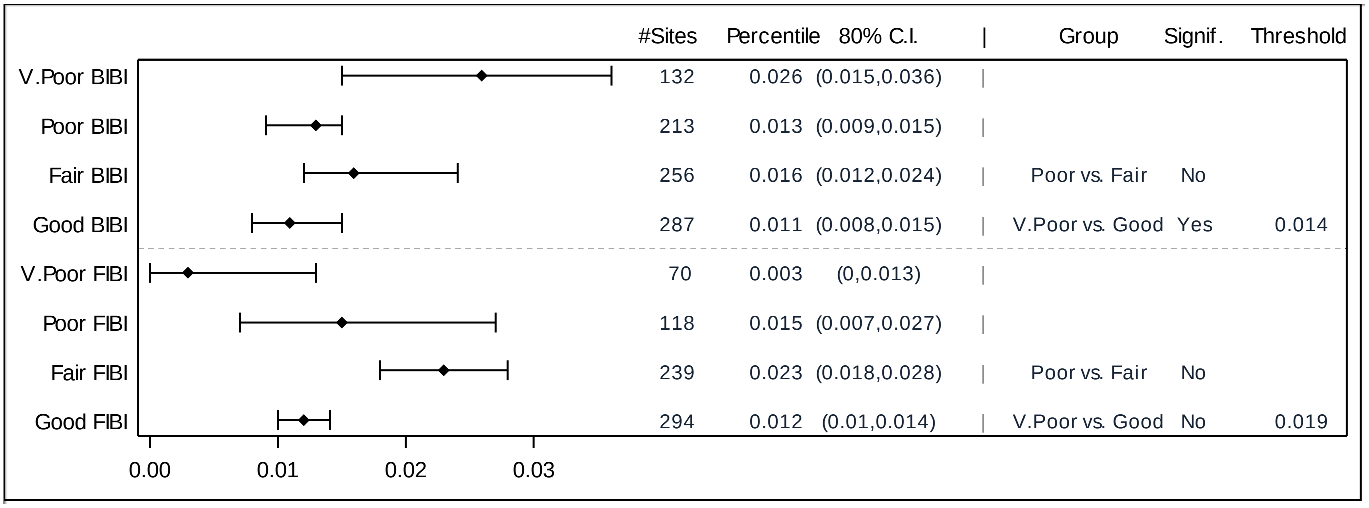
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.065	(0.058,0.069)			
Poor BIBI	244	0.033	(0.024,0.04)			
Fair BIBI	260	0.015	(0.009,0.023)	Poor vs. Fair	Yes	0.015
Good BIBI	150	0.003	(0,0.006)	V.Poor vs. Good	Yes	0.024
V.Poor FIBI	98	0.036	(0.003,0.068)			
Poor FIBI	104	0.076	(0.049,0.106)			
Fair FIBI	204	0.035	(0.027,0.046)	Poor vs. Fair	Yes	0.035
Good FIBI	277	0.018	(0.013,0.02)	V.Poor vs. Good	No	0.056

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.045	(0.039,0.061)			
Poor BIBI	132	0.03	(0.025,0.04)			
Fair BIBI	203	0.022	(0.016,0.024)	Poor vs. Fair	Yes	0.022
Good BIBI	218	0.012	(0.01,0.014)	V.Poor vs. Good	Yes	0.026
V.Poor FIBI	49	0.044	(0.02,0.063)			
Poor FIBI	69	0.024	(0.018,0.025)			
Fair FIBI	187	0.03	(0.025,0.037)	Poor vs. Fair	No	
Good FIBI	302	0.025	(0.022,0.033)	V.Poor vs. Good	No	0.027

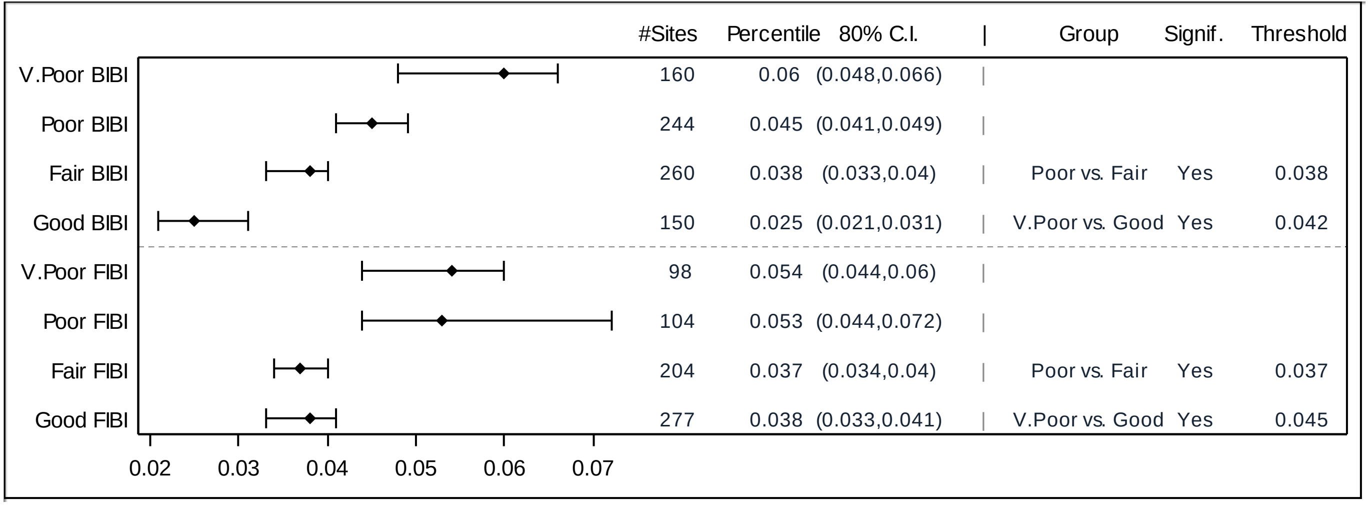
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.026	(0.015,0.036)			
Poor BIBI	213	0.013	(0.009,0.015)			
Fair BIBI	256	0.016	(0.012,0.024)	Poor vs. Fair	No	
Good BIBI	287	0.011	(0.008,0.015)	V.Poor vs. Good	Yes	0.014
V.Poor FIBI	70	0.003	(0,0.013)			
Poor FIBI	118	0.015	(0.007,0.027)			
Fair FIBI	239	0.023	(0.018,0.028)	Poor vs. Fair	No	
Good FIBI	294	0.012	(0.01,0.014)	V.Poor vs. Good	No	0.019

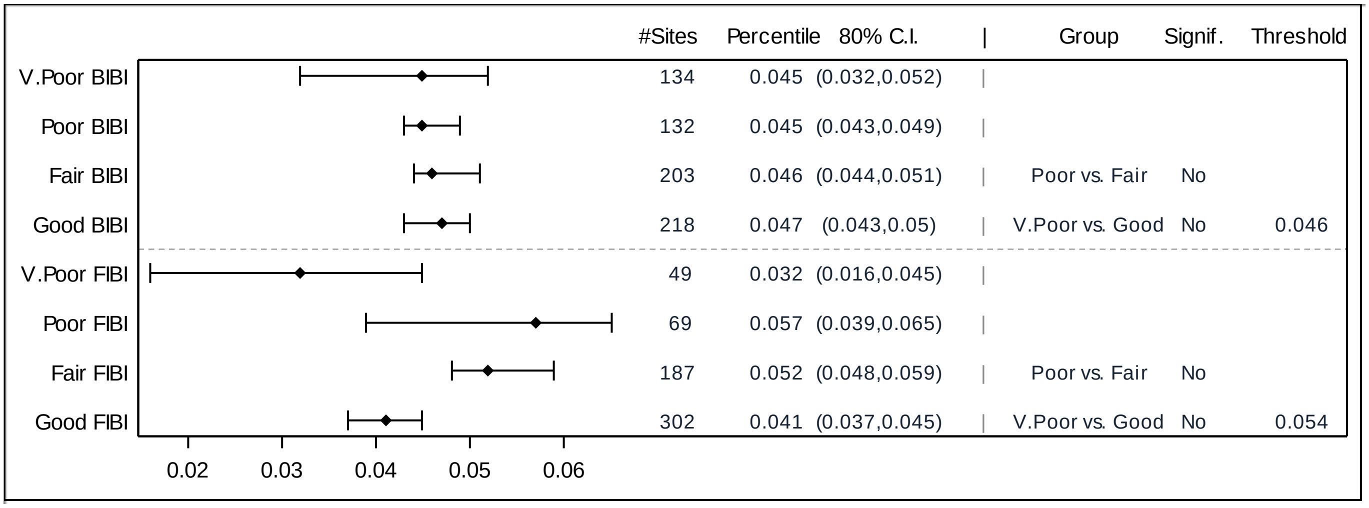
Table A-25. Physiographic Eco-region Analysis for High % of Rural Developed in Watershed

Highland



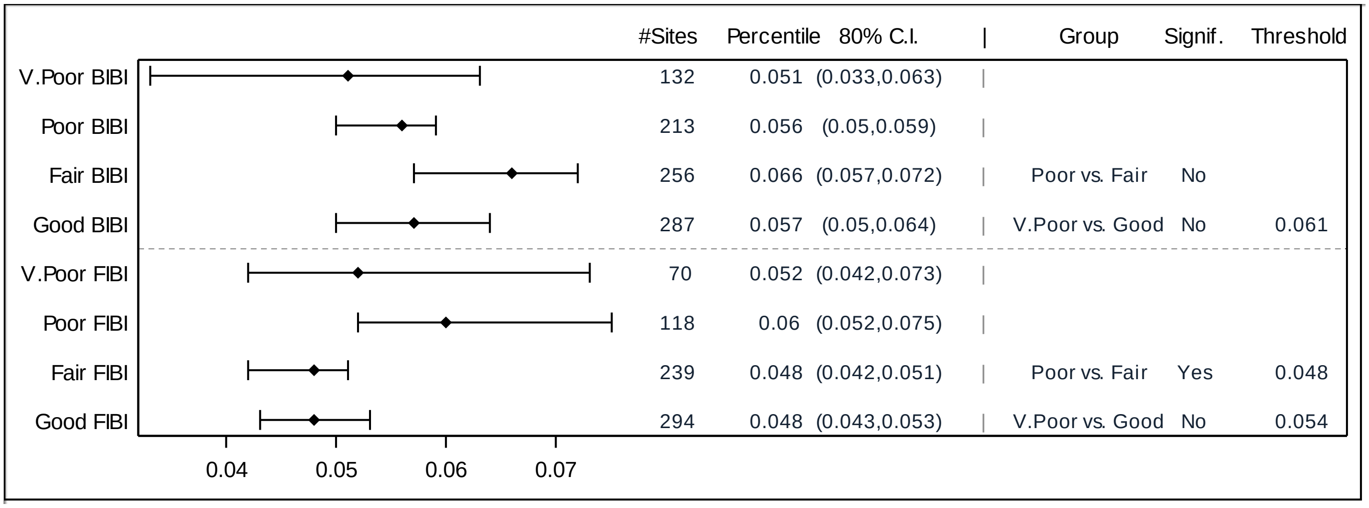
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.06	(0.048,0.066)			
Poor BIBI	244	0.045	(0.041,0.049)			
Fair BIBI	260	0.038	(0.033,0.04)	Poor vs. Fair	Yes	0.038
Good BIBI	150	0.025	(0.021,0.031)	V.Poor vs. Good	Yes	0.042
V.Poor FIBI	98	0.054	(0.044,0.06)			
Poor FIBI	104	0.053	(0.044,0.072)			
Fair FIBI	204	0.037	(0.034,0.04)	Poor vs. Fair	Yes	0.037
Good FIBI	277	0.038	(0.033,0.041)	V.Poor vs. Good	Yes	0.045

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.045	(0.032,0.052)			
Poor BIBI	132	0.045	(0.043,0.049)			
Fair BIBI	203	0.046	(0.044,0.051)	Poor vs. Fair	No	
Good BIBI	218	0.047	(0.043,0.05)	V.Poor vs. Good	No	0.046
V.Poor FIBI	49	0.032	(0.016,0.045)			
Poor FIBI	69	0.057	(0.039,0.065)			
Fair FIBI	187	0.052	(0.048,0.059)	Poor vs. Fair	No	
Good FIBI	302	0.041	(0.037,0.045)	V.Poor vs. Good	No	0.054

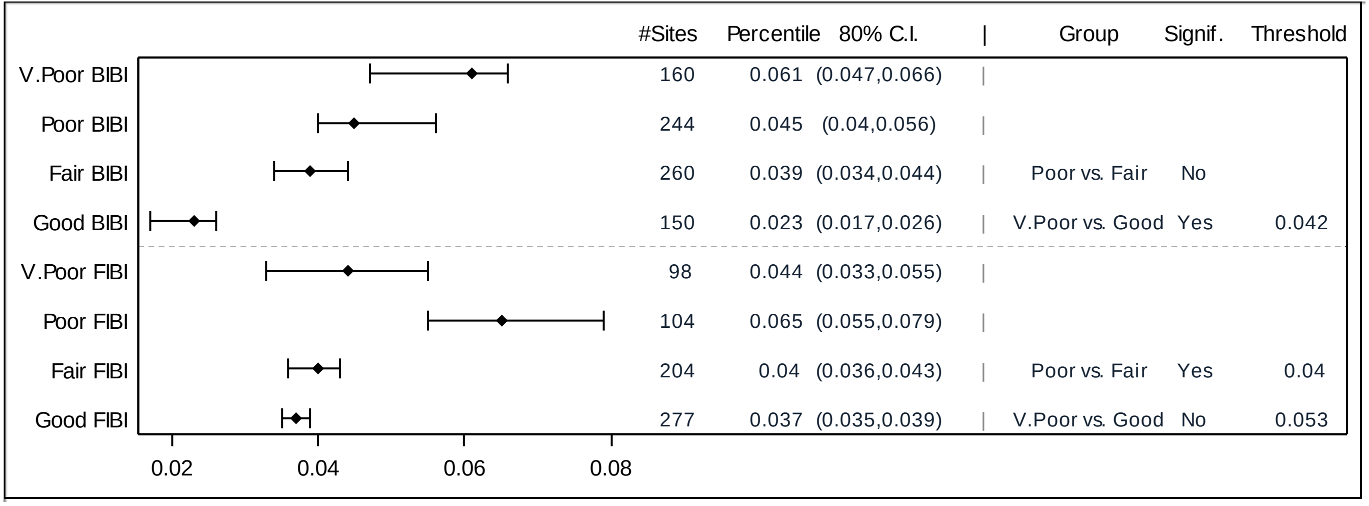
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.051	(0.033,0.063)			
Poor BIBI	213	0.056	(0.05,0.059)			
Fair BIBI	256	0.066	(0.057,0.072)	Poor vs. Fair	No	
Good BIBI	287	0.057	(0.05,0.064)	V.Poor vs. Good	No	0.061
V.Poor FIBI	70	0.052	(0.042,0.073)			
Poor FIBI	118	0.06	(0.052,0.075)			
Fair FIBI	239	0.048	(0.042,0.051)	Poor vs. Fair	Yes	0.048
Good FIBI	294	0.048	(0.043,0.053)	V.Poor vs. Good	No	0.054

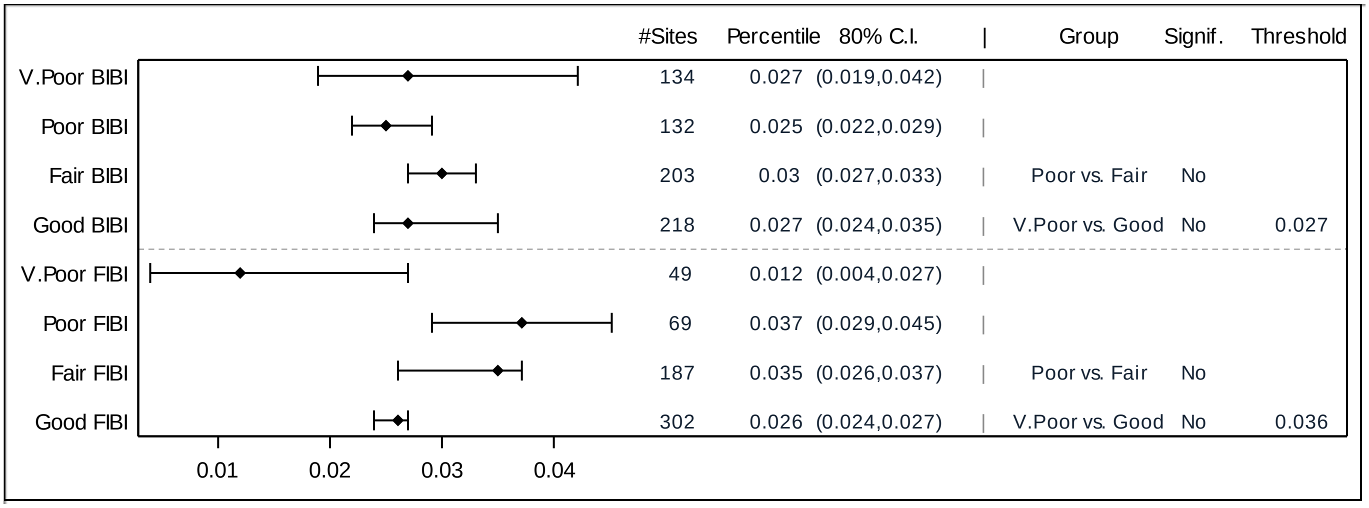
Table A-26. Physiographic Eco-region Analysis for High % of Rural Developed in 60m Buffer

Highland



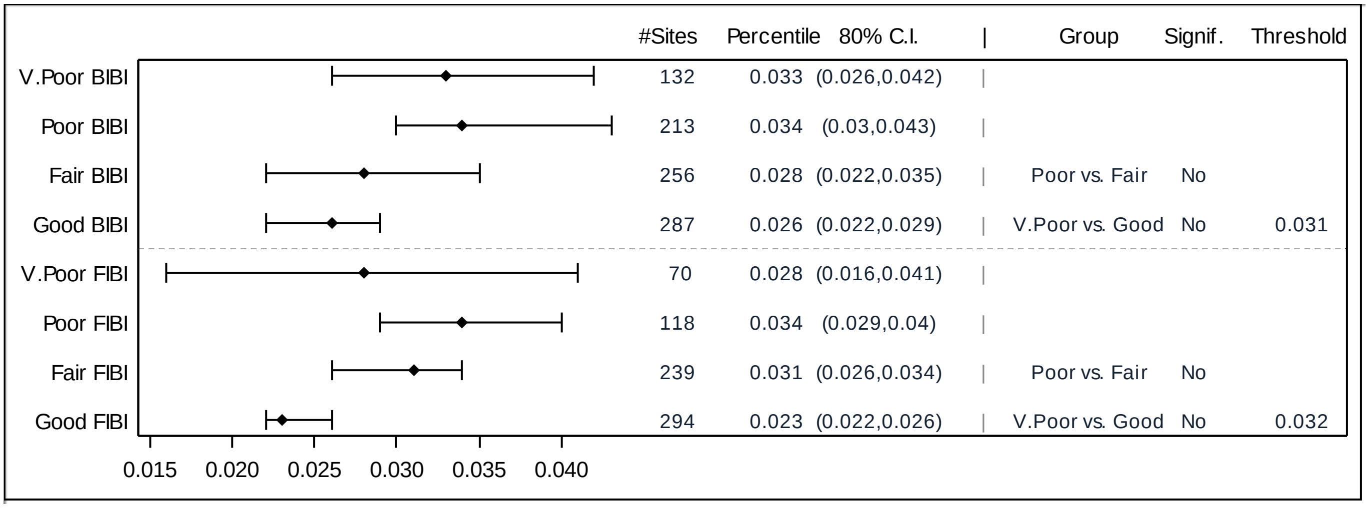
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.061	(0.047,0.066)			
Poor BIBI	244	0.045	(0.04,0.056)			
Fair BIBI	260	0.039	(0.034,0.044)	Poor vs. Fair	No	
Good BIBI	150	0.023	(0.017,0.026)	V.Poor vs. Good	Yes	0.042
V.Poor FIBI	98	0.044	(0.033,0.055)			
Poor FIBI	104	0.065	(0.055,0.079)			
Fair FIBI	204	0.04	(0.036,0.043)	Poor vs. Fair	Yes	0.04
Good FIBI	277	0.037	(0.035,0.039)	V.Poor vs. Good	No	0.053

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.027	(0.019,0.042)			
Poor BIBI	132	0.025	(0.022,0.029)			
Fair BIBI	203	0.03	(0.027,0.033)	Poor vs. Fair	No	
Good BIBI	218	0.027	(0.024,0.035)	V.Poor vs. Good	No	0.027
V.Poor FIBI	49	0.012	(0.004,0.027)			
Poor FIBI	69	0.037	(0.029,0.045)			
Fair FIBI	187	0.035	(0.026,0.037)	Poor vs. Fair	No	
Good FIBI	302	0.026	(0.024,0.027)	V.Poor vs. Good	No	0.036

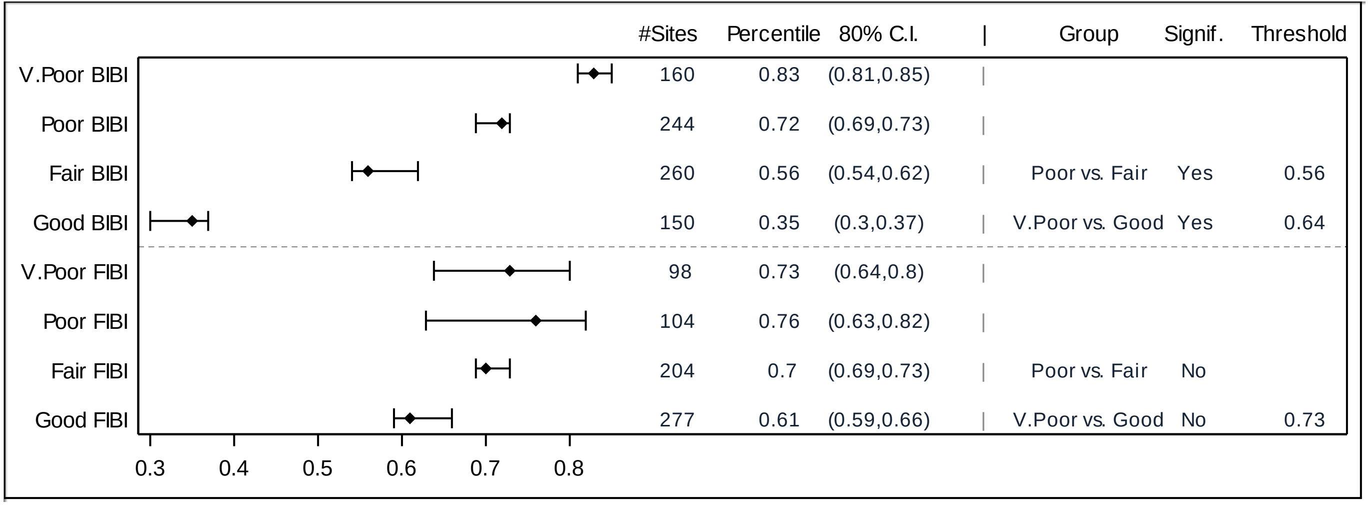
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.033	(0.026,0.042)			
Poor BIBI	213	0.034	(0.03,0.043)			
Fair BIBI	256	0.028	(0.022,0.035)	Poor vs. Fair	No	
Good BIBI	287	0.026	(0.022,0.029)	V.Poor vs. Good	No	0.031
V.Poor FIBI	70	0.028	(0.016,0.041)			
Poor FIBI	118	0.034	(0.029,0.04)			
Fair FIBI	239	0.031	(0.026,0.034)	Poor vs. Fair	No	
Good FIBI	294	0.023	(0.022,0.026)	V.Poor vs. Good	No	0.032

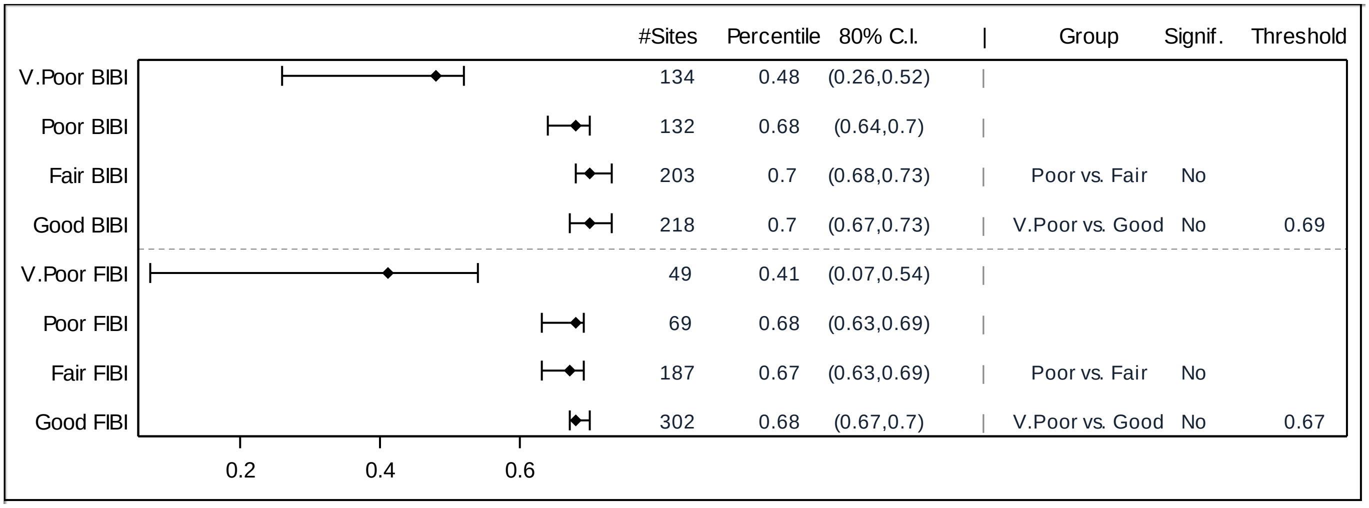
Table A-27. Physiographic Eco-region Analysis for High % of Agriculture in Watershed

Highland



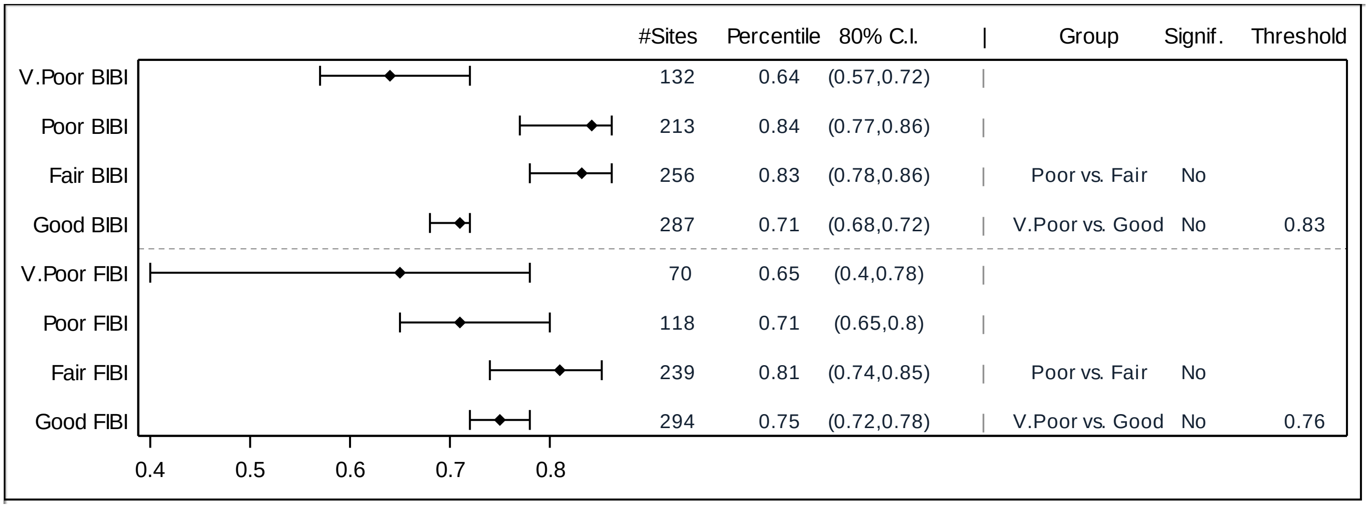
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.83	(0.81,0.85)			
Poor BIBI	244	0.72	(0.69,0.73)			
Fair BIBI	260	0.56	(0.54,0.62)	Poor vs. Fair	Yes	0.56
Good BIBI	150	0.35	(0.3,0.37)	V.Poor vs. Good	Yes	0.64
V.Poor FIBI	98	0.73	(0.64,0.8)			
Poor FIBI	104	0.76	(0.63,0.82)			
Fair FIBI	204	0.7	(0.69,0.73)	Poor vs. Fair	No	
Good FIBI	277	0.61	(0.59,0.66)	V.Poor vs. Good	No	0.73

Eastern Piedmont



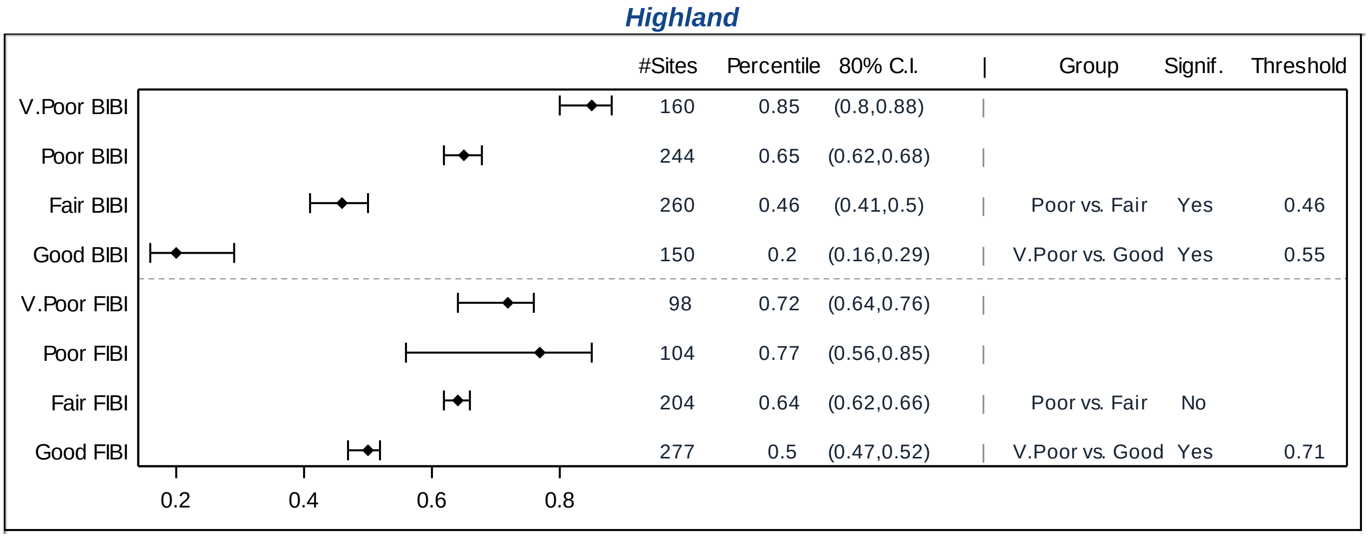
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.48	(0.26,0.52)			
Poor BIBI	132	0.68	(0.64,0.7)			
Fair BIBI	203	0.7	(0.68,0.73)	Poor vs. Fair	No	
Good BIBI	218	0.7	(0.67,0.73)	V.Poor vs. Good	No	0.69
V.Poor FIBI	49	0.41	(0.07,0.54)			
Poor FIBI	69	0.68	(0.63,0.69)			
Fair FIBI	187	0.67	(0.63,0.69)	Poor vs. Fair	No	
Good FIBI	302	0.68	(0.67,0.7)	V.Poor vs. Good	No	0.67

Coastal

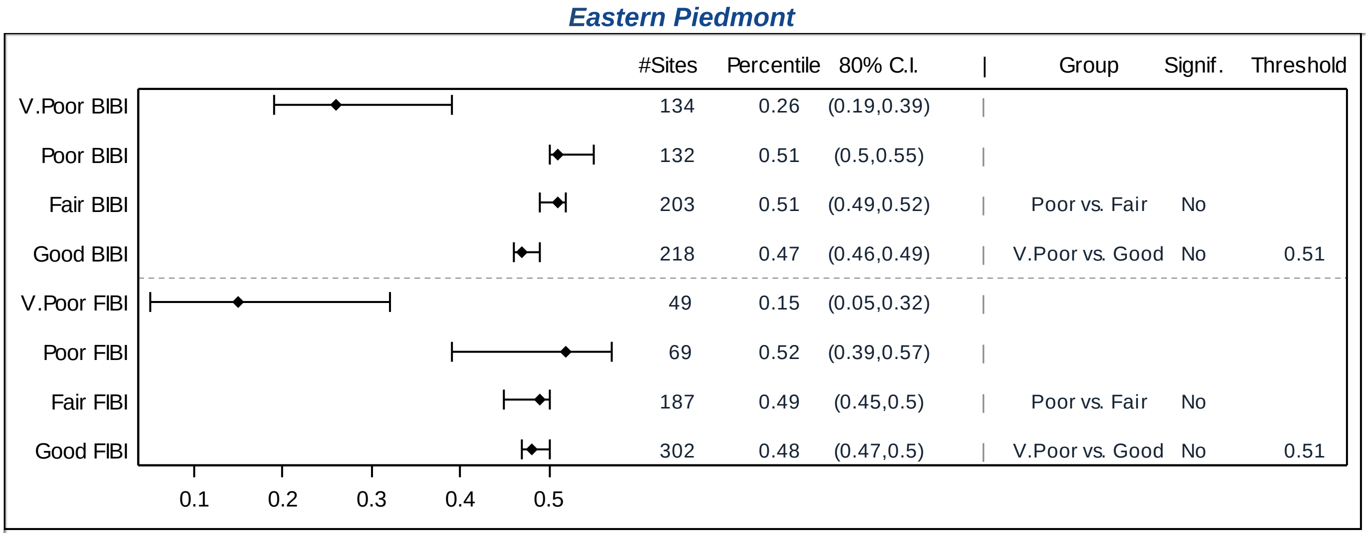


	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.64	(0.57,0.72)			
Poor BIBI	213	0.84	(0.77,0.86)			
Fair BIBI	256	0.83	(0.78,0.86)	Poor vs. Fair	No	
Good BIBI	287	0.71	(0.68,0.72)	V.Poor vs. Good	No	0.83
V.Poor FIBI	70	0.65	(0.4,0.78)			
Poor FIBI	118	0.71	(0.65,0.8)			
Fair FIBI	239	0.81	(0.74,0.85)	Poor vs. Fair	No	
Good FIBI	294	0.75	(0.72,0.78)	V.Poor vs. Good	No	0.76

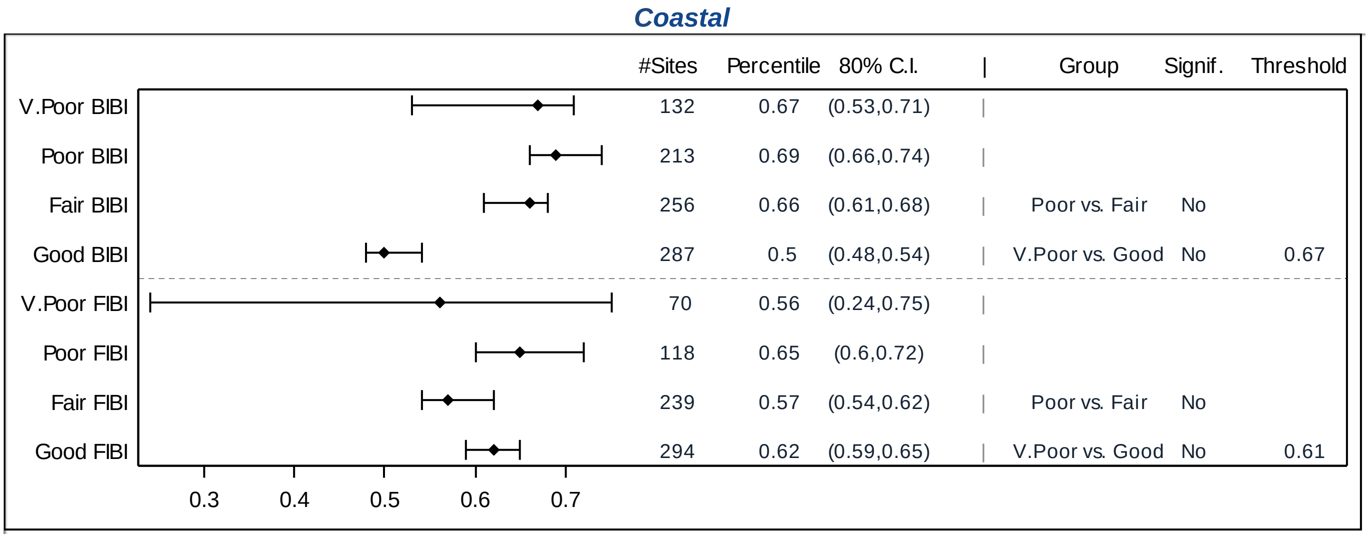
Table A-28. Physiographic Eco-region Analysis for High % of Agriculture in 60m Buffer


Highland

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.85	(0.8,0.88)			
Poor BIBI	244	0.65	(0.62,0.68)			
Fair BIBI	260	0.46	(0.41,0.5)	Poor vs. Fair	Yes	0.46
Good BIBI	150	0.2	(0.16,0.29)	V.Poor vs. Good	Yes	0.55
V.Poor FIBI	98	0.72	(0.64,0.76)			
Poor FIBI	104	0.77	(0.56,0.85)			
Fair FIBI	204	0.64	(0.62,0.66)	Poor vs. Fair	No	
Good FIBI	277	0.5	(0.47,0.52)	V.Poor vs. Good	Yes	0.71


Eastern Piedmont

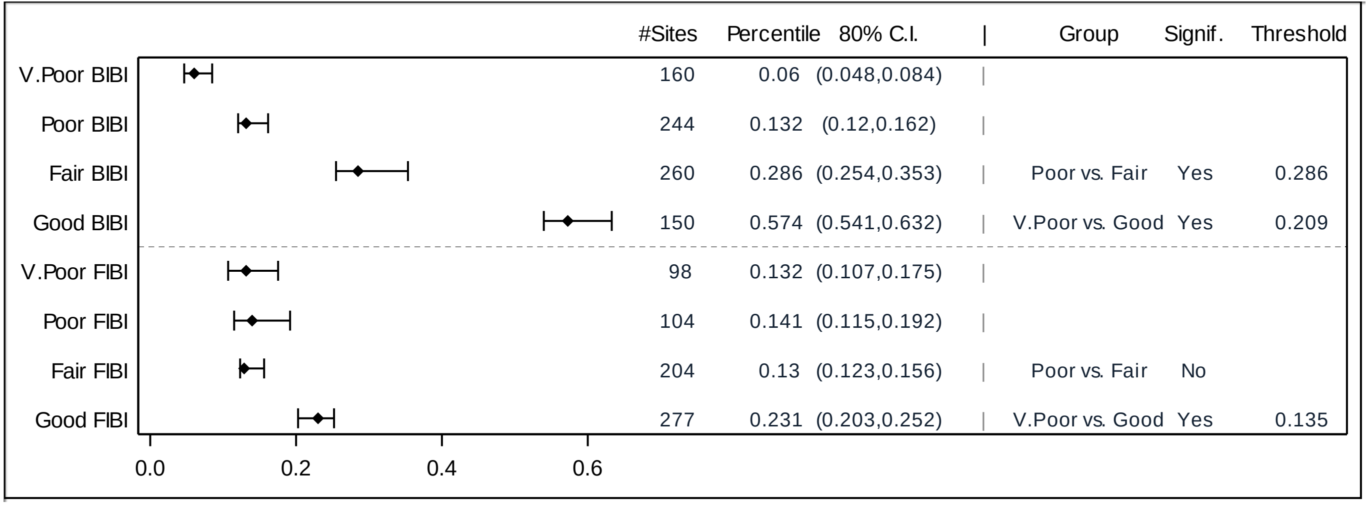
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.26	(0.19,0.39)			
Poor BIBI	132	0.51	(0.5,0.55)			
Fair BIBI	203	0.51	(0.49,0.52)	Poor vs. Fair	No	
Good BIBI	218	0.47	(0.46,0.49)	V.Poor vs. Good	No	0.51
V.Poor FIBI	49	0.15	(0.05,0.32)			
Poor FIBI	69	0.52	(0.39,0.57)			
Fair FIBI	187	0.49	(0.45,0.5)	Poor vs. Fair	No	
Good FIBI	302	0.48	(0.47,0.5)	V.Poor vs. Good	No	0.51


Coastal

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.67	(0.53,0.71)			
Poor BIBI	213	0.69	(0.66,0.74)			
Fair BIBI	256	0.66	(0.61,0.68)	Poor vs. Fair	No	
Good BIBI	287	0.5	(0.48,0.54)	V.Poor vs. Good	No	0.67
V.Poor FIBI	70	0.56	(0.24,0.75)			
Poor FIBI	118	0.65	(0.6,0.72)			
Fair FIBI	239	0.57	(0.54,0.62)	Poor vs. Fair	No	
Good FIBI	294	0.62	(0.59,0.65)	V.Poor vs. Good	No	0.61

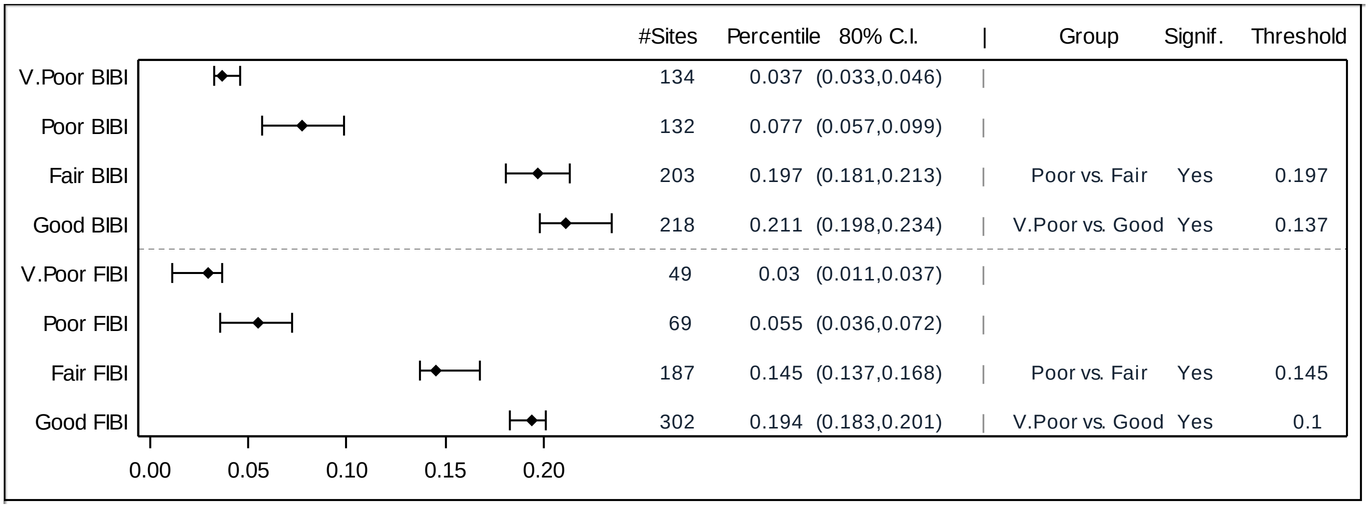
Table A-29. Physiographic Eco-region Analysis for Low % of Forest in Watershed

Highland



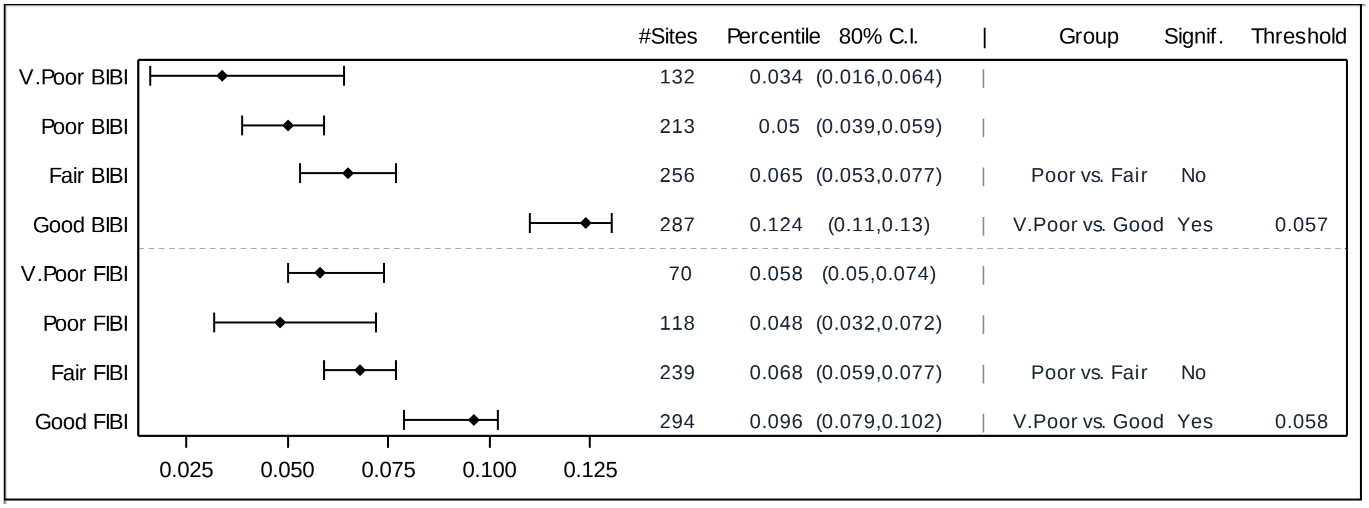
	#Sites	Percentile	80% C.I.	|	Group	Signif.	Threshold
V.Poor BIBI	160	0.06	(0.048,0.084)	|			
Poor BIBI	244	0.132	(0.12,0.162)	|			
Fair BIBI	260	0.286	(0.254,0.353)	|	Poor vs. Fair	Yes	0.286
Good BIBI	150	0.574	(0.541,0.632)	|	V.Poor vs. Good	Yes	0.209
V.Poor FIBI	98	0.132	(0.107,0.175)	|			
Poor FIBI	104	0.141	(0.115,0.192)	|			
Fair FIBI	204	0.13	(0.123,0.156)	|	Poor vs. Fair	No	
Good FIBI	277	0.231	(0.203,0.252)	|	V.Poor vs. Good	Yes	0.135

Eastern Piedmont



	#Sites	Percentile	80% C.I.	|	Group	Signif.	Threshold
V.Poor BIBI	134	0.037	(0.033,0.046)	|			
Poor BIBI	132	0.077	(0.057,0.099)	|			
Fair BIBI	203	0.197	(0.181,0.213)	|	Poor vs. Fair	Yes	0.197
Good BIBI	218	0.211	(0.198,0.234)	|	V.Poor vs. Good	Yes	0.137
V.Poor FIBI	49	0.03	(0.011,0.037)	|			
Poor FIBI	69	0.055	(0.036,0.072)	|			
Fair FIBI	187	0.145	(0.137,0.168)	|	Poor vs. Fair	Yes	0.145
Good FIBI	302	0.194	(0.183,0.201)	|	V.Poor vs. Good	Yes	0.1

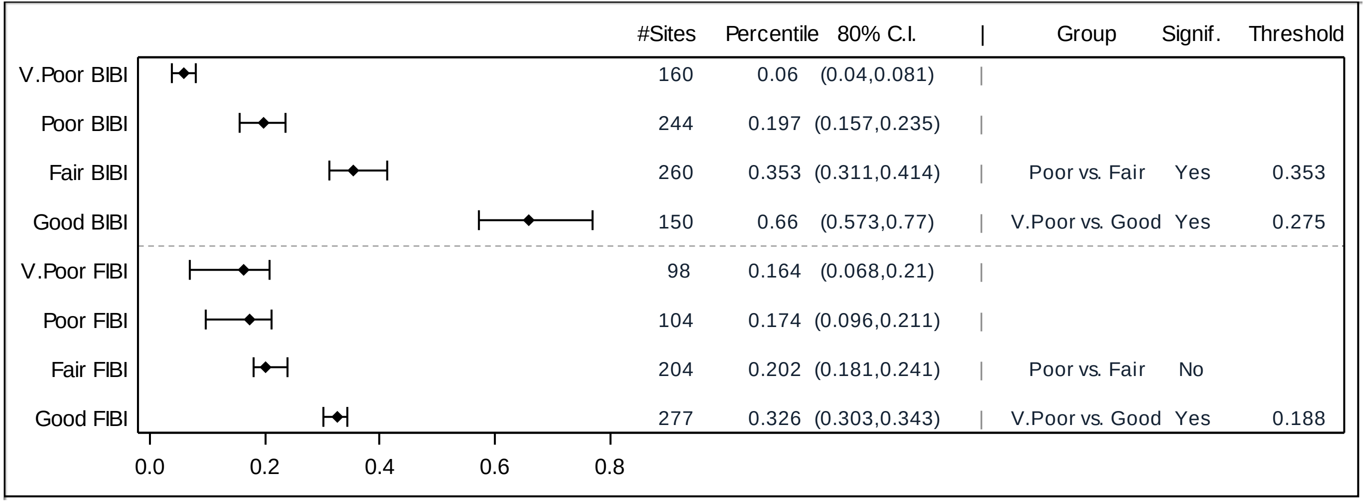
Coastal



	#Sites	Percentile	80% C.I.	|	Group	Signif.	Threshold
V.Poor BIBI	132	0.034	(0.016,0.064)	|			
Poor BIBI	213	0.05	(0.039,0.059)	|			
Fair BIBI	256	0.065	(0.053,0.077)	|	Poor vs. Fair	No	
Good BIBI	287	0.124	(0.11,0.13)	|	V.Poor vs. Good	Yes	0.057
V.Poor FIBI	70	0.058	(0.05,0.074)	|			
Poor FIBI	118	0.048	(0.032,0.072)	|			
Fair FIBI	239	0.068	(0.059,0.077)	|	Poor vs. Fair	No	
Good FIBI	294	0.096	(0.079,0.102)	|	V.Poor vs. Good	Yes	0.058

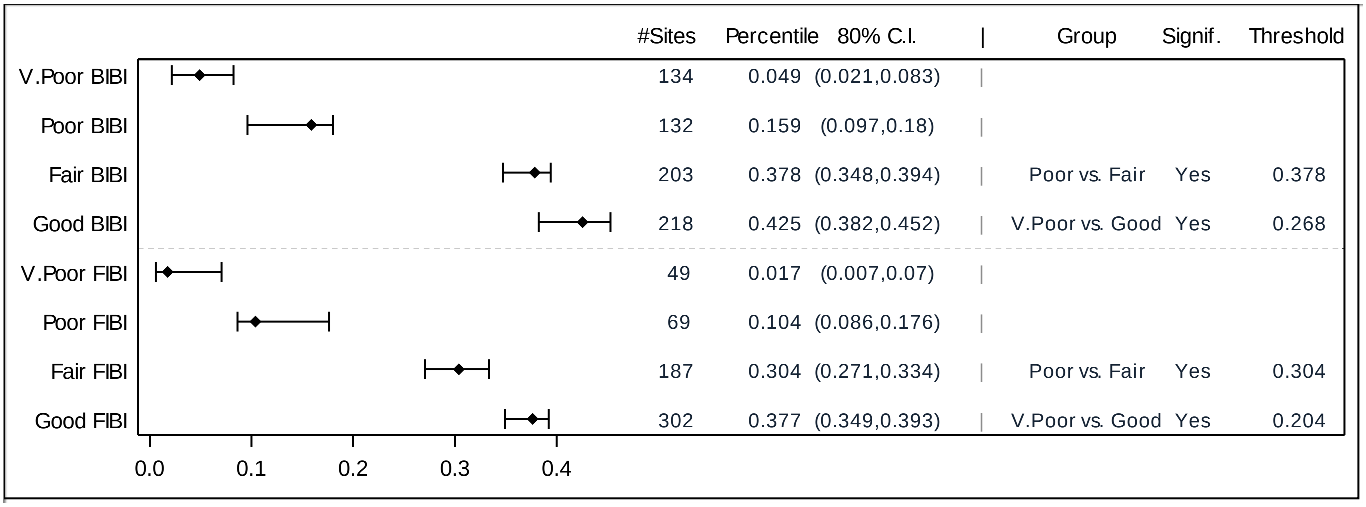
Table A-30. Physiographic Eco-region Analysis for Low % of Forest in 60m Buffer

Highland



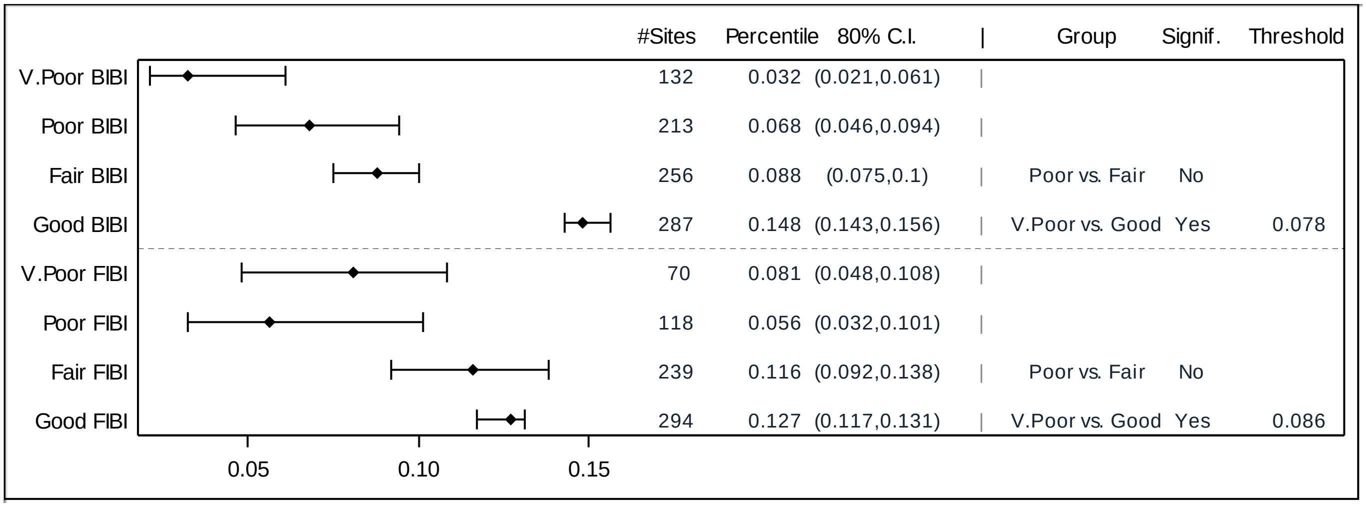
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0.06	(0.04,0.081)			
Poor BIBI	244	0.197	(0.157,0.235)			
Fair BIBI	260	0.353	(0.311,0.414)	Poor vs. Fair	Yes	0.353
Good BIBI	150	0.66	(0.573,0.77)	V.Poor vs. Good	Yes	0.275
V.Poor FIBI	98	0.164	(0.068,0.21)			
Poor FIBI	104	0.174	(0.096,0.211)			
Fair FIBI	204	0.202	(0.181,0.241)	Poor vs. Fair	No	
Good FIBI	277	0.326	(0.303,0.343)	V.Poor vs. Good	Yes	0.188

Eastern Piedmont



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0.049	(0.021,0.083)			
Poor BIBI	132	0.159	(0.097,0.18)			
Fair BIBI	203	0.378	(0.348,0.394)	Poor vs. Fair	Yes	0.378
Good BIBI	218	0.425	(0.382,0.452)	V.Poor vs. Good	Yes	0.268
V.Poor FIBI	49	0.017	(0.007,0.07)			
Poor FIBI	69	0.104	(0.086,0.176)			
Fair FIBI	187	0.304	(0.271,0.334)	Poor vs. Fair	Yes	0.304
Good FIBI	302	0.377	(0.349,0.393)	V.Poor vs. Good	Yes	0.204

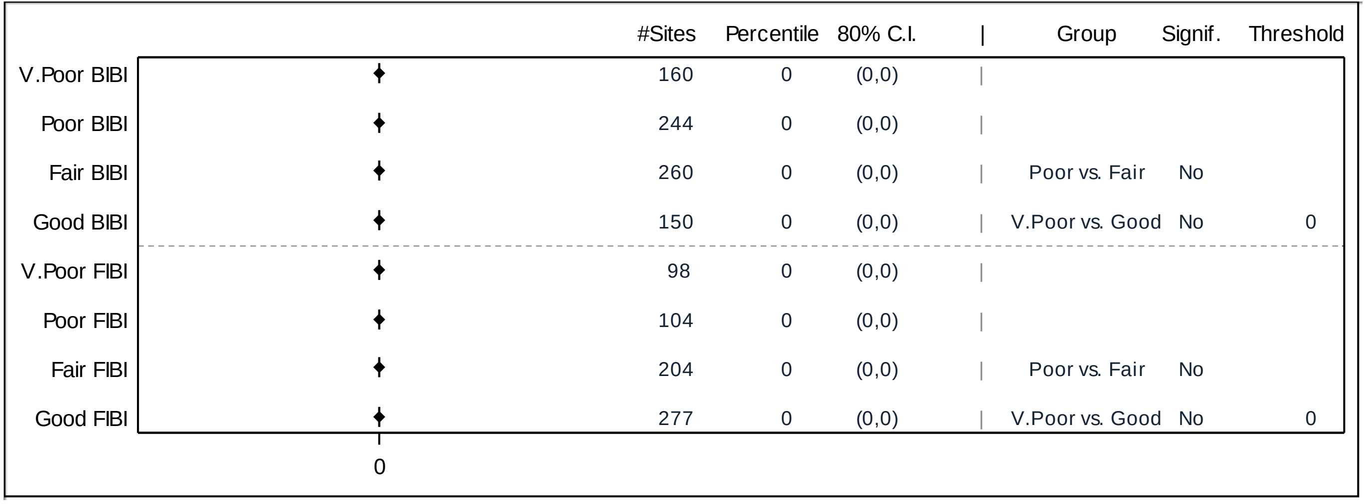
Coastal



	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.032	(0.021,0.061)			
Poor BIBI	213	0.068	(0.046,0.094)			
Fair BIBI	256	0.088	(0.075,0.1)	Poor vs. Fair	No	
Good BIBI	287	0.148	(0.143,0.156)	V.Poor vs. Good	Yes	0.078
V.Poor FIBI	70	0.081	(0.048,0.108)			
Poor FIBI	118	0.056	(0.032,0.101)			
Fair FIBI	239	0.116	(0.092,0.138)	Poor vs. Fair	No	
Good FIBI	294	0.127	(0.117,0.131)	V.Poor vs. Good	Yes	0.086

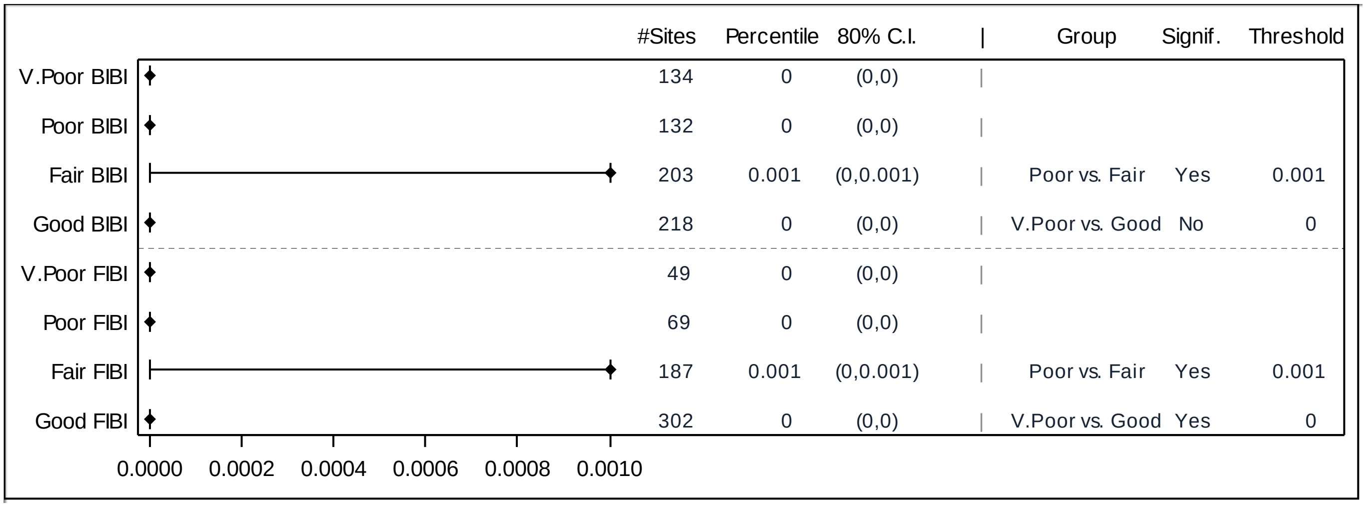
Table A-31. Physiographic Eco-region Analysis for Low % of Wetland in Watershed

Highland



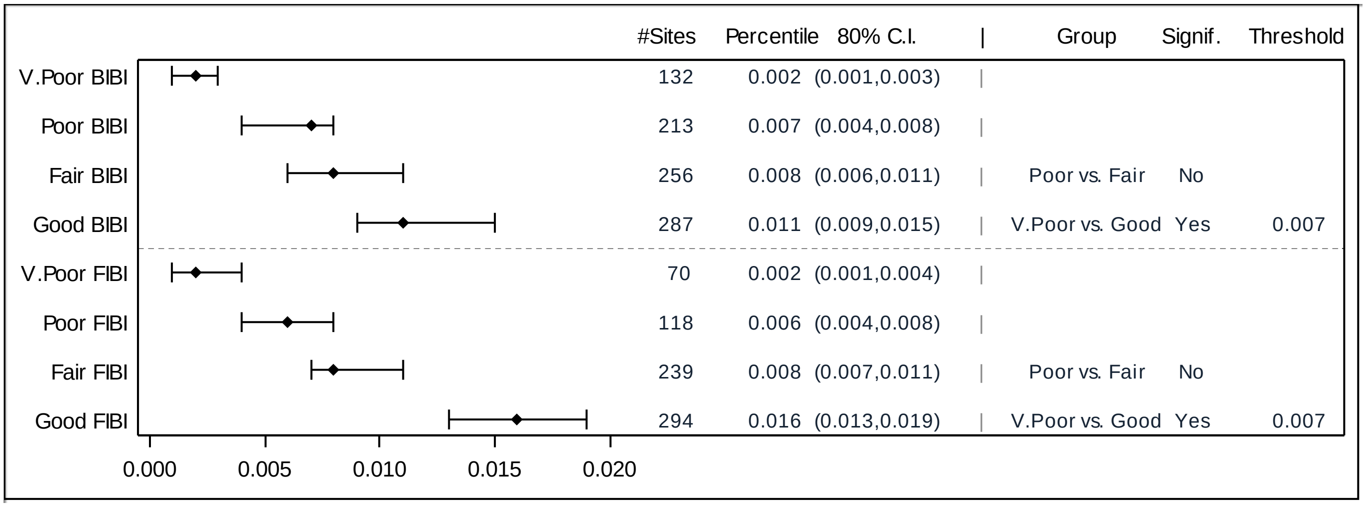
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0	(0,0)			
Poor BIBI	244	0	(0,0)			
Fair BIBI	260	0	(0,0)	Poor vs. Fair	No	
Good BIBI	150	0	(0,0)	V.Poor vs. Good	No	0
V.Poor FIBI	98	0	(0,0)			
Poor FIBI	104	0	(0,0)			
Fair FIBI	204	0	(0,0)	Poor vs. Fair	No	
Good FIBI	277	0	(0,0)	V.Poor vs. Good	No	0

Eastern Piedmont



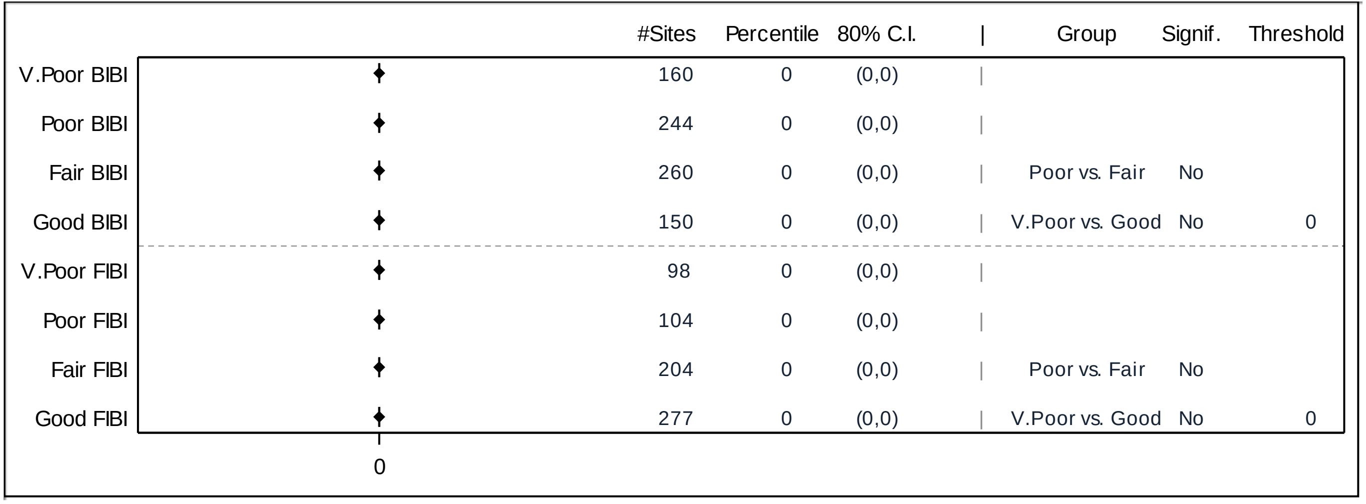
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0	(0,0)			
Poor BIBI	132	0	(0,0)			
Fair BIBI	203	0.001	(0,0.001)	Poor vs. Fair	Yes	0.001
Good BIBI	218	0	(0,0)	V.Poor vs. Good	No	0
V.Poor FIBI	49	0	(0,0)			
Poor FIBI	69	0	(0,0)			
Fair FIBI	187	0.001	(0,0.001)	Poor vs. Fair	Yes	0.001
Good FIBI	302	0	(0,0)	V.Poor vs. Good	Yes	0

Coastal



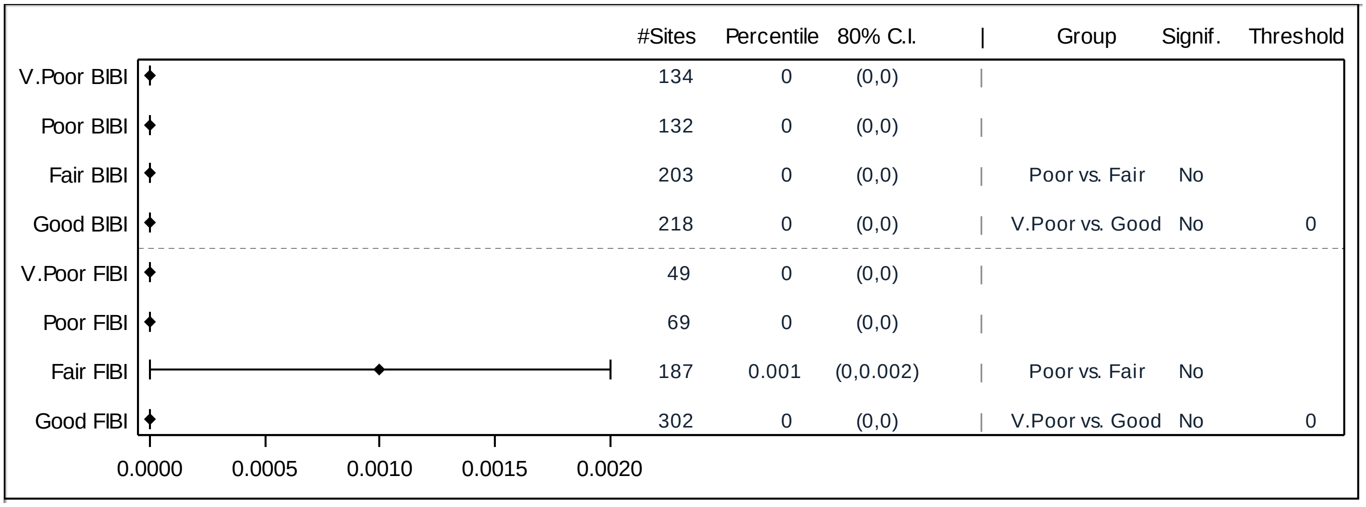
	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0.002	(0.001,0.003)			
Poor BIBI	213	0.007	(0.004,0.008)			
Fair BIBI	256	0.008	(0.006,0.011)	Poor vs. Fair	No	
Good BIBI	287	0.011	(0.009,0.015)	V.Poor vs. Good	Yes	0.007
V.Poor FIBI	70	0.002	(0.001,0.004)			
Poor FIBI	118	0.006	(0.004,0.008)			
Fair FIBI	239	0.008	(0.007,0.011)	Poor vs. Fair	No	
Good FIBI	294	0.016	(0.013,0.019)	V.Poor vs. Good	Yes	0.007

Table A-32. Physiographic Eco-region Analysis for Low % of Wetland in 60m Buffer



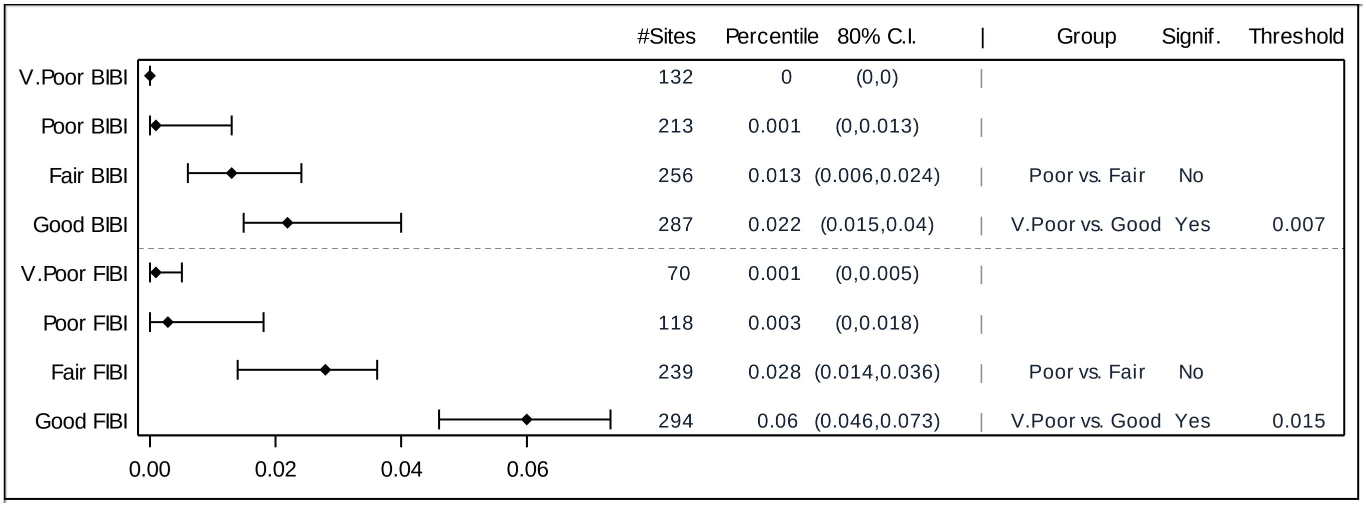
Highland

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	160	0	(0,0)			
Poor BIBI	244	0	(0,0)			
Fair BIBI	260	0	(0,0)	Poor vs. Fair	No	
Good BIBI	150	0	(0,0)	V.Poor vs. Good	No	0
V.Poor FIBI	98	0	(0,0)			
Poor FIBI	104	0	(0,0)			
Fair FIBI	204	0	(0,0)	Poor vs. Fair	No	
Good FIBI	277	0	(0,0)	V.Poor vs. Good	No	0



Eastern Piedmont

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	134	0	(0,0)			
Poor BIBI	132	0	(0,0)			
Fair BIBI	203	0	(0,0)	Poor vs. Fair	No	
Good BIBI	218	0	(0,0)	V.Poor vs. Good	No	0
V.Poor FIBI	49	0	(0,0)			
Poor FIBI	69	0	(0,0)			
Fair FIBI	187	0.001	(0,0.002)	Poor vs. Fair	No	
Good FIBI	302	0	(0,0)	V.Poor vs. Good	No	0



Coastal

	#Sites	Percentile	80% C.I.	Group	Signif.	Threshold
V.Poor BIBI	132	0	(0,0)			
Poor BIBI	213	0.001	(0,0.013)			
Fair BIBI	256	0.013	(0.006,0.024)	Poor vs. Fair	No	
Good BIBI	287	0.022	(0.015,0.04)	V.Poor vs. Good	Yes	0.007
V.Poor FIBI	70	0.001	(0,0.005)			
Poor FIBI	118	0.003	(0,0.018)			
Fair FIBI	239	0.028	(0.014,0.036)	Poor vs. Fair	No	
Good FIBI	294	0.06	(0.046,0.073)	V.Poor vs. Good	Yes	0.015

Appendix B: General Causal Scenario Models

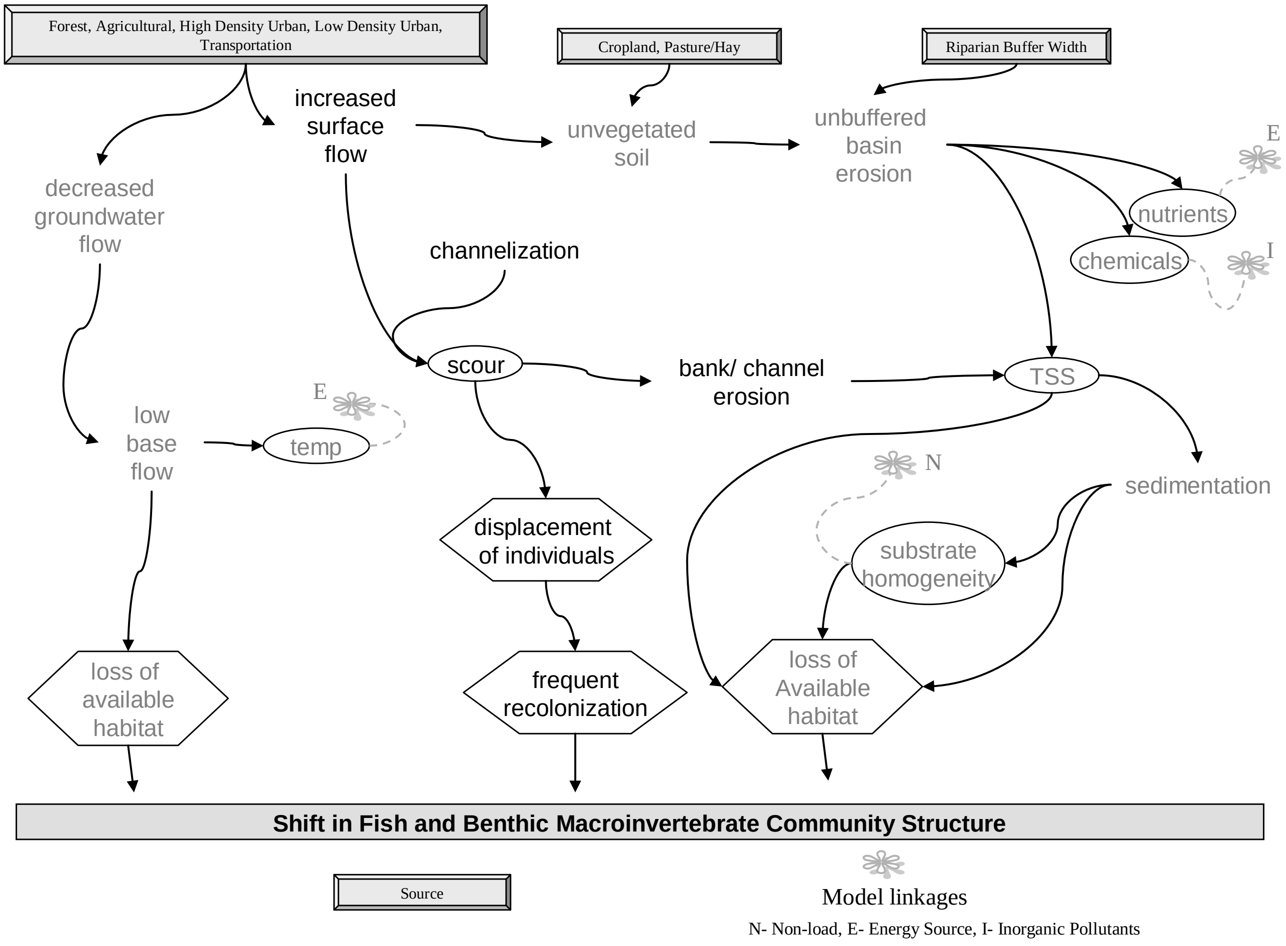


Figure B-1 Flow/Sediment Causal Scenario

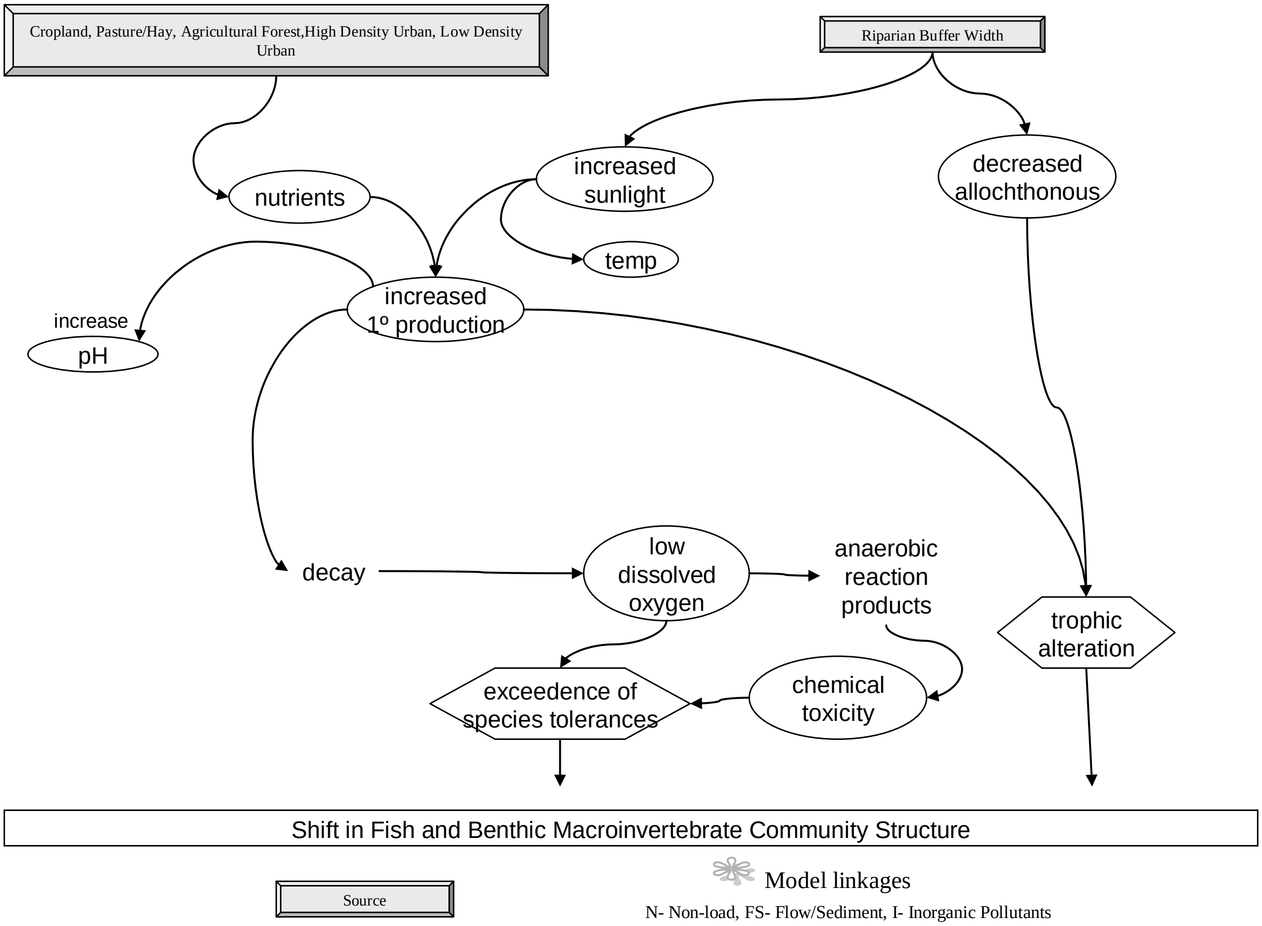


Figure B-2 Energy Source Causal Scenario

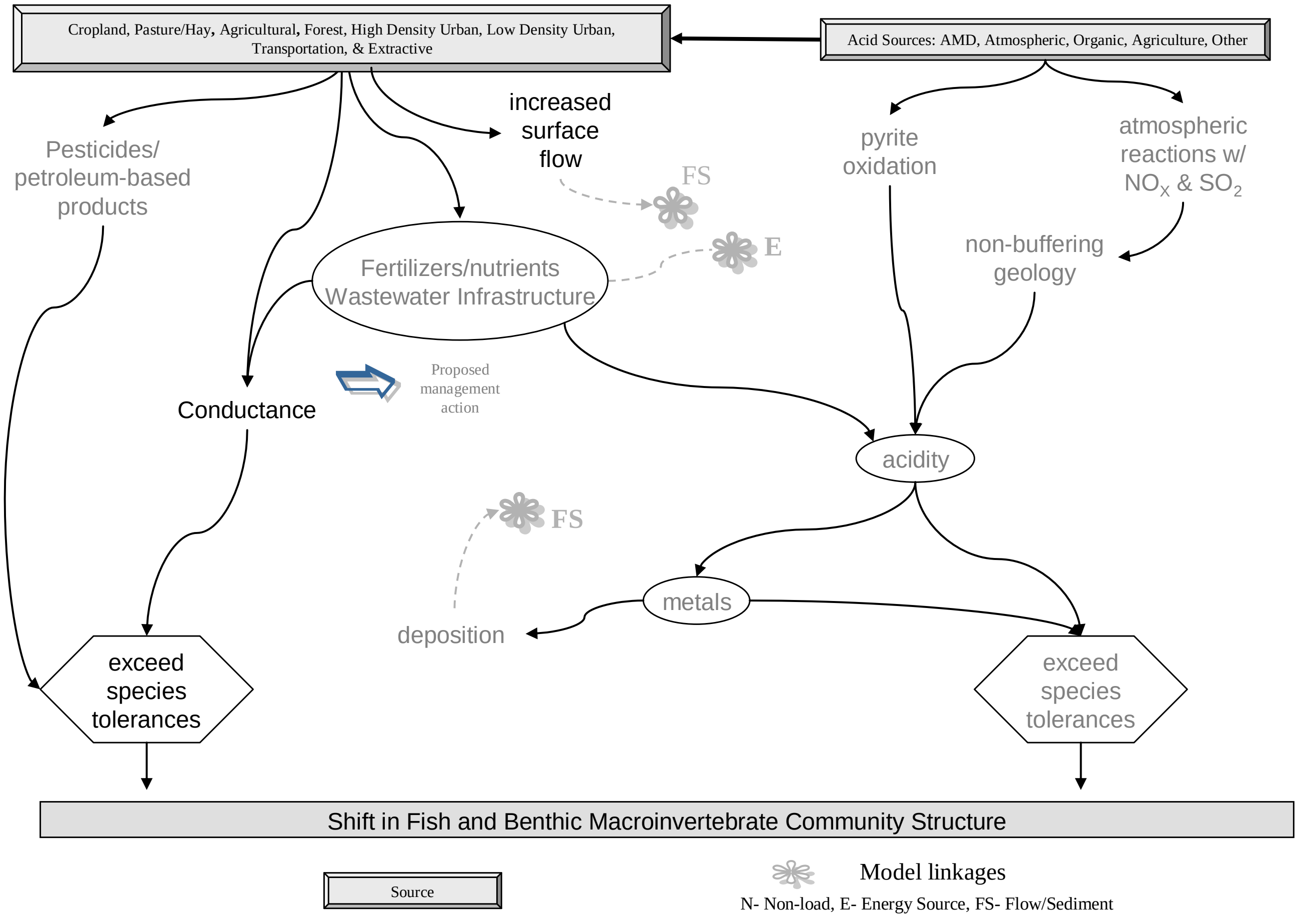


Figure B-3 Inorganic Pollutant Causal Scenario

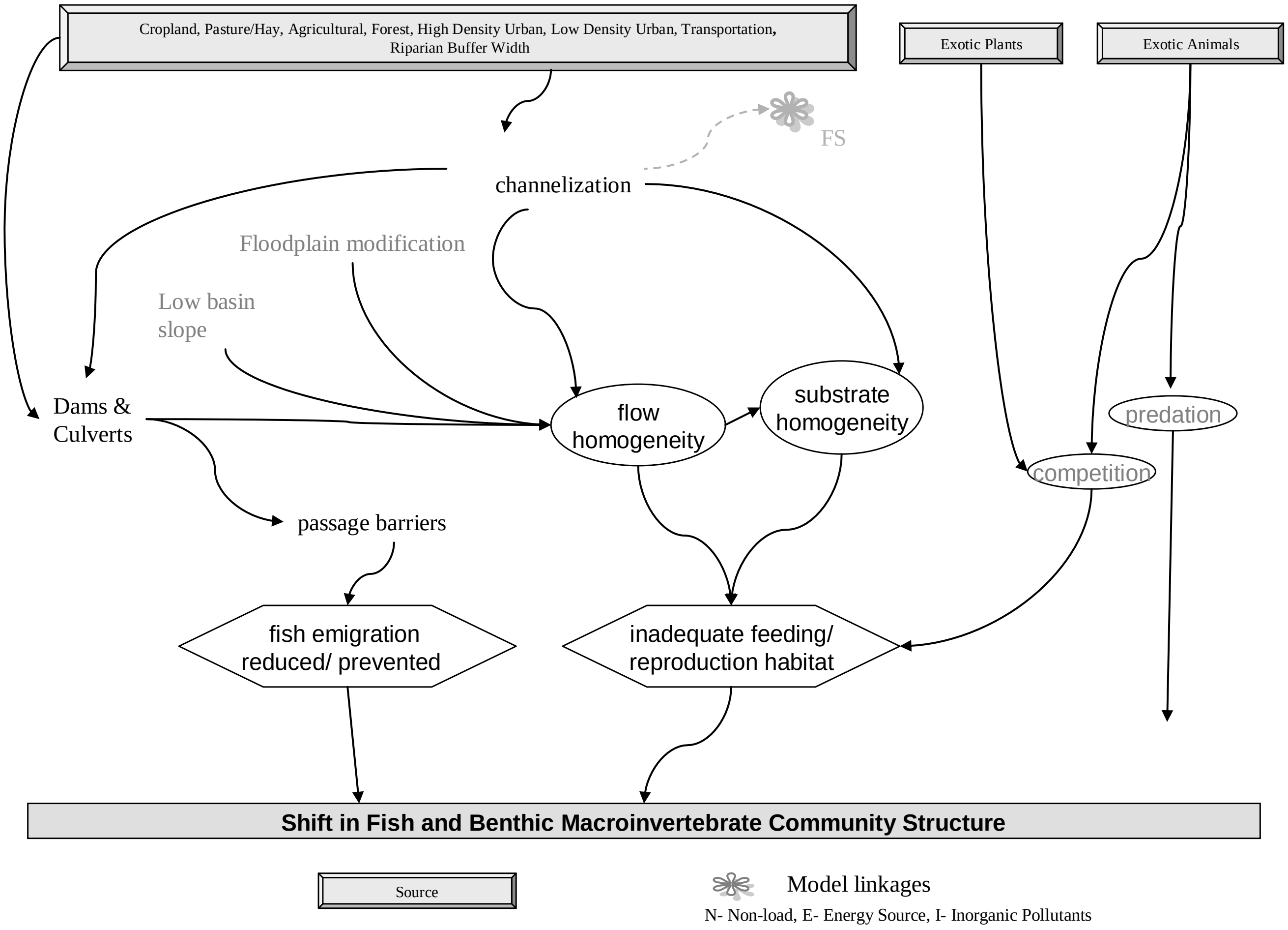


Figure B-4 Non-Load Causal Scenario